No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

RENEWED PETITION FOR A WRIT OF MANDAMUS

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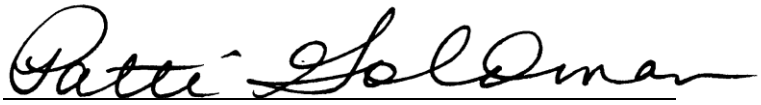
CORPORATE DISCLOSURE STATEMENT REQUIRED BY FRAP 26.1

Petitioners Pesticide Action Network North America and Natural Resources Defense Council, Inc., have no parent, subsidiary, or affiliate that has issued shares or debt securities to the public.

Respectfully submitted this 10th day of September, 2014.



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INTRODUCTION

Pesticide Action Network North America and Natural Resources Defense Council (collectively "PANNA") file this renewed petition for a Writ of Mandamus seeking an order from this Court requiring that the U.S. Environmental Protection Agency ("EPA") finally and fully respond to a 2007 petition to ban uses of chlorpyrifos, a pesticide that causes large numbers of poisonings of workers, children, and rural families every year and that peer-reviewed studies have linked to neurological and behavioral impairments in children. Petition to Revoke All Tolerances and Cancel All Registrations for the Pesticide Chlorpyrifos (September 12, 2007) (the "2007 Petition") (Exhibit B to First Sass Decl. (April 12, 2012)). The petition sought a ban on uses of chlorpyrifos that expose children to acute pesticide poisonings from pesticide spraying and to documented risks of neurological and other impairments from all exposures to chlorpyrifos whether from pesticide spraying, food residues, or other routes of exposure. EPA has initiated several processes to assess the health risks posed by chlorpyrifos as presented in the 2007 Petition and has released some partial responses that address discrete contentions. Jack Housenger Decl. in *In re Pesticide Action Network North America and Natural Resources Defense Council*, No. 12-71125, ECF No. 9-2 (9th Cir. July 24, 2012) ("*In re PANNA*") (Exhibit 1 to this Mandamus Petition). However, it has yet to issue a final and reviewable decision on the

request to ban chlorpyrifos, leaving PANNA in legal limbo and this dangerous pesticide in widespread use.

Over the past seven years, EPA has made commitments to PANNA and the courts to resolve the 2007 Petition and decide whether to ban chlorpyrifos by various deadlines. Without fail, EPA has violated these commitments. When EPA failed to respond, PANNA filed its first lawsuit, leading to a stipulated deadline of November 23, 2011, which EPA missed. The second lawsuit in the form of a petition for a writ of mandamus before this Court extracted two promises from EPA: first, that it would respond by December 2012, and when that deadline passed, that it would fully resolve the petition by February 2014. *In re PANNA*, 532 F. App'x 649, 651 (9th Cir. 2013). In large part based on that commitment, which this Court characterized as "concrete," this Court declined to issue a writ of mandamus, but it explicitly stated that its denial was "without prejudice to seeking the same relief at a future date in the event EPA fails to act." *Id.* at 651-52. EPA's promised February 2014 deadline has come and gone without a final response to the 2007 Petition. Accordingly, PANNA renews its petition for a writ of mandamus. PANNA asks this Court to find that EPA has unreasonably delayed fulfilling its legal obligations and to compel EPA to issue a final decision on the 2007 Petition by EPA's newly promised timeline of December 2014 and summer of 2015, depending on its determination.

STATEMENT OF JURISDICTION AND APPLICABLE LAW

This Court has authority to issue a writ of mandamus pursuant to the All Writs Act, 28 U.S.C. § 1651 (authorizing federal courts to issue all writs appropriate "in aid of their respective jurisdictions") and the Administrative Procedure Act ("APA"), 5 U.S.C. § 706(1) (reviewing court shall "compel agency action unlawfully withheld or unreasonably delayed"). *See In re PANNA*, 532 F. App'x at 650 (citing 5 U.S.C. § 706(1)). As this Court recognized in ruling on the first petition for writ of mandamus, this Court has jurisdiction to review this challenge to the agency's delay because challenges to any final action by EPA would lie in this Court. *See In re PANNA*, 532 F. App'x at 650; *Telecomm. Research & Action Ctr. v. FCC*, 750 F.2d 70, 75 (D.C. Cir. 1984) (hereinafter "*TRAC*").

THE ISSUE PRESENTED

Whether EPA's seven-year delay in deciding whether to ban a hazardous and widely used pesticide that is particularly harmful to children, as requested in the 2007 Petition, is an unreasonable delay warranting an order from this Court requiring EPA to issue a final decision on the schedule EPA has most recently proposed.

STATEMENT OF THE CASE

Chlorpyrifos is a widely used pesticide that has repeatedly been among the top pesticides causing acute pesticide poisonings of workers, their families, and

others who live near places where it is applied. The unacceptable harms to children exposed to chlorpyrifos on lawns and in their homes led EPA to negotiate a phase out-of-home uses in 2000. Inexplicably, EPA neglected to protect rural children from similar harms, despite acknowledging, in the face of litigation and petitions by PANNA and others, its legal obligation to protect rural children from pesticide drift and volatilization. Rural children exposed to chlorpyrifos are often the children of farmworkers, such that this harm falls disproportionately on children in low-income and minority communities. Compounding these harms, a series of peer-reviewed scientific studies has found links between chlorpyrifos and neuro-developmental and behavioral impairments in children at lower levels of exposure than those that cause acute pesticide poisonings. The 2007 Petition presented these risks to EPA. EPA has repeatedly promised to issue a final decision on the Petition, but has repeatedly broken those promises. This statement of the case reviews the pertinent statutory structure, EPA's failure to address serious health impacts to children and bystanders from chlorpyrifos use, and its handling of the 2007 Petition.

I. EPA HAD UNTIL 2006 TO BRING CHLORPYRIFOS INTO COMPLIANCE WITH TWO OVERLAPPING STATUTES REGULATING PESTICIDE USE.

EPA regulates pesticides under two, overlapping statutes, the Federal Food, Drug and Cosmetic Act ("FFDCA") and Federal Insecticide, Rodenticide and

Fungicide Act ("FIFRA"). EPA issues tolerances under the FFDCA, which establish the maximum residue of a pesticide allowed on food. 21 U.S.C. § 346a(b) & (c). EPA may "establish or leave in effect a tolerance for a pesticide chemical residue in or on a food only if the Administrator determines that the tolerance is safe." *Id.* § 346a(b)(2)(A)(i). EPA has the authority to revoke a tolerance if it finds a pesticide residue would not be safe. *Id.* § 346a(b)(2)(A)(i).

Under FIFRA, EPA must establish a registration before a pesticide may generally be sold or used in the United States. 7 U.S.C. § 136a(a). To register or re-register a pesticide, EPA must determine that its use "will not generally cause unreasonable adverse effects on the environment," which includes risks to human health. *Id.* § 136a(c)(5)(D); *see id.* § 136(bb) (definition of "unreasonable adverse effects"). EPA has the authority and the duty to cancel a pesticide registration if the pesticide use "causes unreasonable adverse effects on the environment," including human health. *Id.* § 136d(b).

Congress overhauled our food safety laws in 1996. The overhaul responded to the seminal 1993 National Academy of Sciences ("NAS") report criticizing EPA for treating children like "little adults" by failing to address the unique susceptibility of children to pesticide exposures based on the foods they eat, their play, and sensitive stages of their development. The NAS recommended that EPA revamp and strengthen its pesticide regulations to account for children's

vulnerabilities, consumption patterns, and exposures.[1] In particular, because "[e]xposure to pesticide residues from ambient air sources is generally higher in areas close to agricultural lands," the NAS recommended that "exposure from all sources—not just ingestion—must be considered when estimating total [pesticide] exposure and risk to children."[2]

The Food Quality Protection Act ("FQPA"), passed unanimously in 1996, amends the FFDCA and FIFRA and requires EPA to "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure" to pesticides. 21 U.S.C. § 346a(b)(2)(C)(ii)(I), (II). "Aggregate exposure" includes "all anticipated dietary exposures and all other exposures for which there is reliable information," including pesticide drift exposures. 21 U.S.C. § 346a(b)(2)(A)(ii); *see also id.* § 346a(b)(2)(C)(vi). The FQPA also requires EPA to assess and protect against unsafe risks posed by cumulative exposures to pesticides that share a "common mechanism of toxicity." *See* 21 U.S.C. § 346a(b)(2)(C)-(D). In addition, the FQPA directs EPA to afford added protection to children based on their exposure patterns, their special sensitivities such as during early or adolescent development, and gaps in available data to assess such risks. 21 U.S.C. § 346a(b)(2)(C)-(D).

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[1] NAS, *Pesticides in the Diets of Infants and Children*, Executive Summary at 307-09 (1993) (Exhibit 2) ("NAS Report").
[2] *Id*. at 307, 308-09.

The FQPA also amended FIFRA's "unreasonable adverse effects" definition to include "a human dietary risk from residues that result from a use of a pesticide in or on any food inconsistent with the [FQPA] standard." 7 U.S.C. § 136(bb)(2). Accordingly, EPA can register or re-register a pesticide only if there is a reasonable certainty of no harm from aggregate and cumulative exposures to the pesticide under the FQPA standard.

Congress gave EPA a ten-year deadline, which ended in August 2006, to bring all food-use pesticides into compliance with these protective mandates. 21 U.S.C. § 346a(q)(1). The August 2006 deadline applied to both tolerances established under the FFDCA as amended by the FQPA and re-registration decisions under FIFRA.

II. EPA'S 2001 AND 2006 CHLORPYRIFOS DETERMINATIONS FAILED TO ADDRESS SERIOUS HEALTH IMPACTS TO CHILDREN AND BYSTANDER EXPOSURES.

A. Chlorpyrifos Poses Serious Health Risks to Children.

Chlorpyrifos is a widely used pesticide first registered by EPA in the 1960s. It is an organophosphate pesticide, a class of pesticides developed as nerve agents in World War II. First Sass Decl. ¶ 4. After the war, chlorpyrifos and other organophosphates were adapted for use as insecticides. First Sass Decl. ¶¶ 3-4; 2007 Petition at 1. In setting priorities for reviewing old pesticides under the FQPA, EPA gave priority to organophosphates because they are among the

pesticides that "pose the greatest risk to public health." 62 Fed. Reg. 42,020, 42,021 (Aug. 4, 1997).

Chlorpyrifos poses two types of serious public health risks. First, it is acutely toxic and causes systemic illnesses by inhibiting the body's ability to produce cholinesterase, an enzyme necessary for the proper transmission of nerve impulses. 2007 Petition at 1. Symptoms of cholinesterase inhibition caused by chlorpyrifos poisoning include muscle spasms, confusion, dizziness, loss of consciousness, seizures, abdominal cramps, vomiting, diarrhea, cessation of breathing, paralysis, and death. First Sass Decl. ¶¶ 4-5; 2007 Petition at 1. Year after year, chlorpyrifos has been identified as one of the pesticides associated with an alarming number of pesticide poisonings in many states. Second Reeves Decl. ¶ 7 (Aug. 27, 2014). For example, a PANNA report found that in California, chlorpyrifos was in the top five chemicals for poisoning incidents. First Reeves Decl. ¶ 9. This trend is particularly significant given widespread under-reporting of pesticide poisonings due to such factors as inadequate reporting systems, fear of retaliation from employers, and reluctance to seek medical treatment. Second Reeves Decl. ¶ 7(a). Another recent report showed that chlorpyrifos is the eighth most commonly used hazardous pesticide within ¼ mile of schools. *Id.* at ¶ 7(d).

Second, a growing body of published scientific research links exposure to chlorpyrifos with long-term harmful human health effects, including neuro-

developmental disorders, hyperactivity, attention deficit disorder, low birth weights, and reduced newborn head circumference, which is indicative of impaired cognitive ability. First Sass Decl. ¶¶ 7, 19-20; 2007 Petition at 6-10.

B. EPA's 2001 and 2006 Decisions for Chlorpyrifos and All Organophosphates Failed to Comply with Statutory Obligations.

To comply with the FQPA, EPA conducted an aggregate exposure assessment for chlorpyrifos to add together all of the ways people, and particularly children, are exposed to the pesticide. The FQPA requires an assessment based on aggregation of all exposures to chlorpyrifos, whether from eating foods, drinking water with residues of the pesticide, or uses of the pesticide in and around the home or places like golf courses where people can be exposed. 21 U.S.C. § 346a(b)(2)(A)(ii), (C)(i)(I). EPA developed a "risk cup" approach that compares all of the exposures for specific population groups, including fetuses, infants, and children in different age ranges to what it finds to be unsafe exposure levels. If aggregate exposures to the pesticide "overflow" the risk cup for a particular subpopulation, the pesticide does not meet the FQPA safety standard. EPA must then reduce exposures to levels that no longer exceed what it has deemed to be safe levels by, for example, banning uses.

For chlorpyrifos, EPA found alarmingly high exposures to children from uses of chlorpyrifos in the home, on pets, and in lawns and gardens.[3] EPA, Occupational/Residential Handler and Post Application Residential Risk Assessment for Chlorpyrifos, at 5-7 (Oct. 1999) (Exhibit 3). In 2000, EPA reached an agreement with the registrants to cancel home and garden uses of chlorpyrifos after determining that residential uses of these pesticides cause the child risk cup to overflow. *See* EPA, Administrator Carol M. Browner, Dursban Announcement, Remarks Prepared for Delivery June 8, 2000 (Exhibit 4). Then-Administrator Carol Browner heralded this agreement as "particularly good news for children, who are among the most vulnerable to the risks posed by pesticides." *Id*. at 1.

Inexplicably, EPA failed to assess children's exposures from chlorpyrifos spray drift and volatilization from agricultural sites to homes, schools, daycares, and playfields. By failing to assess the risks to children who are exposed to agricultural pesticide drift and volatilization, EPA maintained a double-standard: protecting kids from pesticides used in urban and residential settings, while leaving kids who live near agricultural sites—often in low-income and minority communities—unprotected and vulnerable to pesticide. This failure to protect farmworker and rural children falls short of the FQPA's requirements and the direction in federal executive orders to address disproportionate health risks to

[3] EPA, Occupational/Residential Handler and Post Application Residential Risk Assessment for Chlorpyrifos, at 5-7 (Oct. 1999) (Exhibit 3).

people of color and low-income populations. Exec. Order No. 12,898, §§ 1-101(b), 2-202(b), 59 Fed. Reg. 7,629 (Feb. 11, 1994) (requiring each federal agency to "ensure that its policies, programs, activities, and standards address disproportionate risks to children that result from environmental health or safety risks . . . that are attributable to products or substances that the child is likely to come in contact with or ingest (such as the air we breath [sic], the food we eat, the water we drink or use for recreation, the soil we live on, and the products we use or are exposed to).").

In 2001, after negotiating the phase-out of residential uses, EPA issued an interim re-registration determination ("IRED") for chlorpyrifos, which allowed chlorpyrifos uses and exposures to continue, although some at reduced levels. EPA, Interim Reregistration Eligibility Decision for Chlorpyrifos at 64-68 (Sept. 2001) (Attach. A to Housenger Decl.). PANNA, NRDC, and others commented on the 2001 IRED, but EPA never responded to these public comments. First Sass Decl. ¶¶ 15-18; Second Sass Decl. Ex. 2; Second Reeves Decl. ¶ 5; 2007 Petition at 3. NRDC and PANNA hoped that EPA would address the concerns raised in its IRED comments when it completed a cumulative risk assessment for all of the organophosphates. 2007 Petition at 3-4. However, EPA made no such changes when it finalized that cumulative risk assessment in 2006, even though by that

time, additional scientific studies and air monitoring confirmed the drift exposures and neuro-developmental risks posed by chlorpyrifos. *See* 2007 Petition at 4.

C. Petitions and Litigation to Obtain EPA Action on Evidence of Chlorpyrifos Health Risks.

Farmworker and health advocates then pursued three legal avenues challenging EPA's failure to protect children from the hazards posed by chlorpyrifos. First, United Farm Workers ("UFW") and other farmworker advocates filed a federal district court challenge to the 2001 chlorpyrifos interim re-registration decision, in part, for failing to protect children and other bystanders from pesticide drift. *UFW v. Adm'r, EPA*, No. 07-3950-JF (N.D. Cal. filed Aug. 1, 2007).[4] The parties negotiated principles on which the case could be settled with a commitment by EPA to make a new regulatory decision for chlorpyrifos by 2010 that would address drift exposures to children and other bystanders and human health risks from chlorpyrifos. However, after the Ninth Circuit ruled that challenges to FIFRA registration determinations must be brought in the courts of appeals within 60 days of the decision, the settlement fell apart, and the farmworker advocates voluntarily dismissed the district court chlorpyrifos challenge. *UFW*, Stipulation of Voluntary Dismissal, Dkt. 98, No. 07-3950-JF (N.D. Cal. filed April 27, 2010).

[4] NRDC and Earthjustice were among the co-counsel for the farmworker advocates.

Second, PANNA joined other farmworker advocates in petitioning EPA to address pesticide drift as mandated by the FQPA. *See* Pesticides In The Air – Kids At Risk: Petition to EPA to Protect Children From Pesticide Drift (October 13, 2009) (the "Kids' Petition") (Exhibit 5 without attachments). The Kids' Petition highlighted EPA's violation of its legal duty to protect children from all aggregate exposures to each pesticide in tolerance and reregistration determinations and asked EPA to expedite adoption of mitigation for airborne routes of exposure to organophosphates and n-methyl carbamates, another nerve poisoning pesticide, because of the heightened poisoning risks posed by those classes of pesticides. As is its pattern, EPA failed to respond to the petition until petitioners filed a writ of mandamus with this Court. *See In re PANNA*, No. 13-72616 (9th Cir. filed July 31, 2013); Agency Response to Pesticides In The Air – Kids At Risk: Petition to EPA to Protect Children From Pesticide Drift (2009) (March 31, 2014) (Exhibit 6). In that response, EPA acknowledged its legal obligation to address pesticide drift under the FQPA and FIFRA; however, it indicated it would not do so until it reviewed pesticide registrations and tolerance decisions as a matter of course and refused to impose interim protections during that years-long delay. EPA Response to the Kids' Petition at 2, 32-33.[5]

[5] PANNA and other farmworker advocacy groups filed an administrative objection on May 28, 2014, and an appeal in this Court challenging EPA's decision, *In re PANNA*, No. 14-71514 (filed May 29, 2014).

Third, on a separate track, PANNA and NRDC filed the 2007 Petition at issue here. That petition and its fate are described below. As with the position EPA took in response to the district court challenge to the 2001 chlorpyrifos registration decision and the Kids' Petition, EPA has acknowledged its legal obligation under the FQPA to address drift and volatilization as aggregate exposures and its failure to do so in the chlorpyrifos reregistration and tolerance decisions made in 2001 and 2006.

III. EPA'S HANDLING OF THE 2007 PETITION TO BAN CHLORPYRIFOS

On September 12, 2007, PANNA and NRDC submitted the 2007 Petition to EPA to compel EPA to ban chlorpyrifos based on the mounting evidence of risks from chlorpyrifos that were left unaddressed in its 2001 and 2006 regulatory decisions. In the absence of a petition (or a successful lawsuit), EPA would review the chlorpyrifos registration as part of its registration review program, which has a statutory deadline of 2022. 7 U.S.C. § 136a(g)(1)(A)(iii).[6] While EPA has again prioritized organophosphates and chlorpyrifos in particular in its schedule for registration review because of the serious public health risks (*see* EPA Response to the Kids' Petition at 14, 35), the 2007 Petition sought an immediate ban because the risks posed by chlorpyrifos cannot wait for the registration review.

[6] While registration review applies to the FIFRA registration, a pesticide may not be registered for a food use unless a tolerance is in place as to that food. Hence, a pesticide's tolerances may implicitly be part of the FIFRA registration review.

At its heart, the 2007 Petition raised two issues. First, the 2007 Petition raised EPA's failure to account for risks to children and bystanders from chlorpyrifos drift and volatilization, as required by the FQPA. In support of this obligation, the petition presented the California Air Resources Board's air monitoring reports and data, which documented concentrations above EPA's levels of concern near fields and in schoolyards, and community air monitoring, which showed widespread contamination in multiple locations and over a period of years, including in schoolyards. 2007 Petition at 17-21.

Second, the 2007 Petition (at 4-16) compiled mounting evidence documenting serious cognitive and behavioral effects from low-dose pre-natal chlorpyrifos exposures not captured in the studies used by EPA in its regulatory decisions. Peer-reviewed scientific studies have shown that children and infants exposed to chlorpyrifos can exhibit long-term neurological and neurodevelopmental difficulties, particularly from early life exposure. 2007 Petition at 6-14; *see also* First Sass Decl. ¶¶ 19-21; Second Sass Decl. ¶¶ 8-11. For example, two studies by Columbia University scientists documented decreases in birth weight, attention deficit disorder, hyperactivity, and delayed development in children exposed to chlorpyrifos *in utero*. 2007 Petition at 6-7. Scientists with Mount Sinai School of Medicine correlated *in utero* exposure to chlorpyrifos with reduced head circumference in newborns, which is predictive of impaired cognitive

ability. *Id*. at 7-8. These studies provide strong evidence that prenatal and early life-stage exposure to chlorpyrifos is associated with not only poor birth outcomes (lower birth weight and length), but also long-lasting, and possibly permanent, impaired cognitive development. *Id.* at 6-9, 11-13. Further, members of EPA's Scientific Advisory Panel expressed concern that EPA failed to account for scientific evidence showing brain impacts from early life exposures to chlorpyrifos at lower doses than those used by EPA in its regulatory decisions. *Id.* at 13, 22-23.

Shortly after PANNA filed the 2007 Petition, EPA found that the petition met the legal requirements for FFDCA petitions and filed a notice in the Federal Register requesting public comments. 72 Fed. Reg. 58,845 (Oct. 17, 2007). For the next three years, EPA failed to resolve the 2007 Petition, and in July 2010, PANNA filed a lawsuit, alleging that EPA unreasonably delayed responding to the 2007 Petition. *NRDC v. EPA*, No. 10-05590-CM, Compl., Dkt. No. 1 (S.D.N.Y. filed July 2010). On December 22, 2010, the parties executed a stipulation in which EPA agreed to complete a preliminary human health risk assessment for chlorpyrifos by June 1, 2011, and to respond to the 2007 Petition on or before November 23, 2011. *Id.* Dkt. No. 17, at 2-3 (Dec. 21, 2010) (Stipulation & Order Transferring Case to the Suspense Docket).

Following that stipulation, EPA released a preliminary human health risk assessment for chlorpyrifos for public comment. 76 Fed. Reg. 39,399 (Jul. 6,

2011). The preliminary human health risk assessment confirmed, as the 2007 Petition insisted, the importance of addressing drift, volatilization, and health impacts to children at low doses. Reader's Guide at 1-3 (July 1, 2011) (Attach. G to Housenger Decl.). The assessment expressed concern that current tolerances may not afford sufficient protection to children from drinking water and drift exposures. *Id*. at 2-3; Chlorpyrifos Preliminary Human Health Risk Assessment for Registration Review at 17 (June 30, 2011) (Attach. F to Housenger Decl.). As to the mounting evidence of neurodevelopmental impacts, EPA concluded that "chlorpyrifos likely played a role in long term neurological effects from early exposures that were evaluated in the epidemiology studies." Reader's Guide at 2-3.

Despite taking these preliminary steps, EPA failed to meet the agreed-upon November 2011 deadline for a final decision on the 2007 Petition. After EPA failed to meet the stipulated deadline, PANNA filed a writ of mandamus in the court of appeals based on a recent decision by the Ninth Circuit holding that jurisdiction over a challenge to the underlying determination would lie in the courts of appeals instead of the district courts. *NRDC v. EPA*, No. 10-05590-CM, ECF No. 21 (S.D.N.Y. April 16, 2012) (keeping case on the district court's suspense docket pending Ninth Circuit's resolution of the mandamus petition); *In*

re PANNA, Petition for Writ of Mandamus and For Relief from Unreasonably Delayed Action by EPA, No. 12-71125 (9th Cir. filed April 12, 2012).

On July 16, 2012, EPA issued a partial response to the 2007 Petition, promising a complete final response in December 2012. Letter of July 16, 2012, from Dr. Steven Bradbury, Director, EPA Office of Pesticide Programs, to Aaron Colangelo and Margaret Reeves, Ph.D ("First Interim Response") (Attach. J to Housenger Decl.). EPA's First Interim Response addressed six points made in the 2007 Petition but did not constitute a final response and did not determine whether EPA would ban chlorpyrifos. *See id.* The only practical effect of EPA's July 2012 partial decision consisted of EPA's announcement that the chlorpyrifos registrants had agreed to a spray drift mitigation package that calls for very small no-spray buffers (most were only ten feet) around school grounds, homes, residential lawns, athletic fields, nursing homes, hospitals, sidewalks, and other places frequented by bystanders. Spray Drift Mitigation Decision for Chlorpyrifos (July 2012) (Attach. K to Housenger Decl.). EPA then missed the December 2012 deadline for issuing a response to the 2007 Petition. *See* Letter of Dec. 18, 2012, from Dr. Steven Bradbury, Director, EPA Office of Pesticide Programs, to Aaron Colangelo and Margaret Reeves, Ph.D (Exhibit 7); Letter of Jan. 25, 2013, from Dr. Steven Bradbury, Director, EPA Office of Pesticide Programs, to Aaron Colangelo and Margaret Reeves, Ph.D ("Second Interim Response") (Exhibit 8).

In briefing before this Court, EPA promised to respond to the 2007 Petition by February 2014. This Court heard argument on the first mandamus petition in February 2013 and directed the parties to engage in mediation with the assistance of the Ninth Circuit mediator. After the mediation proved unsuccessful, the Court denied the mandamus petition on July 10, 2013. *In re PANNA*, 532 F. App'x 649 (9th Cir. 2013). The Court found that EPA "set forth a concrete timeline for final agency action that would resolve the 2007 Petition by February 2014." *Id.* at 651. In addition, the Court pointed to the lack of a statutory deadline for responding to petitions to revoke tolerances and the steps taken by EPA to work toward resolving the 2007 Petition. *Id.* The Court explicitly stated that its denial was "without prejudice to seeking the same relief at a future date in the event EPA fails to act." *Id.* at 652.

EPA missed its February 2014 deadline. In July 2014, EPA issued another partial response and reversed its earlier preliminary determination that chlorpyrifos volatilization presents risks warrant large, no-spray buffers, in some instances many thousands of feet around schools, homes, and other places frequented by people. EPA based this reversal on two new studies conducted by Dow AgroSciences LLC, one of the primary chlorpyrifos registrants. Letter of July 15, 2014, from Jack E, Housenger, Director, EPA Office of Pesticide Programs, to Aaron Colangelo and Margaret Reeves, Ph.D, at 2-4 ("Third Interim Response")

(Exhibit 9). In that partial response, EPA indicates that it now plans to release a revised human health risk assessment for public comment in December 2014, along with either a proposed rule revoking tolerances for chlorpyrifos or a proposed order denying the 2007 Petition. In its latest proposed deadline, EPA claims it will issue any final denial of the 2007 Petition by the summer of 2015. Third Interim Response at 5.

SUMMARY OF ARGUMENT

Seven years ago, PANNA filed a petition seeking a ban on chlorpyrifos based on serious health risks, particularly to children. The 2007 Petition presented scientific evidence of exposures to children from chlorpyrifos drift that EPA ignored when it made its 2001 and 2006 regulatory decisions, even though it now acknowledges it had a legal obligation to address drift exposures. The 2007 Petition also presented evidence of alarming neurodevelopmental impairments to children from chlorpyrifos, which EPA discounted in 2001 and 2006, and which has been further substantiated in the scientific literature since that time. EPA has conducted assessments and internal peer reviews and has made repeated promises to resolve the petition by deadlines that have long since passed, including the "concrete timeline" relied upon by this Court in denying the first mandamus petition. EPA's failure to make a final decision on the 2007 Petition leaves children at risk of harm from chlorpyrifos exposure and leaves PANNA without

legal remedies to challenge EPA's ongoing failure to take necessary steps to protect children.

Under the APA, EPA must act "within a reasonable time." EPA has not. Its delay has grown more unreasonable with each missed deadline and passing month. EPA's enduring delay demonstrates that only an order from this Court will result in final resolution of the 2007 Petition. The Court, therefore, has ample justification for directing EPA to resolve the 2007 Petition according to the timeline EPA has now set for itself.[7]

ARGUMENT

I. A WRIT OF MANDAMUS IS WARRANTED TO COMPEL EPA TO ISSUE A FINAL DETERMINATION ON THE 2007 CHLORPYRIFOS PETITION.

This Court generally employs a three-part test to determine whether to grant mandamus relief: (1) the petitioner's claim is clear and certain; (2) the duty is so plainly prescribed as to be free from doubt; and (3) no other adequate remedy is available. *In re Cal. Power Exch. Corp.*, 245 F.3d 1110, 1120 (9th Cir. 2001)

[7] PANNA and NRDC have standing to pursue this writ of mandamus because they are the organizations that filed the 2007 Petition. Both organizations are dedicated to reducing and eliminating harmful human exposures to hazardous pesticides, and both have members who have been exposed to chlorpyrifos and other organophosphates, who live in close proximity to fields where these pesticides are used, and/or who are concerned about exposure to chlorpyrifos that is not within their control. Decls. of Gina Trujillo, Sattie Clark, Sharon Bolton, Margaret Reeves and Jennifer Sass; *see Friends of the Earth v. Laidlaw Envtl. Servs.*, 528 U.S. 167, 180-81 (2000); *Citizens for Better Forestry v. U.S. Dep't of Agric.,* 341 F.3d 961, 969 (9th Cir. 2003).

(citing *Or. Natural Res. Council v. Harrell*, 52 F.3d 1499, 1508 (9th Cir. 1995); *Fallini v. Hodel*, 783 F.2d 1343, 1345 (9th Cir. 1986)). However, where a petitioner is seeking a writ of mandamus for unreasonable delay, this Court applies the so-called *TRAC* factors established by the D.C. Circuit in *Telecomm. Research & Action Ctr. v. FCC*, 750 F.2d 70, 75 (D.C. Cir. 1984) (hereinafter "*TRAC*"); *see In re Cal. Power Exch. Corp.*, 245 F.3d at 1124-25 (explicitly adopting the *TRAC* factors); *Independence Mining Co. v. Babbitt*, 105 F.3d 502, 507 (9th Cir. 1997) (same). This Court applied the *TRAC* factors to PANNA's earlier petition for writ of mandamus. *In re PANNA*, 532 F. App'x at 650-52.

Before turning to the *TRAC* factors, PANNA satisfies the threshold requirements set out in *In re Cal. Power Exch. Corp*. as EPA has a clear and certain duty to respond to the 2007 Petition that is plain and free of doubt, and there is no other adequate remedy for EPA's failure to do so. The FFDCA lays out a process for the public to petition to revoke a tolerance for a pesticide chemical residue or on a food, 21 U.S.C. § 346a(d)(1)(A), in practical effect banning the pesticide for that food use. The FFDCA directs EPA to take one of three actions in response to such a petition: (1) issue a final regulation modifying or revoking the pesticide tolerance; (2) "issue a proposed regulation" modifying or revoking the tolerance followed by a final regulation after notice and comment; or (3) issue an order denying the petition. *Id.* § 346a(d)(4)(A)(i)-(iii); *see In re PANNA*, 532 F.

App'x at 650 (recognizing EPA's duty to take one of these three actions in response to a petition to revoke pesticide tolerances). EPA has a clear duty to take one of these actions in response to the 2007 Petition. Failing to do anything is not an option.

The issue in this case is whether EPA has unreasonably delayed taking one of these actions by failing to issue a final response to the 2007 Petition after seven years and despite its many promised timeframes. The Administrative Procedure Act requires that federal agencies respond to petitions "within a reasonable time," 5 U.S.C. § 555(b), and authorizes agencies to "compel agency action unlawfully withheld or unreasonably delayed," *id.* § 706(1). To determine whether an agency has unreasonably delayed agency action, this Court applies the six *TRAC* factors:

> (1) the time agencies take to make decisions must be governed by a "rule of reason";
>
> (2) where Congress has provided a timetable or other indication of the speed with which it expects the agency to proceed in the enabling statute, that statutory scheme may supply content for this rule of reason;
>
> (3) delays that might be reasonable in the sphere of economic regulation are less tolerable when human health and welfare are at stake;
>
> (4) the court should consider the effect of expediting delayed action on agency activities of a higher or competing priority;
>
> (5) the court should also take into account the nature and extent of the interests prejudiced by the delay; and

> (6) the court need not "find any impropriety lurking behind agency lassitude in order to hold that agency action is unreasonably delayed."[8]

Independence Mining Co., 105 F.3d at 507 n.7 (quoting *TRAC*, 750 F.2d at 80). In light of EPA's failure to respond by its own, self-imposed "concrete" deadline and the passage of additional time since this Court's ruling on the first mandamus petition, the *TRAC* factors support issuance of a writ of mandamus holding EPA to its newly promised target dates for responding to the 2007 Petition since EPA has shown itself unwilling or unable to hold itself to any timeline.

A. EPA's Seven-Year Delay in Responding to the 2007 Petition is Excessive and Violates the Rule of Reason.

In the first mandamus proceeding, EPA argued that its response to the 2007 Petition was appropriately taking so long because the issues are complex, characterizing the evidence as at the edge of evolving science. Housenger Decl. ¶¶ 11, 20. Given that the issues and scientific studies were presented to EPA in 2007 and that many had been before the agency for many years prior to the 2007 Petition, it is no longer credible for EPA to claim novelty as an excuse for delay.

Moreover, EPA has a process for obtaining reviews from its Scientific Advisory Panel of the scientific evidence, for developing models and methods for integrating the evidence of harm into EPA's chlorpyrifos assessments, and for

[8] In applying these factors in response to the first mandamus petition, this Court noted that factor 6 need not be addressed as no allegation of impropriety has been made. *In re PANNA*, 532 F. App'x at 651-52.

eliciting public and industry input. The Housenger Declaration submitted in the first mandamus case in July 2012 walked through the various Scientific Advisory Panel reviews and EPA assessments of the drift, volatilization, epidemiological studies, and other studies demonstrating neurodevelopmental impacts from chlorpyrifos exposures. Housenger Decl. ¶¶ 12, 14-19. Those various reviews and assessments had either been completed or released for public comment in draft form. No additional Scientific Advisory Panel reviews are underway, which EPA cited as a key reason for the delay in the prior litigation. While this Court might be reluctant to interfere with EPA's chosen process for reviewing the scientific evidence and making a final decision, that process has now largely run its course.

In July 2012, based on the various reviews and assessments underway, EPA asserted that it could respond to the 2007 Petition by the end of 2012. Housenger Decl. ¶ 22. When that date passed, EPA represented to this Court that it could issue a final decision in February 2014; the Court relied on that representation, finding "EPA's subsequent response in this court has set forth a *concrete timeline* for final agency action that would resolve the 2007 Petition by February 2014." *In re PANNA*, 532 F. App'x at 651 (emphasis added). EPA missed that deadline.

After missing those deadlines, EPA now asserts that it can complete the next stage of its decision-making in December 2014 and a subsequent final stage by mid-2015. Third Interim Response at 5. Other courts have held agencies to their

own proposed deadlines, and it is appropriate for the Court to do so here. In one case, the D.C. Circuit held an agency to a deadline the agency proposed because the agency's "timetable representations [had] suffered over the years from a persistent excess of optimism, [and the court shared] petitioners' concerns as to the probable completion date." *Pub. Citizen Health Research Grp. v. Brock*, 823 F.2d 626, 629 (D.C. Cir. 1987). In making an agency's expected timeline mandatory in another case, the D.C. Circuit noted its "grave cause for concern that if [the court did] not insist on a deadline now, some new impediment will be pleaded." *In re Int'l Chemical Workers Union*, 958 F.2d 1144, 1150 (D.C. Cir. 1992). It is appropriate for the Court to hold EPA to this timeline and not let this latest deadline slip like the ones before it.

B. The 2006 Deadline for Ensuring EPA's Pesticide Authorizations Comply with the FQPA Shows that EPA's Delay Is Unreasonable.

TRAC provides that "where Congress has provided a timetable or other indication of the speed with which it expects the agency to proceed in the enabling statute, that statutory scheme may supply content for this rule of reason." *TRAC*, 750 F.2d at 80.[9] Here, although this Court previously found that no specific

[9] This factor does not ask whether Congress established a firm deadline for the challenged inaction, in which case balancing under *TRAC* would not be permitted. *See Biodiversity Legal Found. v. Badgley*, 309 F.3d 1166, 1177 n.11 (9th Cir. 2002). Rather, this factor asks whether the statutory scheme evinces a congressional intent that the agency should act more expeditiously.

deadline existed for responding to APA petitions, *In re PANNA*, 532 F. App'x at 651, the overall scheme of pesticide regulation gives the context and "other indication of speed" necessary to find EPA's delay unreasonable. The FQPA gave EPA ten years to bring all of its pesticide authorizations into compliance with the FQPA's requirements, including its mandate to consider all aggregate exposures and evidence of neurodevelopmental impacts to children and other special vulnerabilities. 21 U.S.C. § 346a(q)(1). While EPA re-registered chlorpyrifos and the other organophosphates by this August 2006 deadline, it did so without considering exposure to children from drift and volatilization and without accounting for the neurodevelopmental impacts to children already demonstrated by published scientific studies.

PANNA and NRDC filed comments on EPA's 2001 chlorpyrifos re-registration decision raising these issues and fully expected EPA to address them in connection with its cumulative risk assessment for the organophosphate pesticides, but EPA did not. First Sass Decl. ¶ 15-18; Second Reeves Decl. ¶ 5; 2007 Petition at 3-4. While the FFDCA and FIFRA establish no deadline for acting on a petition to revoke tolerances or cancel a pesticide registration, the 2007 Petition must be viewed against the backdrop of the FPQA's strict timelines for bringing EPA's pesticide authorizations into compliance with the FQPA's specific mandates for protecting children. *See Pub. Citizen Health Research Grp. v. Auchter*, 702 F.2d

1150, 1154, 1158 n.30 (D.C. Cir. 1983) ("The reasonableness of the delay must be judged 'in the context of the statute' which authorizes the agency's action.").

FIFRA also creates an obligation for EPA to review its pesticide registrations with a goal of doing so every 15 years and a hard deadline of 2022 for completion of the registration reviews of chlorpyrifos and all other pesticides re-authorized under the FQPA. 7 U.S.C. § 136a(g)(1)(A)(iii). However, EPA appropriately accelerated the chlorpyrifos registration review because of the seriousness of the issues presented in the 2007 Petition, Housenger Decl. ¶ 13, and because of the health issues posed by all organophosphates. EPA Response to Kids' Petition at 14, 35.

This statutory scheme supports an order compelling EPA to act by the current timeline it has set. Since the Court's prior order, EPA's delay has only gotten longer and its commitment to *any* self-imposed deadlines has grown even more questionable. When judged against the context of the statute, *Pub. Citizen Health Research Grp. v. Auchter*, 702 F.2d at 1154, 1158 n.30, EPA's seven-year failure to issue a final response is unreasonable. In light of EPA's failure to comply fully with the FQPA's mandates to consider all aggregate exposures and developmental impacts to children by the FQPA's 2006 deadline for doing so and its appropriate expedition of registration review of chlorpyrifos due to the serious

health issues presented in the 2007 Petition, the statutory scheme supports issuance of an order compelling EPA to act by the current timeline EPA has itself set.

C. The Health and Welfare of Those Suffering Ongoing Harms from Chlorpyrifos Support a Finding of Unreasonable Delay.

The 2007 Petition concerns human health and welfare—presenting evidence of major, ongoing health risks from chlorpyrifos that disproportionately affect communities of color and low-income communities—and asks EPA to take urgent action to protect children against ongoing harm from chlorpyrifos. Chlorpyrifos causes acute pesticide poisonings and remains one of the pesticides most often cited in pesticide poisoning reports. Second Reeves Decl. ¶ 7. EPA's 2001 and 2006 regulatory decisions acknowledged that chlorpyrifos exposure "can overstimulate the nervous system causing nausea, dizziness, confusion, and at very high exposures (e.g., accidents or major spills), respiratory paralysis and death." Attach. A to Housenger Decl. at 7. People living near areas where chlorpyrifos has been sprayed have experienced serious flu-like symptoms and other acute health effects, like rashes and difficulties breathing. First Reeves Decl. 1 ¶¶ 5-14.

In addition to acute poisoning effects, numerous published scientific studies correlate exposures of children and infants to chlorpyrifos with long-term neurological and behavioral impairments. 2007 Petition at 6-9; First Sass Decl. ¶¶ 19-21. Low-level exposures to chlorpyrifos early in childhood can lead to attention deficit disorder, hyperactivity, loss of IQ, and other cognitive

impairments. *See* First Sass Decl. ¶¶ 6-9, 19-20; 2007 Petition at 6-8. In its preliminary Human Health Risk Assessment, EPA acknowledges that "there is a growing body of literature with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent behavioral effects into adulthood." Attach. F to Housenger Decl. at 8. Further, EPA explains that "there is consistency across the animal behavior and epidemiology studies, such as delays in cognitive achievement, motor control, social behavior, and intelligence measures." *Id.*

In compelling agencies to put an end to delay, courts have concluded that "[w]hen the public health may be at stake, the agency must move expeditiously to consider and resolve the issues before it." *Pub. Citizen Health Research Grp. v. Comm'r, Food & Drug Admin.*, 740 F.2d 21, 34-35 (D.C. Cir. 1984); *In re Int'l Chemical Workers Union*, 958 F.2d at 1150 (court retained jurisdiction to enforce deadlines for regulating cadmium exposures after six-year delay).

Exposure to chlorpyrifos, a pervasive pesticide, is impossible to avoid. Chlorpyrifos is found in food and drinking water, in the air near agricultural communities, and in breast milk. *See* 2007 Petition at 4; First Sass Decl. ¶ 8. The risk of exposure is not limited to people who choose to buy or use products containing the pesticide; it can travel windborne from where it is sprayed, and it can be tracked inside the home on the shoes and clothes of people who come into

contact with its residues. First Sass Decl. ¶ 8; First Reeves Decl. ¶¶ 5, 9, 14. "Lack of alternative means of eliminating or reducing the hazard necessarily adds to unreasonableness of a delay." *See Cutler v. Hayes*, 818 F.2d 879, 898 (D.C. Cir. 1987).

In denying the first mandamus petition, this Court dismissed the health and welfare factor based on EPA's 2001 and 2006 chlorpyrifos determinations and because EPA operates almost entirely in the realm of human health and welfare. *In re PANNA*, 532 F. App'x at 651. As to the first point, EPA ignored the exposures and health effects identified in the 2007 Petition when it made its 2001 and 2006 decisions, and EPA has acknowledged health risks associated with exposing children to chlorpyrifos in its reviews and evaluations of the evidence presented in the 2007 Petition. *Supra* at 29. EPA's prior work, based on its own acknowledgements, did not consider all relevant paths to chlorpyrifos exposure and is, therefore, unreliable. Indeed, the D.C. Circuit noted in a case where it compelled an agency to act, that "[t]he risk to human life need not be a certainty to justify expedition [of agency action]." *Pub. Citizen Health Research Grp.*, 702 F.2d at 1158 n.26. It would be unseemly to allow EPA to try to minimize those risks in order to avoid a mandamus order before it has made a final determination

on the 2007 Petition based on an objective and complete evaluation of all the evidence.[10]

Moreover, while it is true that much of what EPA does involves human health, here EPA is addressing risks to children. When Congress passed the FQPA in 1996, it recognized that pesticide harm to children had been inadequately addressed. Congress changed that by requiring EPA to address all exposures, special sensitivities of children, and neurodevelopmental impacts, even before a full set of data is in hand. These heightened standards underscore Congress's concern about pesticides and children, above and beyond its normal human health docket.

D. No Higher, Competing Priorities Justify EPA's Delay.

In denying the first mandamus petition, this Court pointed to EPA's obligation to act on registration applications according to statutory deadlines. *In re PANNA*, 532 F. App'x at 650. However, justifications for delay "must always be balanced against the potential for harm," *Cutler*, 818 F.2d at 898, and an agency's "asserted justifications for the delay become less persuasive the longer the delay

[10] Further, if EPA can say with confidence that chlorpyrifos poses little risk, its delay in responding to the 2007 Petition becomes even less explicable. That is, if EPA has somehow determined that exposure to chlorpyrifos is not a major threat, such information should constitute a basis for denial of the 2007 Petition. Rather, as EPA has previously acknowledged and the evidence demonstrates, there are major health risks associated with exposing children and adults to chlorpyrifos, and this petition for a writ of mandamus should be read in light of those risks.

continues." *In re Int'l Chem. Workers Union*, 958 F.2d 1144, 1150 (D.C. Cir. 1992).

Here, EPA has statutory duties to protect children from pesticides and to comply with the FQPA's mandates, and Congress established a 2006 deadline for doing so. EPA failed to address drift, volatilization, and the neurodevelopmental impacts to children when it re-registered chlorpyrifos in 2001 and 2006, and PANNA and NRDC then filed the 2007 Petition to compel EPA to correct its failure.

Against this backdrop, EPA should not be able to claim that any competing priorities allow it to delay further its decision on the 2007 Petition. As the D.C. Circuit stated in *In re United Mine Workers*, "[h]owever many priorities the agency may have, and however modest its personnel and budgetary resources may be, there is a limit to how long it may use these justifications to excuse inaction in the face of the congressional command to act." 190 F.3d 545, 554 (D.C. Cir. 1999). EPA, of course, will always have competing duties, but it has yet to pinpoint any pesticide-related work that must take higher priority than evaluating the seven-year-old petition. EPA's continuing delay cannot be justified by any other priorities.

E. EPA's Delay Is Preventing Petitioners from Pursuing Administrative and Judicial Remedies to Protect Children from Harmful Chlorpyrifos Exposures.

The considerable adverse health risks attributed to chlorpyrifos have been set forth in detail above. The bottom line is more time has now elapsed, resulting in more exposures and greater risk of serious health impairments to children. The longer EPA waits, the more children will be exposed to chlorpyrifos.

It is important to note that a final response to the 2007 Petition will not end but instead begin the administrative process. Only after EPA's response will PANNA be able to begin to exhaust its administrative remedies by filing objections if EPA denies the 2007 Petition or by participating in the tolerance revocation process if EPA grants it. These steps are mandatory prerequisites to seeking judicial review. *See* 40 C.F.R. § 180.30(b). EPA should not be permitted to add its own obstacles by unreasonably delaying its response and thereby frustrating the statutory framework and PANNA's ability to seek judicial relief.

EPA's inaction leaves PANNA "stuck in administrative limbo; it enjoys neither a favorable ruling on its petition nor the opportunity to challenge an unfavorable one." *In re People's Mojahedin Organization of Iran*, 680 F.3d 832, 837 (D.C. Cir. 2012) (observing that the State Department's delay in resolving an organization's petition for revocation of its Foreign Terrorist Organization listing effectively insulated the decision from judicial review); *see also In re American*

Rivers, 372 F.3d 413, 420 (D.C. Cir. 2004) (allowing judicial intervention to end FERC's "marathon round of administrative keep-away").

To date, EPA has released "partial responses" that address some of the arguments and evidence put forward in the 2007 Petition. *See supra* at 18-20.[11] The 2007 Petition, however, sought an outcome—a chlorpyrifos ban—and EPA has yet to decide whether to pursue that outcome. EPA has failed to respond in any of the three legally permissible ways to respond to a petition to revoke tolerances, *see* 21 U.S.C. § 346a(d)(4)(i)-(iii), instead creating a barrier to judicial review through inaction.

EPA's pattern of moving the finish line just beyond the horizon violates the rule of reason. At various points over the last seven years, EPA committed to issue a final response by November 2011, December 2012, and February 2014. *See supra* at 17-20. This Court denied PANNA's first mandamus petition primarily because EPA represented to the Court that final action was forthcoming and would be completed by February 2014. *In re PANNA*, 532 F. App'x at 651 ("EPA's

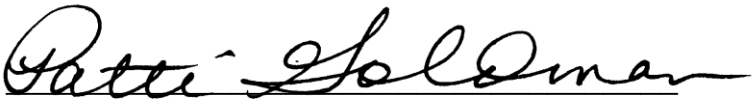
[11] In support of the requested ban, the 2007 Petition offered a series of inter-related and mutually reinforcing rationales supported by evidence. Second Sass Decl. ¶ 7. EPA has parsed the 2007 Petition and tried to divide it into discrete claims, but it would be incomplete and unresponsive to address each one in isolation. *See* Housenger Decl. ¶ 11 (stating that the remaining issues are "fundamentally intertwined" and "should not be addressed in isolation"). Until EPA addresses all shortcomings in EPA's 2001 and 2006 chlorpyrifos decisions by issuing a final decision on the whole of the 2007 Petition, PANNA is without legal recourse regardless of EPA's interim responses to PANNA's legal arguments.

subsequent response in this court has set forth a concrete timeline for final agency action that would resolve the 2007 Petition by February 2014."). EPA missed that timeline and now is proposing a new timeframe that pushes final agency action to mid-2015. It is appropriate for this Court to hold EPA to this new timeline and "let [the] agency know, in no uncertain terms, that enough is enough." *Pub. Citizen Health Research Grp.*, 823 F.2d at 627 ("When lives are at stake," as they are here, the agency "must press forward with energy and perseverance in adopting regulatory protections.").

CONCLUSION

PANNA asks this Court to hold EPA to its latest deadline and order EPA to respond to the 2007 Petition by: (1) releasing the revised human health risk assessment for public comment in December 2014, along with either a proposed revocation rule or a proposed denial of the petition; and (2) a final denial order by July 1, 2015, if that is how EPA decides to resolve the 2007 Petition.

Respectfully submitted this 10th day of September, 2014.



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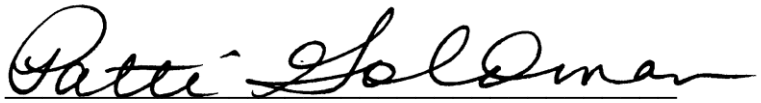
STATEMENT OF RELATED CASES

The undersigned, counsel of record for Petitioners Pesticide Action Network North America and Natural Resources Defense Council, Inc., are aware of no cases related to this petition pending before this court.

Respectfully submitted this 10th day of September, 2014.



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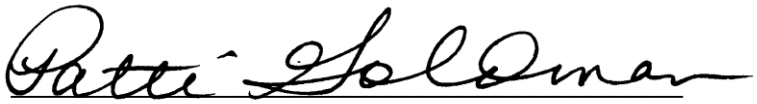
CERTIFICATE OF COMPLIANCE

This petition for writ of mandamus complies with the type-volume limitation of Fed. R. App. P. 32(a)(7)(B) because it contains 7,990 words, excluding the parts exempted by Fed. R. App. P. 32(a)(7)(B)(iii).

This petition for writ of mandamus complies with the typeface requirements of Fed. R. App. P. 32(a)(5) and the type style requirements of Fed. R. App. P. 32(a)(6) because it has been prepared in a proportionally spaced typeface using Microsoft Word 2010 Times New Roman 14 point font.

Respectfully submitted this 10th day of September, 2014.



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CERTIFICATE OF SERVICE

I hereby certify that I served the foregoing documents via United States Postal Service, electronic mail and/or overnight courier:

1. Petition for a Writ of Mandamus, including Corporate Disclosure Statement, Statement of Related Cases and Certificate of Compliance;
2. Declaration of Jennifer Sass, Ph.D., in Support of Petition for a Writ of Mandamus;
3. Declaration of Margaret Reeves, Ph.D., in Support of Petition for a Writ of Mandamus;
4. Declaration of Sattie Clark in Support of Petition for a Writ of Mandamus;
5. Declaration of Sharon Bolton in Support of Petition for a Writ of Mandamus; and
6. Declaration of Gina Trujillo in Support of Petition for a Writ of Mandamus

on the following parties:

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Executed this 10th day of September, 2014, at Seattle, Washington.

Eudora Powell

Eudora Powell

EXHIBIT 1

No. 12-71125

UNITED STATES COURT OF APPEALS

FOR THE NINTH CIRCUIT

PESTICIDE ACTION NETWORK NORTH AMERICA, and

NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

DECLARATION OF JACK HOUSENGER IN SUPPORT OF OPPOSITION TO PETITION FOR A WRIT OF MANDAMUS

I, Jack Housenger, state the following:

1. I declare that the following statements are true and correct to the best of my knowledge and belief and are based upon my personal knowledge and/or on my review of information contained in the records of the United States Environmental Protection Agency ("EPA" or the "Agency") or supplied by current employees.

2. I am the Director of the Health Effects Division ("HED") in EPA's Office of Pesticide Programs ("OPP"). I have worked for EPA for 33 years and previously worked for two years at the United States Department of Agriculture. Since February 1977, I have held various positions within OPP, including: Associate Director of the Special Review and Reregistration Division from March 1997 to June 2003; Associate Director of the Antimicrobials Division from June 2003 to May 2005; Associate Director of HED from May 2005 to January 2009; and acting Director and then Director of the Biological and Economic Analysis Division ("BEAD") from January 2009 to June 2012. I have been the Director of HED since July 2, 2012.

3. HED is the division assigned with the responsibility to develop EPA's human health risk assessments for both the approval (registration) and re-evaluation of conventional pesticides under the Federal Insecticide, Fungicide and Rodenticide Act ("FIFRA") and for the establishment of pesticide residue tolerances under the Federal Food, Drug and Cosmetic Act ("FFDCA"). This

involves evaluating the potential risks (taking into account hazard and exposure) from dietary exposures (food and water), from exposures to workers (including persons exposed when handling pesticides as well as those entering areas previously treated with pesticides (to scout fields, harvest crops, etc.)), and from any non-occupational (e.g., residential) exposures associated with pesticide chemicals.

4. Chlorpyrifos is a broad-spectrum organophosphate ("OP") insecticide, acaricide and miticide that was first registered under FIFRA in 1965 to the Dow Chemical Company. Since that time, numerous additional companies have obtained registrations both for "manufacturing use" products and the "end-use" products used by growers and other consumers.[1]

5. Chlorpyrifos has been and remains one of the most widely used insecticides in the U.S. As a result of EPA risk-reduction activities and changes in agricultural practice, however, overall use of chlorpyrifos in the U.S. is a fraction of historical use, declining from over 20 million pounds of active ingredient annually in the

[1] See U.S. EPA, Chlorpyrifos Interim Reregistration Eligibility Determination, at viii, 3, included as Attachment A.

1980s and early 1990s to roughly 8 millions pounds today. [2] Currently registered uses include primarily food and feed crops.[3]

6. Chlorpyrifos, like all OPs, achieves its pesticidal effect by inhibiting the nervous system enzyme acetylcholinesterase ("AChE"), an enzyme necessary for the proper functioning of the nervous system in target pests as well as non-target organisms, including humans. At high enough doses, chlorpyrifos can reduce AChE activity sufficiently to cause adverse effects to both humans and wildlife. Fortunately, AChE inhibition can be measured at very low levels in the blood, allowing EPA to use AChE inhibition as a sensitive biomarker in developing appropriately safe levels of exposure to humans, consistent with the strict standards required by FIFRA and the FFDCA.

7. In connection with EPA's evaluation and re-evaluation of pesticides under FIFRA and the FFDCA, HED will generally perform a quantitative risk assessment that evaluates both potential exposure and hazard to assess the likelihood that humans – including infants and children -- could be adversely affected from exposures to pesticides. To that end, HED needs both reliable and quantifiable exposure and toxicity data from different life stages to inform hazard endpoint selection, exposure-response assessment, uncertainty factor determination, and

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[2] See id. at 8; Evaluation of the Potential Risks from Spray Drift and the Impacts of Potential Risk Reduction Measures at 7, included as Attachment B.

[3] See IRED at 7-9.

overall risk characterization. Generally, this requires controlled, reproducible scientific data. With respect to the assessment of human health risks due to OPs and other AChE inhibiting pesticides, our risk assessments have taken a conservative approach that tries to find, through appropriate dosing in controlled experiments with laboratory animals, exposure level data where statistically significant AChE inhibition occurs. Using this approach for chlorpyrifos, EPA evaluated laboratory data to determine the dose at which 10% AChE inhibition is expected to occur. That is known as the point of departure ("PoD"), which is then divided by a number of uncertainty factors to account for possible differences among individual humans and between humans and test animals.

8. EPA considers all relevant and reliable sources of scientific data in conducting risk assessments. Although considered qualitatively to inform risk management decisions in the past, the Agency has generally not included epidemiologic data from literature studies in its quantitative risk assessment process for pesticides because these data have traditionally not been sufficiently robust and reliable for quantitative dose response assessment due to the inherent uncertainties and limitations in such information. However, in the past few years, with advancements in design, conduct and analyses (particularly with respect to exposure assessment methods), some newer epidemiologic studies may be adequate for consideration in risk assessment. In acknowledging these more recent

studies, the Office of Pesticide Programs recently (February 2010) developed a draft framework for considering epidemiologic data in hazard and risk characterization to ensure that the data are reviewed in a transparent, consistent and scientifically rigorous manner. While both observational epidemiology and experimental toxicology share the goal of determining whether a relationship exists between a pesticide exposure and an adverse health outcome, there are significant differences in design, conduct, method of analysis and areas of uncertainties (*e.g.*, exposure measurements, potential for systematic biases in epidemiologic data) between the two disciplines which present considerable challenges to integrating the epidemiologic data into regulatory risk assessment. EPA is taking a scientifically defensible and deliberative approach to fully understand and appropriately consider the strengths and weaknesses of currently available pesticide epidemiology, including studies reflecting prenatal exposure to chlorpyrifos and adverse neurodevelopmental effects in children. As this body of information is relatively new to EPA, the issues raised in the petition with respect to the quantitative use of epidemiologic data are truly novel for the regulation of pesticides in the United States.

9. In 2006, EPA completed its most recent re-evaluation of chlorpyrifos consistent with the reregistration requirements of FIFRA section 4 and the food residue tolerance reassessment requirements of FFDCA section 408(q). During the

early stages of that review, in 2000, nearly all residential uses of chlorpyrifos were cancelled and certain crop uses were substantially limited. In 2001, in connection with EPA's development of its interim reregistration eligibility determination ("IRED"), the chlorpyrifos registrants agreed to additional risk mitigation on product labeling, including additional worker and ecological risk protections. With that mitigation in place, EPA found the exposures to chlorpyrifos were consistent with both FIFRA and FFCDA standards. However, because all OPs share a common mechanism of toxicity -- AChE inhibition – EPA was also required to conduct a cumulative risk assessment (CRA) that evaluated the combined human dietary and non-occupational exposures to all the OPs. In August 2006, EPA determined that these cumulative exposures were also safe. Importantly, in both the aggregate assessment and the CRA, EPA also concluded that exposures to infants and children were safe, taking into account the special emphasis that the FFDCA places on ensuring that EPA's decisions are protective for these early life stages. At the same time, EPA announced that it had also completed its FIFRA reregistration eligibility determination for chlorpyrifos, finding that existing registrations were eligible for reregistration under the FIFRA "no unreasonable adverse effects" standard.

10. In September 2007, Petitioners in this action, Pesticide Action Network North America and the Natural Resources Defense Council (hereinafter

"Petitioners"), submitted to EPA a petition seeking the revocation of all FFDCA food residue tolerances for chlorpyrifos and the cancellation of all the chlorpyrifos pesticide product registrations under FIFRA. That petition was in large measure styled as a challenge to the safety findings regarding human health that EPA had made in reregistering the chlorpyrifos registrations and maintaining the chlorpyrifos tolerances in connection with tolerance reassessment under FFDCA section 408(q). Petitioners raised 10 bases or claims for cancellation and revocation that effectively challenged numerous aspects of EPA's human health findings in those decisions: i) EPA ignored genetic evidence of vulnerable populations, ii) EPA needlessly delayed a decision regarding endocrine disrupting effects, iii) EPA ignored data regarding cancer risks, iv) EPA's cumulative risk assessment misrepresented risks and failed to apply an appropriate safety factor for children; v) EPA over-relied on registrant data, vi) EPA failed to properly address the exporting hazard from chlorpyrifos, vii) EPA failed to quantitatively incorporate data demonstrating long-lasting effects from early life exposure to chlorpyrifos in children, viii) EPA disregarded data demonstrating that there is no evidence of a safe level of exposure during pre-birth and early life stages, ix) EPA failed to cite or quantitatively incorporate studies and clinical reports suggesting potential adverse effects below 10% cholinesterase inhibition, and x) EPA failed to incorporate inhalation routes of exposure.

11. While effectively styled as a challenge to the 2006 reregistration and tolerance reassessment decisions, some of the issues involve matters that were, and remain, on the edge of evolving science. For example, the assertion that EPA failed to quantitatively incorporate data on long-lasting early life exposures in its risk assessment raised entirely novel questions about the use of epidemiologic data in risk assessments. Most of the epidemiology studies at issue were published and supplemented over the several years following the issuance of the IRED. Indeed, the declaration of Dr. Jennifer Sass filed by Petitioners acknowledges as much by effectively supplementing the 2007 petition with citations to the three most recent of these epidemiologic studies completed in 2011. Similarly, the extent to which adverse effects can be attributed to doses lower than those that elicit 10% AChE inhibition (the measure currently used as a PoD for regulatory purposes) has been and remains a question of evolving science. The resolution of Petitioners' assertion that "no safe level" of chlorpyrifos exists is fundamentally intertwined with these two issues and therefore EPA believes it should not be addressed in isolation. Finally, the assertion that EPA failed to quantitatively assess inhalation exposure to bystanders from "chlorpyrifos-contaminated air" was effectively a request that EPA require new field studies to measure volatilization exposures and develop a new risk assessment methodology where none previously existed at the time of the petition. While data and methodological tools existed for analyzing

field worker inhalation exposure, field studies and risk assessment methodologies for accurately estimating off-field bystander exposures to volatilizing chlorpyrifos particles following application had not been developed or peer reviewed at the time of the RED decision in 2006.[4]

12. In light of the evolving state of science on the issues identified above, EPA concluded that it could not simply affirm or reject these claims without further evaluation and without the development of new scientific methodologies. Indeed, prior to the petition, EPA had already begun looking at some of the new data cited in the petition and was coming to the conclusion that it would need to review the new data in depth and explore new methodologies for analyzing the data in order to determine whether the data could be used meaningfully in the risk assessment process during the next re-evaluation of chlorpyrifos. Therefore, EPA concluded that the appropriate course of action was to explore Petitioners' assertions in depth and to seek input from the FIFRA Scientific Advisory Panel ("SAP") – the peer review body created by Congress that provides guidance to EPA on pesticide

[4] EPA had previously sought SAP review in 2004 in connection with the evaluation of fumigant pesticide volatilization. Because fumigants are inherently volatile and applied in a manner and under conditions that are considerably different from the techniques used to apply conventional agricultural pesticides like chlorpyrifos (e.g., fumigant applications require the use of techniques such as soil injection and the use of tarps to retain the fumigant whereas pesticides such as chlorpyrifos are sprayed on crops from aerial or ground equipment), the results of that review were not directly transferable.

regulatory science -- in EPA's efforts to develop methodologies for determining whether Petitioners' claims have merit.

13. Under section 3(g) of FIFRA, EPA is tasked with re-evaluating pesticides on a 15-year cycle. Having completed the reregistration of chlorpyrifos and the other OPs in 2006, by law, EPA is not actually required to complete another review of these pesticides under section 3(g) until 2022. However, as noted, EPA understood that there were ongoing science issues with the OPs, and chlorpyrifos in particular, that needed further study. And given EPA's decision to embark on the review of the novel and evolving scientific issues raised in the petition, as discussed above, EPA decided that it was also appropriate to move up the reregistration review of chlorpyrifos in order to complete that review several years in advance of the 2022 deadline. Indeed, EPA's 2009 preliminary work plan for chlorpyrifos identified responding to the petition as one of the purposes of that early review.

14. In September 2008, in connection with both EPA's ongoing re-evaluation of chlorpyrifos and its review of the petition, EPA convened an SAP meeting to review a draft science issue paper on the human health effects of chlorpyrifos to provide a preliminary review of the scientific literature on experimental toxicology and epidemiology available at that time, including studies by Columbia University

and Mount Sinai School of Medicine researchers regarding the effects of chlorpyrifos on birth outcomes and child development. Specifically, the focus was on studies that evaluated the effects of chlorpyrifos on infants and children from *in utero* and/or post-natal exposures and on studies that evaluated population variability with respect to response to paraoxonase ("PON1") – issues raised in the petition.[5] In summary, the SAP expressed confidence that the studies conducted by Columbia University are epidemiologically sound. The SAP agreed with the Agency that human epidemiologic studies have utility for risk characterization, but not as the principal basis for establishing the PoD, in part due to uncertainty in attributing observed adverse neurodevelopmental effects in children solely to chlorpyrifos, when exposure was to multiple anticholinesterase insecticides. Importantly, however, the SAP recommended that EPA "conduct a full formal weight of evidence evaluation for causality of the reported associations between exposure to chlorpyrifos and neurodevelopmental outcomes in the existing epidemiological database."[6] As described in paragraph 18, EPA is currently

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[5] Paraoxonase is an enzyme that detoxifies chlorpyrifos. Paraoxonase activity may differ as between individuals and Petitioners assert that EPA has not properly addressed this potential variability in determining safe levels of human exposure to chlorpyrifos.

[6] Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held September 16-18, 2008 on the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos at 14, included as Attachment C.

following up on this recommendation and has recently presented its preliminary conclusions to the SAP.

15. To help address another issue of evolving science raised in both the petition and the chlorpyrifos registration review, in December 2009, EPA convened the SAP to review scientific issues associated with interpreting risks related to the field volatilization of conventional pesticides.[7] The objectives of the meeting were to review both the exposure and hazard aspects of determining the inhalation risk associated with field volatilization. While Petitioners had submitted some field monitoring data to support their petition, EPA realized that these data were likely too limited in scope to support an assessment to determine the appropriateness of the label use limitations nationwide, as Petitioners' cancellation and revocation petition effectively requested. The primary focus of the discussion on exposure assessment was therefore on methods for predicting emissions from treated fields in lieu of having numerous field volatilization studies as well as how such information should be considered in exposure assessment. With regard to hazard evaluations, the SAP considered the impact on inhalation risk estimates based on differences in how doses are experimentally administered to rodents (oral or inhalation). The Agency's goal for the SAP review was to receive feedback on

[7] See Transmittal of Meeting Minutes of the FIFRA SAP Meeting Held December 1-3, 2009 on the Scientific Issues Associated with "Field Volatilization of Conventional Pesticides,." included as Attachment D.

procedures, methodologies, and data inputs to inform the assessment of bystander exposure resulting from field volatilization of conventional pesticides, such as chlorpyrifos. The procedures, refined in part from the SAP's feedback, will inform the Agency's analysis as it considers chlorpyrifos emissions data identified in the literature, as well as a new field volatility study submitted by Dow AgroSciences early in July 2012.

16. In order for EPA to consider incorporating the chlorpyrifos epidemiologic data into the chlorpyrifos risk assessment (which is what Petitioners are requesting), EPA first had to establish a framework for undertaking such an effort in a transparent and rigorous manner. In February 2010, EPA convened an SAP to review the Agency's draft "Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment,"[8] which provides the conceptual foundation for evaluating and integrating multiple lines of scientific evidence in a human health risk assessment. This draft framework explicitly considers such concepts as strength, consistency, dose response, temporal concordance and biological plausibility. While EPA did not conduct this SAP for the express purpose of addressing the petition (in fact, the pesticide being analyzed in this

[8] See Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting on the Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment, included as Attachment E.

exercise was the herbicide atrazine), EPA could not reasonably have taken steps to evaluate the integration of the epidemiologic data into its risk assessment without building a transparent and scientifically defensible assessment framework for conducting such an evaluation – so this SAP meeting also represented a critical step in responding to the petition.

17. In July 2011, EPA released for public comment its preliminary Human Health Risk Assessment ("HHRA") for chlorpyrifos as part of the registration review process.[9] That assessment also effectively served as an interim response or status report on EPA's progress in responding to the issues raised in the petition. In sum, while the preliminary assessment indicated potential concerns that appropriate margins of safety may not exist in some cases, EPA's analysis did not suggest that serious harm was occurring that required immediate, broad regulatory action of the sort requested by Petitioners. Specifically, the assessment indicated that, in using AChE inhibition as the PoD, aggregate exposures in food were within appropriate margins of safety. However, the assessment indicated the potential for drinking water exposures exceeding appropriate safety margins in sensitive watersheds – which was an issue not raised in the petition. Further, the assessment also indicated that exposure to pesticide drift and volatilization may exceed appropriate margins of safety for bystanders in areas near where chlorpyrifos is

[9] The HHRA is included as Attachment F.

applied to crops. It is important to note that the assessment also made clear that all of these preliminary conclusions were the subject of extensive ongoing review. In the Reader's Guide to the HHRA,[10] EPA identified in detail the specific steps it was planning to take to evaluate the appropriate PoD, refine its drinking water assessment, and develop a broader approach for modeling bystander inhalation exposure on a national level. With respect to the PoD and bystander inhalation exposure, these constituted the same steps EPA was – and is – taking to complete its review of the petition. The HHRA public comment period officially closed on October 6, 2011, with the Agency receiving 48 unique comments totaling over 1000 pages and including citations to a number of significant new studies. The Agency continues to work on reviewing the public comments and studies cited to further inform the final HHRA for chlorpyrifos -- which will include both a dietary and occupational risk assessments -- currently scheduled for completion in 2014.[11]

18. As noted in paragraphs 12-14, since the 2008 SAP on chlorpyrifos, and in part due to the SAP's feedback, the Agency has performed further analyses on the existing and new epidemiologic data (studies published prior to and after the 2008 FIFRA SAP meeting on chlorpyrifos), available biomonitoring data, and experimental toxicology studies evaluating proposed adverse outcome pathways in

[10] The Reader's Guide to the HHRA is included as Attachment G.
[11] As explained in paragraph 21, EPA intends to complete the dietary and non-occupational component of that assessment in 2013.

the context of human health risk assessment. Specifically, consistent with the request in the petition, the Agency is evaluating available literature on the potential for chlorpyrifos to cause long term adverse effects from early life exposure , *in vivo* and *in vitro* studies evaluating mechanistic aspects of chlorpyrifos, and the potential for adverse effects below PoDs established from cholinesterase inhibition that are used for regulatory purposes. This analysis is complicated and multifaceted as it involves many lines of scientific evidence (*i.e.*, *in vivo & in vitro* experimental toxicology studies including neurodevelopmental studies in laboratory animals, explicit consideration of adverse outcome pathway framework analyses, exposure, human epidemiology, and biomonitoring data). As noted in the July 2011 preliminary risk assessment, as the Agency works to finalize the HHRA and respond to the remaining petition issues, the Agency is generating a weight-of-evidence evaluation integrating the epidemiologic data with the experimental toxicology studies for the neurodevelopmental outcomes and AChE inhibition. In connection with that review, EPA convened a FIFRA SAP in April 2012 to review the Agency's preliminary conclusions on these issues.

19. As presented at the April 2012 SAP meeting, EPA's preliminary conclusion is that chlorpyrifos likely played a role in long term neurological effects from early

exposures that were evaluated in the epidemiology studies.[12] It is important to understand, however, that the epidemiology studies at issue were evaluating effects that likely resulted primarily from now banned residential uses of chlorpyrifos, so EPA cannot simply assume that the results of these studies are transferable to the remaining agricultural uses of chlorpyrifos. In order to address whether EPA could quantitatively incorporate the results of the epidemiology studies into its risk assessment for currently approved uses-- either to inform the PoD or EPA's use of uncertainty factors -- EPA explained that additional work would be required to determine whether it is possible to estimate the chlorpyrifos dose levels to which the study participants may have been exposed. EPA would then have to compare those levels to levels resulting in 10% AChE inhibition – the current PoD EPA uses in its risk assessment – to evaluate whether the neurobehavioral effects likely occurred at levels below the existing PoD. That evaluation, if possible, would allow EPA to determine whether the existing PoD is protective of neurodevelopmental effects. On July 12, EPA received the report of the April 2012 SAP.[13] EPA has had insufficient time since then to fully analyze the SAP

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[12] See Meeting of FIFRA Scientific Advisory Panel Draft Issue Paper: Scientific Issues Concerning Health Effects of Chlorpyrifos, April 10-13, 2012, at 102-103, included as Attachment H.

[13] See Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held April 10-12, 2012 on "Chlorpyrifos Health Effects," included as Attachment I.

recommendations and how those recommendations will impact the Agency's work to complete its full weight-of-the-evidence analysis of the effects of early life exposure to chlorpyrifos on long-term child neurodevelopmental effects. EPA will need to carefully consider the SAP report and recommendations before deciding precisely how to proceed.

20. EPA readily acknowledges that it had hoped to complete review of the petition sooner and that it had attempted to resolve Petitioners' earlier legal challenge in the Southern District of New York by completing its review in November of last year. However, in late 2011, following the initial review of the comments on the HHRA, EPA came to the conclusion that while it could complete review of most of the petition issues in a relatively short period of time, addressing the more complex toxicology and inhalation exposure issues raised in the petition still required additional evaluation and peer review given the truly cutting edge nature of these issues. While petitioners have not expressed the desire that EPA respond in serial fashion to their claims, EPA concluded that it would be appropriate to address and resolve certain issues sooner than others and to announce those conclusions when they are completed rather than holding them in reserve until all issues can be resolved. As a result, EPA developed an approach for addressing six of the ten petition issues in a partial response. On July 16, 2012,

EPA issued its partial response to the petition addressing six of the ten issues raised in the petition.[14]

21. The partial response indicated EPA's intention to deny Petitioners' claims (i) through (vi) as identified in paragraph 10 above. Because EPA has not yet published the partial response letter in the Federal Register, as required for FFDCA petition denials, the partial response indicated that it only constitutes final action with regard to the sole issue in the petition that is subject to judicial review under FIFRA – that is, the assertion that EPA must cancel all chlorpyrifos registrations to avoid exporting hazards overseas. For both the convenience of Petitioners and the Agency, EPA does not presently intend to issue a formal denial in the Federal Register for the five issues subject to FFDCA review until after it completes its review of all remaining issues. That way, Petitioners will not be compelled to assess whether to file objections to EPA's responses on separate occasions and EPA will not be compelled to produce separate responses. However, as noted in the partial response to the petition, if Petitioners wish to begin the objections process on EPA's partial response and so notify EPA in writing, EPA will publish a formal denial order for those claims, triggering their right to file objections under FFDCA section 408(g)(2).

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[14] Letter of July 16, 2012, from Dr. Steven Bradbury, Director, EPA Office of Pesticide Programs, to Aaron Colangelo and Margaret Reeves, Ph.D., included as Attachment J.

22. The partial response also explained EPA's approach for addressing the four remaining petition issues, indicating that EPA intends to provide a written response to those issues by December 2012. This response will be in the form of a letter, like the July 16, 2012, partial response. The partial response also explained:

> While [the December] response will address all the specific issues raised in the petition, it is possible that EPA will not be taking final agency action on the petition in connection with [that] response such that it would trigger a party's ability to file objections. While petitioners have raised a number of issues related to the assessment of the toxicity of chlorpyrifos, the petition did not address in any detail, dietary exposure to chlorpyrifos that must be taken into account in determining whether tolerances are safe. Reassessing the exposure to chlorpyrifos is one of the issues EPA intends to address in the registration review assessment of the chemical, which is currently underway. It is possible that if EPA concludes that the toxicity profile of chlorpyrifos needs to be modified based upon this reassessment, the final decision on the petition would need to wait for the conclusion of the chlorpyrifos exposure reassessment under the registration review program. The registration review assessment will include consideration of issues not raised in the petition, including human dietary exposures from drinking water. It is important to note, however, that EPA may take regulatory action at any time if and when it determines that existing tolerances are unsafe or that chlorpyrifos presents unreasonable adverse effects on the environment.

What this language is effectively saying is that to the extent EPA agrees with Petitioners that changes to the chlorpyrifos toxicity profile may be necessary, EPA will quite possibly not be in a position to determine whether all chlorpyrifos exposures remain within appropriate margins of safety until it completes its assessment of dietary and other non-occupational exposures -- including its

consideration of drinking water exposures. Therefore, any rulemaking action to revoke or modify tolerances, if determined to be necessary, may have to await the completion of that exposure assessment. The dietary and non-occupational assessment is scheduled to be completed by the end of 2013 (with the full human health risk assessment to be completed in 2014, as noted in paragraph 17 above). Any rulemaking action to revoke or modify tolerances, if necessary, would then follow the dietary and non-occupational risk assessment, building in sufficient time for EPA to complete the administrative review requirements necessary to publish a proposed rule (including any required review under the Small Business Regulatory Enforcement Fairness Act and Executive Order 12,866). However, to the extent EPA disagrees with the remaining petition claims, EPA would take final agency action denying the entire petition shortly after its planned December 2012 response, building in sufficient time to develop and publish the Federal Register notice announcing the order as required by FFDCA section 408(d)(3). Accordingly, EPA plans to take the following approach for completing action on the remaining issues in the petition: (1) If, in our December 2012 written response to Petitioners, we agree with Petitioners that the toxicity profile of chlorpyrifos needs to be modified, we intend to take one of three actions required by FFDCA 408(d)(4) (denial of the petition[15], a proposed rule revoking or modifying existing

[15] It is possible that even if EPA agrees that the toxicity profile needs to be

tolerances, or a final rule revoking or modifying existing tolerances) not later than February 2014. (2) Conversely, if EPA denies the remaining issues in its December 2012 written response to Petitioners, EPA intends to issue a denial order in the Federal Register not later than February of 2013.

23. In addition to issuing its partial response, on July 16, 2012, EPA also took action to address in part a seventh issue raised in the petition – inhalation risk to bystanders in areas where chlorpyrifos is applied. Specifically, EPA issued its Evaluation of the Potential Risks from Spray Drift and the Impacts of Potential Risk Reduction Measures[16] that updates the preliminary human health risk assessment with respect to bystander inhalation and dermal risk from "primary" spray drift – that is, the drift that occurs at the time of application. In connection with this assessment, the chlorpyrifos registrants have agreed to amend product labeling to reduce application rates and impose no-spray buffers that will mitigate risk from primary drift.[17] EPA acknowledges that its drift assessment and associated risk reduction action do not constitute a complete response to the bystander inhalation risk issues raised in the petition since there are two

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modified, the assessment EPA intends to complete in 2013 may show that exposures are still at safe levels. Because that work has not been completed, EPA must leave open the possibility that it may still deny the petition even if it agrees with Petitioners' claims with respect to toxicity.

[16] This document is included as Attachment B.

[17] See Spray Drift Mitigation Decision for Chlorpyrifos, July 2012, included as Attachment K.

components to that risk: (1) risks from primary drift and (2) risks from chlorpyrifos volatilization following application. EPA is continuing to assess the complex volatilization issue and has recently received data from Dow AgroSciences that it will be evaluating in completing that review. Further, the drift risk assessment will be informed by the continuing assessment EPA is conducting this year to determine whether the current PoD (10% AChE inhibition) is protective. As noted above in paragraph 22, EPA anticipates completing that work and providing a written response to Petitioners regarding its position on all the remaining issues by December of this year.

24. Given the complexity and importance of the scientific issues raised in the petition, we believe our approach for addressing these issues has been appropriate and that the record of significant science reviews of these issues over the past four years demonstrates that EPA has in fact moved diligently to address these issues. As noted throughout this declaration, these efforts have included four SAP reviews[18], the issuance of a preliminary human health risk assessment, an updated

[18] EPA held one additional SAP on chlorpyrifos-related toxicology issues in February 2011. That SAP addressed a source-to-outcome model developed by Dow AgroSciences designed to estimate the probability of inhibition of cholinesterase in humans. Chlorpyrifos was used as an example pesticide because there exists a comparatively rich data set on cholinesterase. While such research efforts may inform future EPA risk assessments, the purpose of this meeting was not intended to address issues in the petition or review EPA science that serves as a basis for EPA's ongoing reregistration review human health risk assessment.

evaluation of pesticide spray drift risk (together with mitigation action to address that risk) and a response to six of Petitioners' ten issues. Petitioners are apparently arguing that it should not reasonably require four and one-half years' time to complete all these activities and respond to the petition. It must be understood, however, that our work on chlorpyrifos has to be balanced with our other pesticide regulatory responsibilities. This includes re-evaluating the over 1000 other pesticide active ingredients contained in registered pesticide products under the periodic registration review requirements of FIFRA section 3(g).[19] In addition, EPA also must devote considerable scientific resources to the evaluation of new and amended products. Under FIFRA section 33, as amended by the Pesticide Registration Improvement Renewal Act of 2007, EPA must make registration decisions on applications for new and amended pesticide products on time frames that run between three months and two years, depending on the nature and complexity of the action requested. In the past three years, EPA has received over 1500 applications annually that are subject to the requirements of section 33.[20] While many of those actions may require only minor scientific review (such as

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[19] See Program Highlights from EPA's Pesticide Registration Review webpage (current as of April 2012), available at http://www.epa.gov/oppsrrd1/registration_review/highlights.htm.

[20] See Implementing the Pesticide Registration Improvement Act 2011 Annual Report, Table III , available at http://www.epa.gov/pesticides/fees/2011annual_report/table3-actions-completed.pdf.

products that are substantially similar to currently registered products), the majority of those actions are either for new products, amended products or other action requiring significant scientific review, including, in many cases, the establishment of new tolerances under the FFDCA. Unlike the petition response, Congress has given EPA a statutory deadlines for completing actions under both section 3(g) and section 33 and EPA must, as a result, devote the vast majority of its scientific review resources to activities under these sections to meet its legal obligations.

25. Finally, it also bears mentioning that EPA's work in responding to the petition over the past several years had to be balanced with the need to address other administrative petitions. As Petitioners mention in their brief, NRDC has in fact filed numerous petitions and/or objections seeking to revoke tolerances and cancel pesticides over the past decade.[21] As is clear, some of these were filed or addressed contemporaneously with the chlorpyrifos petition and, while not as complex as the chlorpyrifos petition, required considerable HED science resources to complete. On top of these petitions, in the years that the chlorpyrifos petition has been pending, EPA has had to review over a dozen other administrative petitions

[21] See, e.g., 73 Fed. Reg. 5439 (January 30, 2008)(denial of NRDC objections to issuance of a tolerance for the fungicide boscalid); 75 Fed. Reg. 55997 (September 15, 2010) (order denying NRDC objections to EPA denial of NRDC petition to revoke tolerances for the insecticide carbaryl); 77 Fed. Reg. 23135 (April 18, 2012) (order denying NRDC petition to revoke tolerances for the herbicide 2, 4-D).

that involve significant scientific review. Had EPA not been required to review those petitions at the same time Petitioners filed the chlorpyrifos petition, more HED resources could have been devoted to addressing the chlorpyrifos petition.

In accordance with 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct.

Executed this 23rd day of July, 2012.

JACK HOUSENGER

LIST OF ATTACHMENTS

A U.S.E.P.A. Chlorpyrifos Interim Reregistration Eligibility Determination.

B Evaluation of the Potential Risks From Spray Drift and the Impacts of Potential Risk Reduction Measures.

C Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held September 16-18, 2008 on the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos.

D Transmittal of Meeting Minutes of the FIFRA SAP Meeting Held December 1-3, 2009 on the Scientific Issues Associated with "Field Volatilization of Conventional Pesticides".

E Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting on the Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment.

F Chlorpyrifos: Preliminary Human Health Risk Assessment for Registration Review

G The Reader's Guide to the HHRA.

H Meeting of FIFRA Scientific Advisory Panel Draft Issue Paper: Scientific Issues Concerning Health Effects of Chlorpyrifos, April 10-13, 2012.

I Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held April 10-12, 2012 on "Chlorpyrifos Health Effects".

J Letter of July 16, 2012, from Dr. Steven Bradbury, Director, EPA Office of Pesticide Programs, to Aaron Colangelo and Margaret Reeves, Ph.D.

K Spray Drift Mitigation Decision for Chlorpyrifos, July 2012.

ATTACHMENT A

US Environmental Protection Agency
Office of Pesticide Programs

Reregistration Eligibility Decision for Chlorpyrifos

When EPA concluded the organophosphate (OP) cumulative risk assessment in July 2006, all tolerance reassessment and reregistration eligibility decisions for individual OP pesticides were considered complete. OP Interim Reregistration Eligibility Decisions (IREDs), therefore, are considered completed REDs. OP tolerance reassessment decisions (TREDs) also are considered completed.

Combined PDF document consists of the following:

- Finalization of Interim Reregistration Eligibility Decisions (IREDs) and Interim Tolerance Reassessment and Risk Management Decisions (TREDs) for the Organophosphate Pesticides, and Completion of the Tolerance Reassessment and Reregistration Eligibility Process for the Organophosphate Pesticides (July 31, 2006)
- Chlorpyrifos IRED

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON D.C., 20460

OFFICE OF
PREVENTION, PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

DATE: July 31, 2006

SUBJECT: Finalization of Interim Reregistration Eligibility Decisions (IREDs) and Interim Tolerance Reassessment and Risk Management Decisions (TREDs) for the Organophosphate Pesticides, and Completion of the Tolerance Reassessment and Reregistration Eligibility Process for the Organophosphate Pesticides

FROM: Debra Edwards, Director
Special Review and Reregistration Division
Office of Pesticide Programs

TO: Jim Jones, Director
Office of Pesticide Programs

As you know, EPA has completed its assessment of the cumulative risks from the organophosphate (OP) class of pesticides as required by the Food Quality Protection Act of 1996. In addition, the individual OPs have also been subject to review through the individual-chemical review process. The Agency's review of individual OPs has resulted in the issuance of Interim Reregistration Eligibility Decisions (IREDs) for 22 OPs, interim Tolerance Reassessment and Risk Management Decisions (TREDs) for 8 OPs, and a Reregistration Eligibility Decision (RED) for one OP, malathion.[1] These 31 OPs are listed in Appendix A.

EPA has concluded, after completing its assessment of the cumulative risks associated with exposures to all of the OPs, that:

(1) the pesticides covered by the IREDs that were pending the results of the OP cumulative assessment (listed in Attachment A) are indeed eligible for reregistration; and

[1] Malathion is included in the OP cumulative assessment. However, the Agency has issued a RED for malathion, rather than an IRED, because the decision was signed on the same day as the completion of the OP cumulative assessment.

ATTACHMENT A

(2) the pesticide tolerances covered by the IREDs and TREDs that were pending the results of the OP cumulative assessment (listed in Attachment A) meet the safety standard under Section 408(b)(2) of the FFDCA.

Thus, with regard to the OPs, EPA has fulfilled its obligations as to FFDCA tolerance reassessment and FIFRA reregistration, other than product-specific reregistration.

The Special Review and Reregistration Division will be issuing data call-in notices for confirmatory data on two OPs, methidathion and phorate, for the reasons described in detail in the OP cumulative assessment. The specific studies that will be required are:

- 28-day repeated-dose toxicity study with methidathion oxon; and
- Drinking water monitoring study for phorate, phorate sulfoxide, and phorate sulfone in both source water (at the intake) and treated water for five community water systems in Palm Beach County, Florida and two near Lake Okechobee, Florida.

The cumulative risk assessment and supporting documents are available on the Agency's website at www.epa.gov/pesticides/cumulative and in the docket (EPA-HQ-OPP-2006-0618).

ATTACHMENT A

Attachment A:
Organophosphates included in the OP Cumulative Assessment

Chemical	Decision Document	Status
Acephate	IRED	IRED completed 9/2001
Azinphos-methyl (AZM)	IRED	IRED completed 10/2001
Bensulide	IRED	IRED completed 9/2000
Cadusafos	TRED	TRED completed 9/2000
Chlorethoxyphos	TRED	TRED completed 9/2000
Chlorpyrifos	IRED	IRED completed 9/2001
Coumaphos	TRED	TRED completed 2/2000
DDVP (Dichlorvos)	IRED	IRED completed 6/2006
Diazinon	IRED	IRED completed 7/2002
Dicrotophos	IRED	IRED completed 4/2002
Dimethoate	IRED	IRED completed 6/2006
Disulfoton	IRED	IRED completed 3/2002
Ethoprop	IRED	IRED completed 9/2001 IRED addendum completed 2/2006
Fenitrothion	TRED	TRED completed 10/2000
Malathion	RED	RED completed 8/2006
Methamidophos	IRED	IRED completed 4/2002
Methidathion	IRED	IRED completed 4/2002
Methyl Parathion	IRED	IRED completed 5/2003
Naled	IRED	IRED completed 1/2002
Oxydemeton-methyl	IRED	IRED completed 8/2002
Phorate	IRED	IRED completed 3/2001
Phosalone	TRED	TRED completed 1/2001
Phosmet	IRED	IRED completed 10/2001
Phostebupirim	TRED	TRED completed 12/2000
Pirimiphos-methyl	IRED	IRED completed 6/2001
Profenofos	IRED	IRED completed 9/2000
Propetamphos	IRED	IRED completed 12/2000
Terbufos	IRED	IRED completed 9/2001
Tetrachlorvinphos	TRED	TRED completed 12/2002
Tribufos	IRED	IRED completed 12/2000
Trichlorfon	TRED	TRED completed 9/2001

United States Environmental Protection Agency | Prevention, Pesticides and Toxic Substances (7508C) | EPA 738-R-01-007 February 2002

Interim Reregistration Eligibility Decision for Chlorpyrifos

United States Environmental Protection Agency | Prevention, Pesticides and Toxic Substances (7508C) | EPA 738-F-01-006 February 2002

Chlorpyrifos Facts

EPA has assessed the risks of chlorpyrifos and reached an Interim Reregistration Eligibility Decision (IRED) for this organophosphate (OP) pesticide. Provided that risk mitigation measures are adopted, chlorpyrifos fits into its own "risk cup"-- its individual, aggregate risks are within acceptable levels. Chlorpyrifos also is eligible for reregistration, pending a full reassessment of the cumulative risk from all OPs.

Used on a variety of food and feed crops, golf courses, as a non-structural wood treatment, and as an adult mosquitocide, chlorpyrifos residues in food and drinking water do not pose risk concerns. With mitigation eliminating virtually all homeowner uses, chlorpyrifos fits into its own "risk cup." With other mitigation measures, chlorpyrifos worker and ecological risks also will be below levels of concern for reregistration.

EPA's next step under the Food Quality Protection Act (FQPA) is to complete a cumulative risk assessment and risk management decision encompassing all the OP pesticides, which share a common mechanism of toxicity. The interim decision on chlorpyrifos cannot be considered final until this cumulative assessment is complete. Further risk mitigation may be warranted at that time.

EPA is reviewing the OP pesticides to determine whether they meet current health and safety standards. Older OPs need decisions about their eligibility for reregistration under FIFRA. OPs with residues in food, drinking water, and other non-occupational exposures also must be reassessed to make sure they meet the new FQPA safety standard.

The OP Pilot Public Participation Process

The organophosphates are a group of related pesticides that affect the functioning of the nervous system. They are among EPA's highest priority for review under the Food Quality Protection Act.

EPA is encouraging the public to participate in the review of the OP pesticides. Through a six-phased pilot public participation process, the Agency is releasing for review and comment its preliminary and revised scientific risk assessments for individual OPs. (Please contact the OP Docket, telephone 703-305-5805, or see EPA's web site, www.epa.gov/pesticides/op .)

EPA is exchanging information with stakeholders and the public about the OPs, their uses, and risks through Technical Briefings, stakeholder meetings, and other fora. USDA is coordinating input from growers and other OP pesticide users.

Based on current information from interested stakeholders and the public, EPA is making interim risk management decisions for individual OP pesticides, and will make final decisions through a cumulative OP assessment.

The chlorpyrifos interim decision was made through the OP pilot public participation process, which increases transparency and maximizes stakeholder involvement in EPA's development of risk assessments and risk management decisions. EPA worked extensively with affected parties to reach

the decisions presented in this interim decision document, which concludes the OP pilot process for chlorpyrifos.

Uses

- Chlorpyrifos is an organophosphate insecticide, acaricide and miticide used to control foliage and soil-borne insect pests on a variety of food and feed crops.

- Approximately 10 million pounds are applied annually in agricultural settings. The largest agricultural market for chlorpyrifos in terms of total pounds ai is corn (~5.5 million).

Health Effects

- Chlorpyrifos can cause cholinesterase inhibition in humans; that is, it can overstimulate the nervous system causing nausea, dizziness, confusion, and at very high exposures (e.g., accidents or major spills), respiratory paralysis and death.

Risks

- Dietary exposures from eating food crops treated with chlorpyrifos are below the level of concern for the entire U.S. population, including infants and children. Drinking water risk estimates based on screening models and monitoring data from both ground and surface water for acute and chronic exposures are generally not of concern.

- In June, 2000, the Agency entered into an agreement with the technical registrants to eliminate virtually all homeowner uses, except ant and roach baits in child resistent packaging.

- Residential postapplication exposures may occur after termiticide use in residential structures. To mitigate risks from this use, the technical registrants agreed in June 2000 to limit termiticide treatments to 0.5% solution, and cancel all postconstruction uses. Pre-construction use will remain until 2005, unless acceptable exposure data are submitted that show that residential postapplication risks from this use are not a concern.

- Occupational exposure to chlorpyrifos is of concern to the Agency. Exposures of concern include mixing/loading liquids for aerial/chemigation and groundboom application, mixing wettable powder for groundboom application, aerial application, and application by backpack sprayer, high-pressure handwand, and hand-held sprayer or duster. Generally, these risks can be mitigated by a combination of additional personal protective equipment and engineering controls, and by reductions in application rates. Additionally, the Agricultural Handler Task Force will be developing exposure data to better characterize the risk from certain uses (e.g., applying granulars by air).

- Risk quotients indicate that a single application of chlorpyrifos poses risks to small mammals, birds, fish and aquatic invertebrate species for nearly all registered outdoor uses. Multiple applications increase the risks to wildlife and prolong exposures to toxic concentrations. To address these risks, a number of measures including reduced application rates, increased retreatment intervals, reduced seasonal maximum amounts applied per acre, and no-spray setback zones around water bodies will be needed.

Risk Mitigation

In order to support a reregistration eligibility decision for chlorpyrifos, the following risk mitigation measures are necessary:

- To mitigate risks to agricultural workers PPE consisting of double layers, chemical resistant gloves, chemical resistant shoes plus socks, chemical resistant headgear for overhead exposure, chemical resistant apron when cleaning and mixing or loading and a dust/mist respirator are required for the following scenarios: mixing/loading liquids for groundboom and airblast application, loading granulars for ground application, tractor drawn granular spreader, and low pressure handwand.

- engineering controls are required for the following scenarions: mixing wettable powder for groundboom application (water soluble packaging), mixing wettable powder for airblast application (water soluble packaging), and aerial application of sprays (enclosed cockpit).

- There are still some occupational risk scenarios that are still below the target MOE of 100, even with all feasible PPE or engineering controls. The risk assessments for these uses will be refined with additional data.

- To mitigate ecological risks the technical registrants have agreed to label amendments which include the use of buffer zones to protect water quality, fish and wildlife, reductions in application rates, number of applications per season, seasonal maximum amounts applied, and increases in the minimum intervals for retreatment.

- The mitigation measures prescribed in the IRED along with mitigation that is already being implemented as a result of the June, 2000, Memorandum of Agreement, will reduce risk to both terrestrial and aquatic species. For example, many of the reported incidents of wildlife mortality associated with chlorpyrifos use were related to residential lawn and termite uses and use on golf courses. The residential uses have been eliminated, the termiticide use is being phased out, and the application rate on golf courses has been reduced from 4 to 1 lb/ai/A. Additionally, no-spray buffers around surface water bodies, as well as rate reductions for agricultural uses will be implemented as a result of this IRED and will further reduce the environmental burden of chlorpyrifos.

Next Steps

- Numerous opportunities for public comment were offered as this decision was being developed. In addition, the chlorpyrifos IRED has been issued with a public comment period (see www.epa.gov/REDs/ or www.epa.gov/pesticides/op).

- When the cumulative risk assessment for all organophosphate pesticides is completed, EPA will issue its final tolerance reassessment decision for chlorpyrifos and may request further risk mitigation measures. The Agency will revoke the tomato tolerance and amend the grape and apple tolerances for chlorpyrifos. For all OPs, raising and/or establishing tolerances will be considered once a cumulative assessment is completed.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

CERTIFIED MAIL

Dear Registrant:

This is to inform you that the Environmental Protection Agency (hereafter referred to as EPA or the Agency) has completed its review of the available data and public comments received related to the preliminary and revised risk assessments for the organophosphate pesticide chlorpyrifos. The public comment period on the revised risk assessment phase of the reregistration process is closed. Based on comments received during the public comment period and additional data received from the technical registrants, the Agency revised the human health and environmental effects risk assessments and made them available to the public on August 16, 2000. Additionally, the Agency held a Technical Briefing on June 8, 2000, where the results of the revised human health and environmental effects risk assessments and interim mitigation measures were presented to the general public. This Technical Briefing concluded Phase 4 of the OP Public Participation Pilot Process developed by the Tolerance Reassessment Advisory Committee, and initiated Phase 5 of that process. During Phase 5, all interested parties were invited to participate and provide comments and suggestions on ways the Agency might mitigate the estimated risks presented in the revised risk assessments. This public participation and comment period commenced on August 16, 2000, and closed on October 16, 2000.

Based on its review, EPA has identified risk mitigation measures that the Agency believes are necessary to address the human health and environmental risks associated with the current use of chlorpyrifos. The EPA is now publishing its interim decision on the reregistration eligibility of and risk management decision for the current uses of chlorpyrifos and its associated human health and environmental risks. The reregistration eligibility and tolerance reassessment decisions for chlorpyrifos will be finalized once the cumulative risks for all of the organophosphate pesticides are considered. The enclosed "Interim Reregistration Eligibility Decision for Chlorpyrifos," which was approved on September 28, 2001, contains the Agency's decision on the individual chemical chlorpyrifos.

A Notice of Availability for this Interim Reregistration Eligibility Decision (IRED) for chlorpyrifos was being published in the *Federal Register*. To obtain a copy of the interim RED document, please contact the OPP Public Regulatory Docket (7502C), US EPA, Ariel Rios Building, 1200 Pennsylvania Avenue NW, Washington, DC 20460, telephone (703) 305-5805. Electronic copies of the interim RED and all supporting documents are available on the Internet. See http:www.epa.gov/pesticides/op.

This IRED for chlorpyrifos has been revised based on comments received during the public comment period following the announcement of the availability of the chlorpyrifos IRED in the *Federal Register* (66 FR 57073). This revised IRED incorporates many of the comments that were received, other comments will be addressed under separate cover.

The interim RED is based on the updated technical information found in the chlorpyrifos public docket. The docket not only includes background information and comments on the Agency's preliminary risk assessments, it also includes the Agency's revised risk assessments for chlorpyrifos (revised as of June 8, 2000), and a document summarizing the Agency's Response to Comments. The Response to Comments document addresses corrections to the preliminary risk assessments submitted by chemical registrants, as well as responds to comments submitted by the general public and stakeholders during the comment period on the risk assessment. The docket will also include comments on the revised risk assessment, and any risk mitigation proposals submitted during Phase 5. During Phase 5, EPA and the technical registrants of chlorpyrifos entered into an agreement to implement interim risk mitigation.

This document and the process used to develop it are the result of a pilot process to facilitate greater public involvement and participation in the reregistration and/or tolerance reassessment decisions for these pesticides. As part of the Agency's effort to involve the public in the implementation of the Food Quality Protection Act of 1996 (FQPA), the Agency is undertaking a special effort to maintain open public dockets on the organophosphate pesticides and to engage the public in the reregistration and tolerance reassessment processes for these chemicals. This open process follows the guidance developed by the Tolerance Reassessment Advisory Committee (TRAC), a large multi-stakeholder advisory body that advised the Agency on implementing the new provisions of the FQPA. The reregistration and tolerance reassessment reviews for the organophosphate pesticides are following this new process.

Please note that the chlorpyrifos risk assessments and the attached interim RED concern only this particular organophosphate. This interim RED presents the Agency's conclusions on the dietary risks posed by exposure to chlorpyrifos alone. The Agency has also concluded its assessment of the ecological and worker risks associated with the use of chlorpyrifos. Because the FQPA directs the Agency to consider available information on the basis of cumulative risk from substances sharing a common mechanism of toxicity, such as the toxicity expressed by the organophosphates through a common biochemical interaction with the cholinesterase enzyme, the Agency will evaluate the cumulative risk posed by the entire organophosphate class of chemicals after considering the risks for the individual organophosphates. The Agency is working towards completion of a methodology to assess cumulative risk and the individual risk assessments for each organophosphate are likely to be necessary elements of any cumulative assessment. The Agency has decided to move forward with individual assessments and to identify mitigation measures necessary to address those human health and environmental risks associated with the current uses of chlorpyrifos. The Agency will issue the final tolerance reassessment decision for chlorpyrifos and finalize decisions on reregistration eligibility once the cumulative risks for all of the organophophates are considered.

This document contains generic and product-specific Data Call-Ins (DCIs) that outline further data requirements for this chemical. Note that a complete DCI, with all pertinent

instructions, is being sent to registrants under separate cover. Additionally, for product-specific DCIs, the first set of required responses is due 90 days from the receipt of the DCI letter. The second set of required responses is due eight months from the date of the DCI.

In this interim RED, the Agency has determined that, with the exception of open-pour dust formulations for fire ant control, chlorpyrifos products will be eligible for reregistration provided that all the conditions identified in this document are satisfied, including implementation of the risk mitigation measures outlined in Section IV of the document. The Agency believes that current uses of chlorpyrifos may pose unreasonable adverse effects to human health and the environment, and that such effects can be mitigated with the risk mitigation measures identified in this interim RED. Accordingly, the Agency recommends that registrants implement these risk mitigation measures immediately. Sections IV and V of this interim RED describe labeling amendments for end-use products and data requirements necessary to implement these mitigation measures. Instructions for registrants on submitting the revised labeling can be found in the set of instructions for product-specific data that accompanies this interim RED.

Should a registrant choose not to implement any of the risk mitigation measures outlined in this document, the Agency will continue to have concerns about the risks posed by chlorpyrifos. Where the Agency has identified any unreasonable adverse effect to human health or the environment, the Agency intends to initiate appropriate regulatory action to address this concern. At that time, any affected person(s) may challenge the Agency's action.

If you have questions on this document, the label changes necessary for reregistration, or the generic DCI, please contact the Chemical Review Manager, Tom Myers, at (703) 308-8589. For questions about product reregistration and/or the Product DCI that accompanies this document, please contact Venus Eagle at (703) 308-8045.

Sincerely,

Lois A. Rossi, Director
Special Review and Reregistration Division

Attachment

Interim Reregistration Eligibility Decision
for
Chlorpyrifos

Case No. (0100)

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GLOSSARY OF TERMS AND ABBREVIATIONS

AE	Acid Equivalent
a.i.	Active Ingredient
AGDCI	Agricultural Data Call-In
ai	Active Ingredient
aPAD	Acute Population Adjusted Dose
AR	Anticipated Residue
ARC	Anticipated Residue Contribution
BCF	Bioconcentration Factor
CAS	Chemical Abstracts Service
CI	Cation
CNS	Central Nervous System
cPAD	Chronic Population Adjusted Dose
CSF	Confidential Statement of Formula
CFR	Code of Federal Regulations
CSFII	USDA Continuing Surveys for Food Intake by Individuals
DCI	Data Call-In
DEEM	Dietary Exposure Evaluation Model
DFR	Dislodgeable Foliar Residue
DRES	Dietary Risk Evaluation System
DWEL	Drinking Water Equivalent Level (DWEL) The DWEL represents a medium specific (i.e., drinking water) lifetime exposure at which adverse, noncarcinogenic health effects are not anticipated to occur.
DWLOC	Drinking Water Level of Comparison.
EC	Emulsifiable Concentrate Formulation
EEC	Estimated Environmental Concentration. The estimated pesticide concentration in an environment, such as a terrestrial ecosystem.
EP	End-Use Product
EPA	U.S. Environmental Protection Agency
FAO	Food and Agriculture Organization
FDA	Food and Drug Administration
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
FFDCA	Federal Food, Drug, and Cosmetic Act
FQPA	Food Quality Protection Act
FOB	Functional Observation Battery
G	Granular Formulation
GENEEC	Tier I Surface Water Computer Model
GLC	Gas Liquid Chromatography
GLN	Guideline Number
GM	Geometric Mean
GRAS	Generally Recognized as Safe as Designated by FDA

HA	Health Advisory (HA). The HA values are used as informal guidance to municipalities and other organizations when emergency spills or contamination situations occur.
HAFT	Highest Average Field Trial
HDT	Highest Dose Tested
IR	Index Reservoir
LC_{50}	Median Lethal Concentration. A statistically derived concentration of a substance that can be expected to cause death in 50% of test animals. It is usually expressed as the weight of substance per weight or volume of water, air or feed, e.g., mg/l, mg/kg or ppm.
LD_{50}	Median Lethal Dose. A statistically derived single dose that can be expected to cause death in 50% of the test animals when administered by the route indicated (oral, dermal, inhalation). It is expressed as a weight of substance per unit weight of animal, e.g., mg/kg.
LEL	Lowest Effect Level
LOC	Level of Concern
LOD	Limit of Detection
LOAEL	Lowest Observed Adverse Effect Level
MATC	Maximum Acceptable Toxicant Concentration
MCLG	Maximum Contaminant Level Goal (MCLG) The MCLG is used by the Agency to regulate contaminants in drinking water under the Safe Drinking Water Act.
mg/kg/day	Milligram Per Kilogram Per Day
mg/L	Milligrams Per Liter
MOE	Margin of Exposure
MP	Manufacturing-Use Product
MPI	Maximum Permissible Intake
MRID	Master Record Identification (number). EPA's system of recording and tracking studies submitted.
NA or N/A	Not Applicable
NAWQA	USGS National Water Quality Assessment
NOEC	No Observable Effect Concentration
NOEL	No Observed Effect Level
NOAEL	No Observed Adverse Effect Level
NPDES	National Pollutant Discharge Elimination System
OP	Organophosphate
OPP	EPA Office of Pesticide Programs
OPPTS	EPA Office of Prevention, Pesticides and Toxic Substances
Pa	pascal, the pressure exerted by a force of one newton acting on an area of one square meter.
PAD	Population Adjusted Dose
PADI	Provisional Acceptable Daily Intake
PAG	Pesticide Assessment Guideline
PAM	Pesticide Analytical Method
PCA	Percent Crop Area

PDP	USDA Pesticide Data Program
PHED	Pesticide Handler's Exposure Data
PHI	Preharvest Interval
ppb	Parts Per Billion
PPE	Personal Protective Equipment
ppm	Parts Per Million
PRN	Pesticide Registration Notice
PRZM/ EXAMS	Tier II Surface Water Computer Model
Q_1*	The Carcinogenic Potential of a Compound, Quantified by the EPA's Cancer Risk Model
RAC	Raw Agriculture Commodity
RBC	Red Blood Cell
RED	Reregistration Eligibility Decision
REI	Restricted Entry Interval
RfD	Reference Dose
RQ	Risk Quotient
RS	Registration Standard
RUP	Restricted Use Pesticide
SAP	Science Advisory Panel
SCI-GROW	Tier I Ground Water Computer Model
SF	Safety Factor
SLC	Single Layer Clothing
SLN	Special Local Need (Registrations Under Section 24(c) of FIFRA)
TC	Toxic Concentration. The concentration at which a substance produces a toxic effect.
TD	Toxic Dose. The dose at which a substance produces a toxic effect.
TEP	Typical End-Use Product
TGAI	Technical Grade Active Ingredient
TLC	Thin Layer Chromatography
TMRC	Theoretical Maximum Residue Contribution
torr	A unit of pressure needed to support a column of mercury 1 mm high under standard conditions.
TRR	Total Radioactive Residue
UF	Uncertainty Factor
μg/g	Micrograms Per Gram
μg/L	Micrograms Per Liter
USDA	United States Department of Agriculture
USGS	United States Geological Survey
WHO	World Health Organization
WP	Wettable Powder
WPS	Worker Protection Standard

Executive Summary

EPA has completed its review of public comments on the revised risk assessments and is issuing its interim reregistration eligibility decision for chlorpyrifos. The decisions outlined in this document do not include the final tolerance reassessment decision for chlorpyrifos; however, some tolerance actions will be undertaken prior to completion of the final tolerance reassessment. EPA intends to revoke the tolerance for tomatoes, because that use is being canceled, and to reduce the tolerances for grapes and apples. The final tolerance reassessment and reregistration eligibility decision for this chemical will be issued once the cumulative risks for all of the organophosphates are considered. The Agency may need to pursue further risk management measures for chlorpyrifos once cumulative risks are considered.

The revised risk assessments are based on review of the required target data base supporting the use patterns of currently registered products and new information received. The Agency invited stakeholders to provide proposals, ideas or suggestions on appropriate mitigation measures before the Agency issued its risk mitigation decision on chlorpyrifos. After considering the revised risks taking into account the interim mitigation as well as additional mitigation proposed by Dow AgroSciences (DAS), one of the technical registrants of chlorpyrifos, and comments and mitigation suggestions from other interested parties, EPA developed its risk management decision for remaining uses of chlorpyrifos that pose risks of concern. This decision is discussed fully in this document.

Chlorpyrifos is an organophosphate insecticide, acaricide and miticide used to control a variety of insects, first registered in 1965 for control of foliage and soil-borne insect pests on a variety of food and feed crops. Technical registrants include Dow AgroSciences, Cheminova, Inc., Gharda USA, Inc., Luxembourg-Pamol, Inc., Makhteshim-Agan of North America, Inc. and Platte Chemical Company, Inc. Chlorpyrifos is one of the most widely used organophosphate insecticides in the U.S. and, until 2000 when nearly all residential uses were cancelled, was one of the major insecticides used in residential settings. Currently registered uses include food and feed crops, golf course turf, greenhouses, non-structural wood treatments such as utility poles and fence posts, and as an adult mosquitocide. Structural treatments for termites are also currently registered, but are being phased out. All use of products for structural termite control will be prohibited after December 31, 2005, unless acceptable data demonstrate that risks from these exposures are not of concern. Indoor non-residential uses include shipholds, railroad boxcars, industrial plants and manufacturing plants.

Based on data reflecting usage for the years 1987 through 1998, the Agency estimates that the annual total domestic usage of chlorpyrifos was approximately 21 to 24 million pounds active ingredient (ai) for 8 million acres treated in the U.S. Approximately 11 million pounds were applied annually in non-agricultural settings (i.e., residences, schools, golf courses, parks) prior to the implementation of interim mitigation in 2000. The largest agricultural market for chlorpyrifos in terms of total pounds ai is corn (~5.5 million). The largest non-agricultural markets in terms of total pounds ai applied were pest control operators (PCOs) for termite control (5 million), and turf (2.5 million). Crops with a high average percentage of their total

U.S. planted acres treated include Brussels sprouts (73%), cranberries (46%), apples (44%), broccoli (41%) and cauliflower (31%).

In June, 2000, the Agency released its revised human health risk assessment and entered into an agreement with the technical registrants to eliminate and phase out certain uses of chlorpyrifos. The agreement was established at that time in order to expeditiously address food, drinking water, residential and non-residential uses posing the greatest risks to children. The mitigation contained in the agreement also reduced some occupational and ecological exposures by eliminating use sites and reducing application rates. Details of the interim risk mitigation can be found on the internet at http://www.epa.gov/pesticides/op/.

The technical registrants have since agreed to additional mitigation measures addressing occupational and ecological risks not addressed in the June, 2000 agreement. These measures are the result of discussion between the Agency and the technical registrants during Phase 5 of the public participation process, and are in the process of being implemented.

Overall Risk Summary

EPA's preliminary human health risk assessment for chlorpyrifos indicated dietary (food and drinking water), occupational and residential risk concerns. The revised risk assessment indicates that, with implementation of the June 2000 mitigation agreement, dietary risks from food are not of concern. Drinking water risk estimates based on screening models and monitoring data from both ground and surface water for acute and chronic exposures are generally not of concern. The exception is incidents of contamination resulting from termiticide use, which are highly localized and expected to be declining because the termiticide use is being phased out. There are concerns for some workers who mix, load, and apply chlorpyrifos to agricultural and other non-residential sites.

Application of chlorpyrifos poses acute and reproductive risks to many non-target aquatic and terrestrial animals for all outdoor uses reviewed. The risk quotients for all chlorpyrifos uses exceed the levels of concern for most terrestrial and aquatic categories. In general, risk quotients are greater among estuarine species than freshwater species. Terrestrial animals are at less risk than aquatic species. Birds appear to be more at risk than most mammalian species. Aquatic risk quotients for ground spray applications are less than aerial spray applications at the same application rate.

Results of the risk assessments, and the label amendments that EPA believes will mitigate risks to acceptable levels taking into account the benefits of chlorpyrifos use, are presented in this interim RED.

Dietary Risk

The preliminary risk assessment showed that acute dietary risks from food exceeded the acute population adjusted dose (aPAD) for infants, all children, and nursing females of child-bearing age (13-50 years old). To address these risks, the technical registrants agreed to eliminate use on tomatoes and restrict use on apples. EPA will revoke the tomato tolerance and lower the apple tolerance to ensure that both domestic and imported commodities do not contain residues of concern. Use on apples is restricted to dormant (pre-bloom) applications; the tolerance will be lowered to reflect this. In addition, the tolerance on grapes will be lowered to reflect the currently registered use. The proposed tolerance actions be announced in the *Federal Register* and will have a public comment period separate from the comment period for this IRED. With this mitigation, acute risks from food are not a concern for any population subgroup.

Acute and chronic exposures to drinking water do not exceed the DWLOCs and are therefore not of concern. Drinking water risk estimates based on screening models and monitoring data from both ground and surface water for acute and chronic exposures are generally not of concern. The exception is incidents of contamination resulting from termiticide use, which are highly localized and expected to be declining with the phasing out of the termiticide use and implementation of generic risk mitigation for termiticides (reduction of the concentration during the phase-out period.)

Chronic dietary risk from food and drinking water does not exceed the Agency's level of concern for the general U.S. population or for any population subgroup.

Occupational Risk

Occupational exposure to chlorpyrifos is of concern to the Agency. Exposures of concern include mixing/loading liquids for aerial/chemigation and groundboom application, mixing wettable powder for groundboom application, aerial application, and application by backpack sprayer, high-pressure handwand, bulbous duster and hand-held sprayer. Generally, these risks can be mitigated by a combination of additional personal protective equipment and engineering controls, and by reductions in application rates. Additionally, the Agricultural Handler Task Force will be developing exposure data to better characterize the risk from certain uses (e.g., applying granulars by air).

Postapplication risks can be mitigated by reducing application rates for a number of uses and in some cases by the establishment of new restricted entry intervals, i.e., the amount of time that must elapse before risks are not of concern to workers re-entering treated fields.

Residential Risk

Risks to residents, particularly children, from chlorpyrifos use in the home, as well as residential postapplication risks following residential treatments are a concern. To mitigate these risks, the technical registrants agreed in June 2000 to cancel almost all indoor and outdoor residential uses. Virtually all products labeled for homeowner use have been canceled effective December 31, 2001, except containerized ant and roach baits in child-resistant packaging which have not been canceled because they present minimal exposure. Distribution and sale of products for all other residential uses will be prohibited after December 31, 2001. The application rate for termite treatments was reduced as of December 1, 2000. Full-barrier (whole-house) termite treatment products may no longer be distributed or sold after December 31, 2001. Spot and local post-construction use will be canceled on December 31, 2002, and pre-construction termiticide uses will be canceled on December 31, 2005, unless acceptable exposure data are submitted and demonstrate that postapplication risks to residents are not of concern.

Non-Agricultural Non-Residential Risk

Risks to children in schools and parks, both indoors and outdoors, are of concern to the Agency. Therefore, per the mitigation agreement signed in June 2000, distribution and sale of products bearing these uses will be prohibited effective December 31, 2001. The only non-agricultural non-residential uses that will be reregistered are golf course turf, shipholds, railroad boxcars, industrial plants, manufacturing plants, and processed wood products, none of which are expected to result in risks to children. Exposure data are required to confirm that exposure to residents from chlorpyrifos-treated wood products is not of concern.

Aggregate Risk

Acute, short-term and chronic aggregate assessments were conducted. Taking into account residential risk mitigation, aggregate risks are not a concern for any of these scenarios.

Ecological Risk

Risk quotients indicate that a single application of chlorpyrifos poses risks to small mammals, birds, fish and aquatic invertebrate species for nearly all registered outdoor uses. Multiple applications increase the risks to wildlife and prolong exposures to toxic concentrations. In most cases, acute risk quotients exceed 1 for the most sensitive, small mammals and birds. All aquatic acute and reproductive risk quotients exceed 1; many aquatic risk quotients exceed 10 and 100, and both acute and reproductive risk quotients for estuarine invertebrates exceed 1,000 on some crops. In a few cases at maximum application rates, chlorpyrifos may bioconcentrate in the tissues of fish and aquatic invertebrates to levels that exceed acute LC_{50} values for sensitive bird species and reproductive NOAELs for birds and small mammalian species. Hence without mitigation to reduce levels in shallow waters,

bioconcentration of chlorpyrifos in ponds and estuarine areas may pose acute and/or reproductive risks to aquatic birds and mammals feeding adjacent to treated areas.

To address these risks, a number of measures including reduced application rates, increased retreatment intervals, reduced seasonal maximum amounts applied per acre, and no-spray setback zones around water bodies will be needed.

Interim Reregistration Eligibility Decision

With the addition of the label restrictions and amendments detailed in this document, the Agency has determined that all currently registered uses of chlorpyrifos, except open-pour dust formulations, may continue until the cumulative risks for all of the organophosphates have been considered.

The Agency is issuing this interim Reregistration Eligibility Decision (RED) for chlorpyrifos, as announced in a Notice of Availability published in the *Federal Register*. This interim RED document includes guidance and time frames for making label changes for products containing chlorpyrifos. There will be a 60-day public comment period for this interim RED. Phase 6 of the pilot process did not include a public comment period; however, for some chemicals, the Agency may provide for another comment period, depending on the content of the risk management decision. Neither the tolerance reassessment nor the reregistration eligibility decision for chlorpyrifos can be considered final, however, until the cumulative risks for all organophosphate pesticides are considered. The cumulative assessment may result in further risk mitigation measures for chlorpyrifos.

I. Introduction

The Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) was amended in 1988 to accelerate the reregistration of products with active ingredients registered prior to November 1, 1984. The amended Act calls for the development and submission of data to support the reregistration of an active ingredient, as well as a review of all submitted data by the U.S. Environmental Protection Agency (referred to as EPA or "the Agency"). Reregistration involves a thorough review of the scientific database underlying a pesticide's registration. The purpose of the Agency's review is to reassess the potential hazards arising from the currently registered uses of the pesticide; to determine the need for additional data on health and environmental effects; and to determine whether the pesticide meets the "no unreasonable adverse effects" criteria of FIFRA.

On August 3, 1996, the Food Quality Protection Act of 1996 (FQPA) was signed into law. This Act amends FIFRA to require tolerance reassessment of all existing tolerances. The Agency had decided that, for those chemicals that have tolerances and are undergoing reregistration, the tolerance reassessment will be initiated through this reregistration process. It also requires that by 2006, EPA must review all tolerances in effect on the day before the date of the enactment of the FQPA. FQPA also amends the FFDCA to require a safety finding in tolerance reassessment based on factors including an assessment of cumulative effects of chemicals with a common mechanism of toxicity. Chlorpyrifos belongs to a group of pesticides called organophosphates, which share a common mechanism of toxicity--they all affect the nervous system by inhibiting cholinesterase. Although FQPA significantly affects the Agency's reregistration process, it does not amend any of the existing reregistration deadlines. Therefore, the Agency is continuing its reregistration program while it resolves the remaining issues associated with the implementation of FQPA.

This document presents the Agency's revised human health and ecological risk assessments; its progress toward tolerance reassessment; and the interim decision on the reregistration eligibility of chlorpyrifos. It is intended to be only the first phase in the reregistration process for chlorpyrifos. The Agency will eventually proceed with its assessment of the cumulative risk of the OP pesticides and issue a final reregistration eligibility decision for chlorpyrifos.

The implementation of FQPA has required the Agency to revisit some of its existing policies relating to the determination and regulation of dietary risk, and has also raised a number of new issues for which policies need to be created. These issues were refined and developed through collaboration between the Agency and the Tolerance Reassessment Advisory Committee (TRAC), which was composed of representatives from industry, environmental groups, and other interested parties. The TRAC identified the following science policy issues it believed were key to the implementation of FQPA and tolerance reassessment:

- Applying the FQPA 10-Fold Safety Factor
- Whether and How to Use "Monte Carlo" Analyses in Dietary Exposure Assessments
- How to Interpret "No Detectable Residues" in Dietary Exposure Assessments
- Refining Dietary (Food) Exposure Estimates
- Refining Dietary (Drinking Water) Exposure Estimates
- Assessing Residential Exposure
- Aggregating Exposure from all Non-Occupational Sources
- How to Conduct a Cumulative Risk Assessment for Organophosphate or Other Pesticides with a Common Mechanism of Toxicity
- Selection of Appropriate Toxicity Endpoints for Risk Assessments of Organophosphates
- Whether and How to Use Data Derived from Human Studies

The process developed by the TRAC calls for EPA to provide one or more documents for public comment on each of the policy issues described above. Each of these issues is evolving and in a different stage of refinement. Some issue papers have already been published for comment in the *Federal Register* and others will be published shortly.

In addition to the policy issues that resulted from the TRAC process, the Agency issued, on September 29, 2000, a Pesticide Registration Notice (PR 2000-9, Worker Risk Mitigation for Organophosphate Pesticides, hereafter referred to as the Worker PR Notice) that presents EPA's approach for managing risks from organophosphate pesticides to occupational users. The Worker PR Notice describes the Agency's baseline approach to managing risks to handlers and workers who may be exposed to organophosphate pesticides, and the Agency expects that other types of chemicals will be handled similarly. Generally, basic protective measures such as closed mixing and loading systems, enclosed cab equipment, or protective clothing, as well as increased reentry intervals will be necessary for most uses where current risk assessments indicate a risk and such protective measures are feasible. The policy also states that the Agency will assess each pesticide individually, and based upon the risk assessment, determine the need for specific measures tailored to the potential risks of the chemical. The measures included in this interim RED are consistent with the Worker PR Notice.

This document consists of six sections. Section I contains the regulatory framework for reregistration/tolerance reassessment as well as descriptions of the process developed by TRAC for public comment on science policy issues for the organophosphate pesticides and the Worker PR notice. Section II provides a profile of the use and usage of the chemical. Section III gives an overview of the revised human health and environmental effects risk assessments resulting from public comments and other information. Section IV presents the Agency's interim decision on reregistration eligibility and risk management decisions. Section V summarizes the label changes necessary to implement the risk mitigation measures outlined in Section IV. Section VI provides information on how to access related documents. Finally, the Appendices include Data Call-In (DCI) information. The revised risk assessments and related addenda are not included in this document, but are available on the Agency's web page www.epa.gov/pesticides/op, and in the public docket.

II. Chemical Overview

A. Regulatory History

Chlorpyrifos, [0,0-diethyl 0-(3,5,6-trichloro-2-pyridinyl)-phosphorothioate], is a broad-spectrum, chlorinated organophosphate insecticide, acaricide and nematicide that was first registered in 1965 to control foliage- and soil-borne insect pests on a variety of food and feed crops. Chlorpyrifos' most common trade names are Dursban®, Empire 20®, Equity®, and Whitmire PT 270®. Lorsban® is a trade name for agricultural-use products. It is one of the most widely used organophosphate insecticides in the U.S., and until recently was one of the major insecticides used in residential settings. During the years 1987 to 1998, approximately 21 to 24 million pounds were used annually in the U.S., of which approximately 11 million pounds were applied in non-agricultural settings. At one time there were over 400 registered products containing chlorpyrifos on the market. Registered uses included: a variety of food crops (i.e., there are approximately 112 tolerances for food/feed commodities); golf course turf; non-residential sites such as industrial plants and vehicles; non-structural wood treatments such as utility poles, fence posts, and processed wood products; and public health uses (to control mosquitoes and fire ants) and impregnated in ear tags for cattle. Chlorpyrifos is also registered for structural pest control for termites; however, this use is being phased out and will be prohibited effective December 31, 2005, unless acceptable data demonstrate that exposures from this use are not of concern.

In January, 1997, the technical registrants entered into an agreement with the Agency to reduce indoor exposures to chlorpyrifos, especially to children and other sensitive groups. Indoor broadcast treatments, indoor total release aerosols/foggers, direct application to pets via shampoos, dips and sprays, and paint additives were eliminated.

In June 2000, the technical registrants entered into an agreement with the Agency to eliminate and phase out nearly all uses that result in residential exposures. The only exceptions are containerized baits and public health uses such as mosquito and fire ant control, which do not pose risks of concern and provide important public health benefits. The agreement phased in the various restrictions and cancellations to address higher risk uses of chlorpyrifos first. Because much of the risk reduction involves increasing margins of safety, the agreement focused first on mitigation that achieved the greatest risk reduction for children. Allowing uses with lower risks to continue for a specific period of time will help ensure that appropriate alternatives are available for a reasonable and orderly transition. The provisions of the agreement are summarized in Table 1 below. This document does not present the risks for those uses that will be phased out and/or have been canceled. Discussion of the risks associated with these uses can be found in the *Human Health Risk Assessment*, June 8, 2000, which is located in the public docket and on the internet at www.epa/gov/pesticides/op.

Table 1. Provisions of the June 2000 Memorandum of Agreement

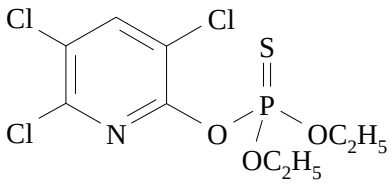
Food Uses		
Crop	**Mitigation Measures**	**Effective Dates**
Apples	Production of chlorpyrifos products labeled for post-bloom application is prohibited (only production for pre-bloom, dormant application is allowed) Post-bloom use is prohibited	August - September 2000 Stop use (use prohibited) as of 12-31-00
Tomatoes	Production of products for tomato use is prohibited	August - September 2000 Stop use as of 12-31-00
All Agricultural Uses	Classify new end-use products for restricted use or package in large containers New end-use products must bear revised Restricted Entry Intervals (REIs)	As of 12-1-00 As of 12-1-00

Home Uses		
Home lawn and most other outdoor uses	Classify new end-use products for restricted use or package in large containers (except baits in child resistant packaging) Use will be canceled	As of 12-1-00 Stop formulation 12-1-00 Formulators stop sale 2-1-01 Retailers stop sale 12-31-01
Crack and crevice and most other indoor uses	Classify new end-use products for restricted use or package in large containers Use will be canceled	As of 12-1-00 Stop formulation 12-1-00 Formulators stop sale 2-1-01 Retailers stop sale 12-31-01

Home Uses		
Termiticides	Classify new products for restricted use or package in large containers	As of 12-1-00
	Limit use to 0.5% solution	In label directions as of 12-1-00
❑ Full barrier (whole house) post-construction use	Use will be canceled	Stop formulation 12-1-00 Formulators stop sale 2-1-01 Retailers stop sale 12-31-01
❑ Spot and local post-construction use	Use will be canceled	Stop formulation 12-1-00 unless label has stop use date of 12-31-02
❑ Pre-construction use	Use will be canceled unless acceptable exposure data show that risks are not of concern	Stop production 12-31-04 Stop use 12-31-05
Non-Residential Uses		
Indoor areas where children could be exposed (such as schools)	Uses will be canceled	Stop formulation 12-1-00 Formulators stop sale 2-1-01 Retailers stop sale 12-31-01
Outdoor areas where children could be exposed (such as parks)	Uses will be canceled	Stop formulation 12-1-00 Formulators stop sale 2-1-01 Retailers stop sale 12-31-01
Non-Agricultural Uses that Will Remain		
Residential use of containerized baits	In child resistant packaging	(Use allowed to continue)
Indoor areas where children will not be exposed, including only ship holds, railroad boxcars, industrial plants, manufacturing plants, or food processing plants		New end-use product labels must reflect only these uses as of 12-1-00

Non-Agricultural Uses that Will Remain		
Outdoor areas where children will not be exposed, including only:		New end-use product labels must reflect only these uses as of 12-1-00
❑ Golf course turf	Reduce application rate from 4 lbs/acre to 1 lb/acre	
❑ Road medians	Reduce maximum application rate to 1 lb ai/acre	
❑ Industrial plant sites	Reduce maximum application rate to 1 lb ai/acre	
❑ Non-structural wood treatments including fenceposts, utility poles, railroad ties, landscape timbers, logs, pallets, wooden containers, poles, posts, and processed wood products	(Continue at current rate)	
Public health uses:		
❑ Fire ant mounds (drench and granular treatment)	For professional use only	
❑ Mosquito control	For professional use only	

B. Chemical Identification



- **Common name:** Chlorpyrifos
- **Chemical name:** [0,0-diethyl 0-(3,5,6-trichloro-2-pyridinyl-phosphorothioate]
- **Chemical family:** Organophosphate
- **Case number:** 0100
- **CAS registry number:** 2921-88-2
- **OPP chemical code:** 059101
- **Empirical formula:** $C_9H_{11}Cl_3NO_3PS$

- **Molecular weight:** 350.6
- **Trade and other names:** Dursban®, Lorsban®, Empire 20®, Equity®, Whitmire PT270®
- **Basic manufacturer:** Dow AgroSciences

Technical chlorpyrifos is a white crystalline solid with a melting point of 41.5-42.5°C. Chlorpyrifos is stable in neutral and acidic aqueous solutions; however, stability decreases with increasing pH. Chlorpyrifos is practically insoluble in water, but is soluble in most organic solvents (i.e. acetone, xylene and methylene chloride). Chlorpyrifos is not particularly volatile based on its low vapor pressure of 1.87×10^{-5}mm Hg at 20°C (Merck Index, 11th Edition). Its maximum attainable vapor concentration is 25 ppb at 25°C.

C. Use Profile

The following information is based on the currently registered uses of chlorpyrifos.

- **Type of Pesticide:** Insecticide, acaricide and nematicide
- **Summary of Use Sites:**

  Food/Feed: Registered for use on the following crops/sites: cranberries, strawberries, citrus, apples, figs, pears, nectarines, cherries, peaches, plums, grapes, almonds, pecans, walnuts, nut trees, onions, peppers, kale, broccoli, Brussels sprouts, cabbage, cauliflower, collards, cucurbits, asparagus, roots/tubers, corn, lentils, beans, peas, sorghum, tobacco, wheat, alfalfa, peanuts, soybeans, sunflower, cotton, sugar beets, mint, bananas, pasture

  Other agricultural sites: Cattle ear tags, Christmas trees, woodland

  Residential: Structural treament for termites, containerized baits

  Public Health: Fire ant mounds, mosquito adulticides

  Other Nonfood: Golf courses, shipholds, boxcars, industrial plants, processed wood products

- **Target Pests:** A wide variety of insects and related organisms, and root-knot nematodes

- **Formulation Types Registered**: Formulated as a liquid emulsifiable concentrate, granular, wettable powder, dry flowable, pressurized liquid, dust, ready-to-use solution, microencapsulated material, pellets/tablets, soluble concentrate and impregnated materials (eartags).

- **Method and Rates of Application**:

  Equipment: Applied by aerial, chemigation, groundboom, tractor-drawn granular spreader, airblast sprayer, low and high pressure hand wands, hydraulic hand-held sprayer, shaker can, belly grinder, push-type spreader, large tank sprayer, compressed air sprayer, hose-end sprayer, aerosol sprayer, hand, and eartags.

  Method: Foliar, bark, seed and soil-incorporated band or broadcast treatments

  Rates: Maximum application rates range from 0.5 lb/ai/A to 8 lb/ai/A. The maximum number of applications per year range from 1 to 3. Up to 4 applications are permissible in some citrus growing areas (grove floor treatment).

  Timing: Dormant, delayed dormant, preplant, at-planting, transplanting, postplant, post-transplant, preemergence and postemergence.

- **Use Classification**: Any emulsifiable concentrate (EC) end-use product formulated from chlorpyrifos must be labeled as a restricted use product. All other end-use products (other than containerized baits in child-resistant packaging) must either be labeled as restricted use or packaged in containers no smaller than 15 gallons of a liquid formulation or 25 pounds of a dry formulation.

D. Estimated Usage of Pesticide

This section summarizes the best estimates available for many of the pesticide uses of chlorpyrifos, based on available pesticide usage information for 1987-1998. Approximately 21 million pounds a.i. of chlorpyrifos were used annually, according to Agency and registrant estimates. As a result of the June 7, 2000 MOA, which eliminated residential uses and phased out the termite uses, approximately 10 million pounds of chlorpyrifos will be phased out of the market place. Table 2 provides usage estimates for selected use sites. A full list of all uses of chlorpyrifos, with the corresponding use and usage data for each site, has been completed and is in the "Quantitative Use Analysis," March 30, 2000, which is available in the public docket and on the internet. The data, reported on an aggregate and site (crop) basis, reflect annual

fluctuations in use patterns as well as the variability in using data from various information sources. These estimates do not reflect reductions in use from mitigation that has been implemented as a result of the Memorandum of Agreement.

Table 2. Chlorpyrifos Estimated Usage for Representative Sites

Crop	Lbs. Active Ingredient Applied (Wt. Avg.)[1]	Percent Crop Treated (Likely Maximum)	Percent Crop Treated (Wt. Avg.)
Cranberries	26,000	60	47
Oranges	460,000	19	14
Oranges, Fresh	350,000	54	41
Oranges, Processed	110,000	10	7
Apples	550,000	53	44
Pecans	240,000	36	20
Walnuts	197,000	39	30
Sweet Corn	120,000	13	11
Sweet Corn, Fresh	74,000	22	18
Sweet Corn, Processed	46,000	9	7
Corn	5,527,000	8	7
Broccoli	73,000	51	41
Brussels Sprouts	9,000	91	73
Cauliflower	27,000	36	31
Tobacco	146,000	14	11
Wheat, Winter	170,000	1	1
Alfalfa	480,000	3	3
Peanuts	316,000	15	10
Cotton	670,000	6	5
Sugar Beets	169,000	10	8
Nursery/Greenhouse	277,000	–	–
PCOs, Termite Control[2]	5,003,000	–	–
PCOs, Other (Roaches, Ants, Fleas, etc.)[2]	1,946,000	–	–

Crop	Lbs. Active Ingredient Applied (Wt. Avg.)[1]	Percent Crop Treated (Likely Maximum)	Percent Crop Treated (Wt. Avg.)
Mosquito Abatement Districts	29,000	–	–
Turf[3, 4]	2,519,000	–	–
Households, Outdoor[4]	1,112,000	–	–

[1] Weighted average is based on data for 1987-1998; the most recent years and more reliable data are weighted more heavily.
[2] Mitigation implemented in June 2000 included phase-out or cancellation of products for this use.
[3] Includes golf courses, turf farms, institutional turf, lawncare control operators, and landscape contractors.
[4] Products registered for residential use were cancelled effective December 31, 2000.

III. Summary of Chlorpyrifos Risk Assessment

Following is a summary of EPA's revised human health and ecological risk findings and conclusions for the organophosphate pesticide chlorpyrifos, as fully presented in the documents, *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000, and *Fate and Environmental Risk Assessment*, dated June 2000, and addenda thereto. The purpose of this summary is to assist the reader by identifying the key features and findings of these risk assessments, and to better understand the conclusions reached in the assessments.

These risk assessments for chlorpyrifos were presented at a Technical Briefing on June 8, 2000, which was followed by an opportunity for public comment on risk management for this pesticide. The risk assessments presented here form the basis of the Agency's risk management decision for chlorpyrifos only; the Agency must consider cumulative risks of all the organophosphate pesticides before any final decisions can be made.

A. Human Health Risk Assessment

EPA issued its preliminary risk assessments for chlorpyrifos in Phase 3 of the public participation process on October 18, 1999. In response to comments and new studies submitted during Phase 3, and mitigation measures agreed to by the technical registrants to address risks identified in the preliminary assessments, the risk assessments were updated and refined. The major revision to the human health risk assessment was the reassessment of acute dietary risks to reflect the cancellation of the tomato use and reduction of the grape and apple tolerances to 0.01 ppm; inclusion of new data from the Agricultural Reentry Task Force (ARTF); and preliminary consideration of a new acute study with human subjects and a new oral dog study with peripheral nervous system measurements. The registrant has submitted a rebuttal to the modification of the tolerances. This rebuttal is under review.

1. Dietary Risk from Food

a. Toxicity

The Agency has reviewed all toxicity studies submitted and has determined that the toxicity database is complete, and that it supports an interim reregistration eligibility determination. A brief overview of the studies used for the dietary risk assessment is outlined in Table 3 in this document. Further details on the toxicity of chlorpyrifos can be found in the Human Health Risk Assessment for Chlorpyrifos, June 8, 2000.

Table 3. Summary of Doses and Endpoints Selected for Chlorpyrifos Dietary Risk Assessment

Exposure Scenario	NOAEL/Dose (mg/kg/day)	Endpoint	Study
Acute Dietary	NOAEL=0.5 UF = 100 FQPA = 10 (infants, children and females 13-50)	Significant (28-40%) plasma ChE inhibition at peak time of (3-6 hours post exposure) at 1 mg/kg/day (Mendrala and Brzak 1998). Significant 30% RBC ChE inhibition 4 hours post exposure at the LOAEL of 1.5 mg/kg/day (Zheng et al. 2000).	Acute Blood Time Course Study in male rats (Mendrala and Brzak 1998) with support from Zheng et al. (2000)
	Acute RfD =0.005 mg/kg/day **Acute PAD (children and females 13-50) = 0.0005 or $5x10^{-4}$ mg/kg/day** **Acute PAD (general population) = 0.005 or $5x10^{-3}$ mg/kg/day**		
Chronic Dietary	NOAEL= 0.03 UF= 100 FQPA = 10 (infants, children and females 13-50)	Significant plasma and RBC cholinesterase inhibition at the LOAEL of 0.22 to 0.3 mg/kg/day	Weight of Evidence from 5 studies: 2 year dog 90 day dog 2 year rat 90 day rat developmental neurotoxicity (DNT) rat study (at 2 weeks)
	Chronic RfD =0.0003 mg/kg/day **Chronic PAD (children and females 13-50) = 0.00003 or $3x10^{-5}$ mg/kg/day** **Chronic PAD (general population) = 0.0003 or $3x10^{-4}$ mg/kg/day**		

NOAEL = No Observed Adverse Effect Level
RBC = red blood cell
UF = Uncertainty Factor
PAD = Population Adjusted Dose (includes UF and FQPA safety factor)

The Agency has evaluated the potential impact on the acute dietary risk assessment following the submission of an acute (single oral dose) toxicity study with chlorpyrifos in humans. The following observations can be made on the potential impact of these data on the chlorpyrifos risk assessment. Because the study is a single oral dose, it could be used in a weight-of-evidence approach to inform the selection of the inter-species uncertainty factor for

acute dietary risk assessment. The Agency's evaluation did not include an independent review of the ethical standards under which this study was conducted. The acute human study could be compared to existing acute animal data to determine if the full ten-fold inter-species uncertainty factor is needed to account for variation between species in the acute dietary assessment. However, because of its limited duration, this study would not be adequate for use in short-term or intermediate-term risk assessments, such as those used to estimate worker risk from chlorpyrifos use, nor would it be appropriate for the chronic dietary assessment.

The Agency has concluded that the primary metabolite of chlorpyrifos, 3,5,6-trichloro-2-pyridinol (TCP), does not induce cholinesterase inhibition, and exhibits effects only at doses high than those producing ChEI with chlorpyrifos, and therefore is less toxic than chlorpyrifos (58 FR 19354, April 14, 1993). The primary toxicological effect after subchronic and chronic exposure to TCP was alterations in liver enzymes seen at 30 mg/kg/day and increases in liver and kidney weights at 100 mg/kg/day. Because of the potential exposure to TCP in food and residential settings, and evidence of increased susceptibility of rabbit fetuses relative to dams, a screening-level dietary risk assessment for TCP resulting from chlorpyrifos, chlorpyrifos-methyl and trichlorpyr was conducted. That assessment indicated that the percentage of the acute PAD occupied for females 13+ years old (the population subgroup of concern for acute toxicity effects) was 2.4%. The percentage of the chronic PAD occupied ranged from 0.3% for the general U.S. population to 0.7% for children 1-6 years old. Upper-bound estimated environmental concentrations of TCP exceeded chronic DWLOCs for children. However, the Agency believes that actual concentrations are probably considerably lower than modeled values primarily because the acres treated with chlorpyrifos in any watershed is expected to be much lower than 100% assumed in the modeling. Uncertainties with surface and groundwater modeling are discussed more fully in the Summary of Risks to Nontarget Organisms later in this document. More detailed information on TCP and the screening assessment can be found in the "Preliminary Risk Assessment for Trichloropyridinol (TCP) Metabolite," June 5, 2000, which is available in the public docket and on the internet at www.epa.gov/pesticides/op.

b. FQPA Safety Factor

The FQPA 10X Safety Factor has been retained due to increased susceptibility and sensitivity to chlorpyrifos among neonates when compared with adults, and for the qualitative increased susceptibility occurring at the high dose in the developmental neurotoxicity (DNT) study (cholinesterase inhibition in dams versus structural effects on developing brain of the offspring). In addition, recent data in the literature suggest that the inhibition of cholinesterase may not be essential for adverse effects on brain development. Further uncertainty arises from the lack of an offspring No Observed Adverse Effect Level (NOAEL) in the DNT. In that study, structural alterations in brain development were the toxicity endpoint of concern and were seen at the lowest dose tested. The registrant has submitted a rebuttal to the EPA review of the DNT study. This rebuttal is under review.

The FQPA Safety Factor is applicable to females 13-50 as well as infants and children, for all exposure durations. The FQPA Safety Factor is applicable to the following assessments:

- Acute Dietary Assessment - The FQPA safety factor is applicable to the Females 13-50 and Infants and Children population subgroups for the acute dietary assessment because adverse effects could result from a single exposure to chlorpyrifos (as demonstrated in several open literature studies including Zheng et al.).

- Chronic Dietary Assessment - The FQPA safety factor is applicable to the Females 13-50 and Infants and Children population subgroups due to the concern that potential adverse effects could result from repeated exposure to chlorpyrifos (as demonstrated, for example, in the developmental neurotoxicity study in rats).

- Residential and Other Non-Occupational Exposure Assessment - The FQPA safety factor is applicable for Females 13-50 and the Infants and Children population subgroups for all exposure durations due to the adverse effects resulting from single and repeated exposure(s) to this organophosphate insecticide in and around residential (non-occupational) settings.

c. Population Adjusted Dose (PAD)

The Population Adjusted Dose, or PAD, is a term that characterizes the dietary risk of a chemical, and reflects the Reference Dose (RfD), either acute or chronic, that has been adjusted to account for the FQPA safety factor (i.e., RfD/FQPA safety factor). A risk estimate that is less than 100% of the acute or chronic PAD does not exceed the Agency's risk concern.

d. Exposure Assumptions

Chlorpyrifos is registered for use on a wide variety of food crops, and has approximately 112 tolerances for food and/or feed commodities (which translates to approximately 700 food forms in the dietary analysis). Food uses evaluated in this analysis were those reflected by the established tolerances in/on raw agricultural, animal, and processed food/feed commodities for chlorpyrifos as listed in 40 CFR §180.342. Food handling establishment (FHE) tolerances were also included as cited in 40 CFR §180.342(a)(4) for the chronic dietary analysis (i.e., as a result of the registered use in FHE, all foods have an established tolerance of 0.1 ppm, unless they are covered by higher tolerances). The established tolerances in/on raw agricultural, animal, and processed food/feed commodities are expressed either in terms of the combined residues of chlorpyrifos and its metabolite TCP or as chlorpyrifos *per se*. The Agency has determined that residues of TCP are not of concern for the chlorpyrifos dietary assessment, and concluded that it can therefore be excluded from the tolerance expression. Proposed tolerances are supported by available residue chemistry data and are expressed in terms of chlorpyrifos *per se*. Thus, for purposes of this analysis, only residues of chlorpyrifos *per se* were considered, when data were available. Whenever possible, data for anticipated residues (ARs) reflect levels of chlorpyrifos *per se*.

Highly refined acute and chronic dietary risk analyses for chlorpyrifos were conducted with the Dietary Exposure Evaluation Model (DEEM™). DEEM incorporates consumption data generated in USDA's Continuing Surveys of Food Intakes by Individuals (CSFII), 1989-91. For

chlorpyrifos, inputs to the DEEM analysis also include DAS's National Food Survey (NFS, 1993-1994), U.S. Department of Agriculture's Pesticide Data Program (PDP) monitoring data (1994-1999), the Food and Drug Administration (FDA) Surveillance Monitoring Program data (1992-1998), and field trial residue data. Percent crop treated data were supplied by EPA's Biological and Economic Analysis Division (see Quantitative Usage Analysis for Chlorpyrifos, March 30, 2000, available in the public docket). Where percent crop treated estimates indicated no chlorpyrifos use, a default assumption of 1% crop treated was applied. In general, when residues on commodities were nondetectable, one-half the limit of detection (LOD) was assumed. All available processing and cooking factors were incorporated into the dietary exposure analysis.

For chronic dietary risk assessments, the three-day average of the consumption data for each subpopulation is combined with average residues in commodities to determine the average exposure in mg/kg/day. For acute dietary risk assessment, the entire distribution of single day food consumption events is combined with a distribution of residues (probabilistic analysis, referred to as "Monte Carlo") to obtain a distribution of exposures in mg/kg/day.

e. Food Risk Characterization

Generally, a dietary risk estimate that is less than 100% of the acute or chronic PAD does not exceed the Agency's risk concerns. A summary of acute dietary risk estimates is shown in Table 4. Based on use patterns before the June 2000 mitigation agreement, the chlorpyrifos acute dietary risk from food at the 99.9th percentile for the most highly exposed subpopulation, children 1-6 years old, was 355% of the aPAD.

Commodities that contribute the most to that risk estimate are apples (residues resulting from post-bloom uses), grapes (residues primarily on imported crops) and fresh tomatoes (residues primarily on imported crops). Measures agreed to in the June 2000 agreement addressed these risks by canceling use on tomatoes and revoking the associated tolerance; restricting use on apples to pre-bloom (dormant) applications and reducing the tolerance to 0.01 ppm to reflect this new use pattern; and reducing the tolerance on grapes to 0.01 ppm to reflect the domestic dormant use pattern. The registrant has submitted a rebuttal to the modification of the tolerances. This rebuttal is under review.

With these measures in place, at the 99.9th percentile, the dietary risk from food alone is below 100% of the aPAD for all population subgroups, including the most sensitive population subgroup, children 1-6 years old, with 82% of the aPAD occupied. Thus acute dietary risks from food alone are not of concern.

Table 4. Acute Dietary (Food Only) Risk Estimates for Chlorpyrifos as Percent of aPAD

Subpopulation	Pre-Mitigation[1] 99.9th Percentile	Post-Mitigation[2] 99.9th Percentile
U.S. population	16%	4.1%
All infants	130%	50%
Children 1-6	355%	82%
Children 7-12	270%	62%
Females 13+ , nursing	130%	39%

[1]Pre-mitigation refers to uses/use patterns in effect prior to the June 2000 mitigation agreement.
[2]Post-mitigation reflects changes in use/use patterns for tomatoes, apples and grapes as set forth in the June 2000 mitigation agreement.

The chronic dietary risk from food alone is not of concern, as shown in Table 5. Input values included PDP, FDA and Dow AgroSciences' (DAS')1993 National Food Survey (NFS) (a market basket survey), average residues from field trials, and percent crop treated data compiled by the Agency. Exposure estimates were below 100% of the cPAD for the most highly exposed subgroup, children 1-6 years old. With mitigation measures for apples, tomatoes and grapes in place per the June 2000 agreement and assuming use in food handling establishments, exposure for children 1-6 years old, the highest exposure subgroup, occupies 51% of the cPAD, and thus is not of concern.

Table 5. Chronic Dietary (food only) Risk Estimates for Chlorpyrifos as Percent of cPAD

Subpopulation	Pre-Mitigation[1] 99.9th Percentile	Post-Mitigation[2] 99.9th Percentile
U.S. population	4%	2.5%
All infants	45%	33%
Children 1-6	81%	51%
Children 7-12	59%	36%
Females 13+ , nursing	30%	20%

[1]Pre-mitigation refers to uses/use patterns in effect prior to the June 2000 mitigation agreement.
[2]Post-mitigation reflects changes in use/use patterns for tomatoes, apples and grapes as set forth in the June 2000 mitigation agreement.

These assessments are the most refined estimates of risk from exposure to chlorpyrifos through food, although some uncertainties exist. PDP data indicate that chlorpyrifos residues were detected in several commodities for which tolerances do not exist, specifically spinach, carrots, squash, lettuce, potatoes and celery. These residues were not included in the Agency's risk estimates because they represent misuse of chlorpyrifos. However, additional assessments

were conducted using spinach, carrots and squash, the commodities most frequently fed to children. These assessments were not significantly different from the mitigated acute or chronic dietary assessments and thus are not of concern.

A tolerance also does not exist for chlorpyrifos in freshwater fish. In a screening level assessment of the health risks to individuals who consume freshwater fish conducted by the EPA Office of Water in 1992, residues of chlorpyrifos were detected in fish from 26% of 388 sample collection sites. These data suggest that consumption of freshwater fish could contribute to the dietary exposures and risks from chlorpyrifos for sports fishermen and subsistence populations. Risk estimates could be of concern for an individual who consumed the maximum detected residue level daily for 70 years at a rate of 170 g/day; however, the Agency considers this unlikely. Subsistence populations are not expected to have exposures or risk that exceed the Agency's level of concern following chronic ingestion of fish fillets containing the mean detected residue level. For a more detailed discussion of risks from freshwater fish consumption, please refer to the *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000.

2. Dietary Risk from Drinking Water

Drinking water exposure to pesticides can occur through ground water and surface water contamination. EPA considers both acute (one day) and chronic (lifetime) drinking water risks and uses either modeling or actual monitoring data, if available, to estimate those risks. For chlorpyrifos, ground and surface water monitoring data were used as well as conservative Tier 1 and Tier 2 modeling. Modeling is considered to be an unrefined assessment and can provide a high-end estimate of risk.

The GENEEC and PRZM-EXAMS models were used to estimate surface water concentrations, and SCI-GROW was used to estimate groundwater concentrations. All of these are considered to be screening models, with the PRZM-EXAMS model being somewhat more refined than the other two.

The available environmental fate data suggest that chlorpyrifos has a low potential to leach to groundwater in measurable quantities from most typical agricultural uses, except following termiticide use. Chlorpyrifos is persistent in concentrated applications used in termiticide treatments. The available data indicate that the primary metabolite of chlorpyrifos, TCP is more mobile and significantly more persistent in many soils, especially under anaerobic conditions. A screening-level dietary risk assessment for TCP indicated that drinking water exposure following termiticide use may pose risks of concern to children. Generic risk mitigation action for termiticides has been implemented. The technical registrants agreed in June 2000 to a suite of mitigation measures for the termiticide products that will reduce the potential for exposure from this use. By December 31, 2000, the application rate was reduced to a 0.5% solution, and use was restricted to professional applicators. After December 31, 2001, whole house (post-construction) treatment will not be allowed. The preconstruction termiticide use will be eliminated by December 31, 2005, unless the registrants submit acceptable exposure data that demonstrate that risks are not of concern.

a. Surface Water

The Agency examined data of over 3000 samples from 20 of the U.S. Geological Survey's National Water Quality Assessment (NAWQA) Program study units for flowing surface water collected from rivers and streams. Chlorpyrifos was detected in 15% of 1530 agricultural streams, 26% of 604 urban stream samples in 1997 and in 65% of 57 urban stream samples from Georgia, Alabama and Florida in 1994. The maximum reported dissolved chlorpyrifos concentration in surface water was 0.4 ppb, with the majority of detections below 0.1 ppb. Although the data represent a large part of the U.S., they may not represent the most vulnerable watersheds where chlorpyrifos use is pervasive. A limited number of watersheds in the U.S. may have chlorpyrifos concentrations greater than 0.4 ppb due to higher usage rates or greater pesticide runoff. In particular, acute exposure levels could be higher for streams draining watersheds with more intense chlorpyrifos use or for lakes and reservoirs for which there are little data.

For comparison, the Agency developed screening-level model estimates of chlorpyrifos concentrations in surface water such as lakes and reservoirs using Tier I GENEEC and Tier II PRZM/EXAMS. Inputs to the models included high exposure agricultural scenarios for major crops (alfalfa, corn, citrus, and tobacco) at the maximum application rates. Estimated 90-day average and peak concentrations of chlorpyrifos in surface water using the PRZM/EXAMS screening model were 6.7 ppb and 40.6 ppb, respectively. The modeled estimates represent a pond draining an adjacent 100% treated field. These estimates should be highly conservative for most surface waters and all drinking water because it is unlikely that 100% of a watershed constituting a major drinking water source would be treated with chlorpyrifos in a given year.

After comparison of the NAWQA monitoring data and modeled estimates, an upper-bound range of concentrations was selected from the NAWQA study to assess acute and chronic risks associated with non-termiticide uses for surface water. For the acute assessment, a range of 0.026 to 0.4 ppb was used. The 0.026 ppb represents the 95th percentile chlorpyrifos concentration, while the 0.4 ppb concentration is the maximum detected concentration from streams and rivers. Estimated environmental concentrations (EECs) used in the assessments are shown in Table 6.

Table 6. Surface and Groundwater EECs for Chlorpyrifos

Drinking Water Source	Estimated Environmental Concentration (ppb)	
	Acute	Chronic
Groundwater	0.007 to 0.103 (a)	
Surface water	0.026 to 0.4 (b)	0.026 (c)

(a) Concentrations predicted by screening-level model SCI-GROW. The value is considered an upper bound concentration estimate.

(b) Based on the 95th percentile and maximum detected concentrations from surface water monitoring data.

(c) Based on the 95th percentile surface water concentration from monitoring data

To assess chronic risks, 0.026 ppb was used. As indicated above, 0.026 ppb represents the 95th percentile concentration from the NAWQA study. Although PRZM/EXAMS predicted a peak concentration of 40.6 ppb for lakes and reservoirs, this estimate was not used to assess chronic risks for the following reasons: 1) multi-month or annual mean concentrations in a reservoir are expected to be less than the maximum reported concentrations in the flowing water feeding the reservoir, which in this case is 0.4 ppb; therefore 40.6 ppb is unlikely to occur; and 2) the monitoring data demonstrate that chronic concentrations of chlorpyrifos in surface water are unlikely to exceed 0.1 ppb.

b. Ground Water

The Agency examined data of over 3000 samples of filtered well monitoring samples from the NAWQA database, and in the Agency's Pesticides in Ground Water Data Base (PGWDB). The NAWQA data showed that chlorpyrifos was detected in groundwater in fewer than 1% of the 3000 wells sampled, with the majority of concentrations reported at <0.01 ppb, and occasional detections at a maximum level of 0.026 ppb. Although the available monitoring data represent a large part of the U.S., it is not clear that they represent the most vulnerable groundwater where chlorpyrifos is used most intensively. The PGWDB reports a maximum detected concentration of 0.65 ppb.

Chlorpyrifos concentrations in groundwater were also estimated using the screening-level model SCI-GROW for four crops (corn, cotton, alfalfa and citrus). SCI-GROW predicted chlorpyrifos concentrations ranging from 0.007 ppb (typical application to alfalfa) to 0.103 ppb (maximum multiple applications to sweet corn). An analysis of both monitoring and modeling data suggest that chlorpyrifos concentrations in 99% of potable water in the U.S. are unlikely to exceed 0.1 ppb. Based on these data, EECs ranging from 0.007 to 0.103 ppb were used to evaluate both acute and chronic exposures for groundwater. The NAWQA monitoring data support that the SCI-GROW estimates are conservative.

Chlorpyrifos use as a termiticide is significant, with a recent estimate of seven million pounds ai applied annually, constituting about 30% of the total annual use. Chlorpyrifos groundwater exposure from termiticidal use occurs only in wells located within 100 feet of the treatment area and when the well casing is cracked. The maximum reported dissolved concentration following termiticide use is 2090 ppb. The current U.S. EPA Health Advisory for a child is 30 ppb. Therefore, acute concentrations are estimated at 30 to 2090 ppb. Chronic concentrations are presumably significantly lower but persistent at detectable levels for at least six months. Chronic concentrations following this use are estimated at 8.3 to 578 ppb. These values were derived by adjusting the acute concentrations for partial environmental degradation.

The Agency is concerned about exposure associated with termiticide use. However, because these exposures are isolated incidents and because termiticide use is being phased down with immediate reduction in applied concentrations, these exposures were not included in the dietary risk assessment. The following points support this determination. First, the technical

registrants state that this exposure only occurs in homes where the well is near or in the foundation and the well casing is cracked. The Agency has determined that because of changes made to termiticide labels as a result of the Label Improvement Process for Termiticides (PR Notice 96-7 for termiticides), potential exposure from incidents of this type has been reduced. For example, reported incidents associated with termiticide use were 28.2 per 100,000 homes in 1997 (before PR 96-7), and were 8.3 per 100,000 homes in 1998 (after PR 96-7).

Secondly, the technical registrants agreed in June 2000 to a suite of mitigation measures for termiticide products that reduced the potential for exposures from this use. By December 31, 2000, the application rate was reduced to a 0.5% solution, and use was restricted to professional applicators. After December 31, 2001, whole house (post-construction) treatment will not be allowed. By December 31, 2005, all residential termiticide use will be canceled.

c. Drinking Water Levels of Comparison (DWLOCs)

To determine the maximum allowable contribution of water-containing pesticide residues permitted in the diet, EPA first looks at how much of the overall allowable risk is contributed by food (and if appropriate, residential uses), and then determines a "drinking water level of comparison" (DWLOC) to determine whether modeled or monitored concentrations exceed this level. The Agency uses the DWLOC to estimate risk associated with exposure to pesticides in drinking water. The DWLOC is the maximum concentration in drinking water which, when considered together with dietary exposure, does not exceed a level of concern.

For acute risk, the potential drinking water exposure derived from either ground or surface water is not of concern for any population subgroup. Long-term exposure to chlorpyrifos as a result of well contamination from termiticide use could result in exposures of concern; however, these incidents are unlikely given ongoing mitigation. In addition, the technical registrants have agreed to reductions in use in the interim until all termiticide use is canceled. This is discussed in greater detail above and in Section IV of this document.

Table 7 presents the calculations for the acute and chronic drinking water assessment. Details of this analysis are found in the *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000.

Table 7. Drinking Water DWLOC and EEC Comparisons (Excluding Well Contamination)

Population Subgroup	DWLOCS (ppb)		Estimated Environmental Concentrations (ppb)		
			Ground Water	Surface Water	
	Acute	Chronic	Acute and Chronic	Acute	Chronic
U.S. Population	166	10	0.007-0.103	0.026-0.4	0.026
All Infants (<1 year)	2.4	0.2			
Children (1-6 years)	0.9	0.15			
Females (13-50 years)	9	0.72			

3. Occupational and Residential Risk

a. Toxicity

All risk calculations in this assessment are based on the most current toxicity information available for chlorpyrifos, including a 21-day dermal toxicity study. The toxicological endpoints and other factors used in the occupational and residential risk assessments for chlorpyrifos are shown in Table 8.

Table 8. Toxicological Endpoints and Other Factors Used in the Occupational and Residential Risk Assessment for Chlorpyrifos

Exposure Scenario	NOAEL Dose (mg/kg/day)	Endpoint	Study	Target MOE for Occupa-tional	Target MOE for Residential/Homeowner Exposures
Dermal Short-Term 1-30 days	Dermal NOAEL =5 Absorbed Dermal NOAEL = 0.15 (for biomonitoring) (a)	Plasma and RBC cholinesterase inhibition of 45 and 16%, respectively at LOAEL of 10 mg/kg/day after 4 days. (Dermal absorption factor not necessary)	21-day dermal rat study	100	1000 (infants, children and females 13-50) 100 (all other subpopulations)
Dermal Intermediate-Term (1–6 months) Long-Term (>6 months)	Oral NOAEL = 0.03 (3% dermal absorption)	Plasma and RBC cholinesterase inhibition at LOAEL of 0.22 to 0.3 mg/kg/day	Weight of Evidence from 5 studies: 2 year dog , 90 day dog, 2 year rat, 90 day rat, DNT study (at 2 weeks)	100	1000 (infants, children and females 13-50) 100 (all other subpopulations)
Inhalation Short-Term (1-30 days) Intermediate-Term (1–6 months)	Inhalation NOAEL = 0.1	Lack of effects in 2 rat inhalation studies at the highest dose tested; 43% plasma and 41% RBC cholinesterase inhibition following oral doses of 0.3 mg/kg/day for 2 weeks in the DNT study	Two 90 day rat inhalation studies (NOAEL) and DNT (LOAEL)	100	1000 (infants, children and females 13-50) 100 (all other subpopulations)

Exposure Scenario	NOAEL Dose (mg/kg/day)	Endpoint	Study	Target MOE for Occupa-tional	Target MOE for Residential/Homeowner Exposures
Inhalation Long-Term (>6 months)	Oral NOAEL= 0.03 (assume inhalation absorption is 100% of oral absorption)	Significant plasma and RBC cholinesterase inhibition at 0.22 to 0.3 mg/kg/day	Weight of Evidence from 5 studies: 2 year dog, 90 day dog, 2 year rat, 90 day rat, DNT (at 2 weeks)	100	1000 (infants, children and females 13-50) 100 (all other subpopulations)

NOAEL = No Observed Adverse Effect Level
RBC = red blood cell
UF = Uncertainty Factor
PAD = Population Adjusted Dose (includes UF and FQPA safety factor)
(a) For comparison with absorbed biomonitoring data, use dermal NOAEL of 0.15 mg/kg/day * 0.03 dermal absorption factor

The Agency has evaluated a 6-week dietary study in dogs designed to assess cholinesterase inhibition (ChEI) in peripheral nervous system (PNS) tissues, such as the heart and leg muscles, as well as measure cholinesterase activity in the blood and brain. The study was conducted by DAS in Michigan to address regulatory requirements in the United Kingdom. This type of study is not required under current EPA guidelines, but the Agency has recommended direct measurement of ChEI in the target peripheral nervous system tissues as a potential alternative to measuring ChEI in the blood only.

This study conducted with beagle dogs was designed to assess for inhibition of red blood cell (RBC), peripheral tissue (brain, nodose ganglion, left atrium, diaphragm and quadriceps muscle) and brain acetylcholinesterase (AChE). A separate report presented a histopathological evaluation of the adrenal gland.

All dogs survived the six week study and there were no clinical signs or effects on body weight or food consumption. There were also no histopathological alterations in the adrenal gland noted in the special assessment of this organ. The results of this study demonstrates that in the dog, RBC AChE is more sensitive than brain or peripheral tissue AchE. Overall, the peripheral tissue data were considered too variable and the cohort of dogs too small to make a meaningful evaluation of potentially small changes in AChE activity in these structures. There were, however, sufficient data to *imply* that peripheral tissue was not demonstrated to be inhibited by chlorpyrifos. No *definite* conclusions that chlorpyrifos inhibits peripheral tissue AChE can be drawn from the data with the four peripheral tissue preparations. The peripheral tissue aspects of the study cannot be upgraded due to the small number of animals assessed and the variability of the data.

If another study was conducted that addressed the study deficiencies and limitations as described in the data evaluation record and found to be acceptable, the following observations could be made on the potential impact of these data on the chlorpyrifos risk assessment. Because the study would be a repeat dose over a 6 week period, it could be used in a weight-of-evidence approach to inform the selection of short and intermediate term endpoints for the chlorpyrifos worker risk assessment. Taking into account the established dermal absorption rate of rate of 3%, this study would yield MOEs 3-6 times greater than those currently shown in EPA's assessment. At a minimum, if the data are reliable, they could increase the confidence that EPA's current assessment does not underestimate worker risk.

The Agency uses the results of acute toxicity studies to determine early entry PPE and other labeling requirements. Acute toxicity values and categories for the technical grade of chlorpyrifos are summarized in Table 9. Chlorpyrifos is moderately toxic following acute oral, dermal and inhalation exposures, and is classified in toxicity category II for all three routes of exposure for rats.

Table 9. Acute Toxicity Profile for Occupational Exposure for Chlorpyrifos

Study	MRID Number	Results	Toxicity Category
Acute Oral LD_{50} - rat	44209101	223 mg/kg M&F	II
Acute Dermal LD_{50} - rat	Accession No. 112115	202 mg/kg	II
Acute Dermal LD_{50} - rabbit	44209102	>5000 mg/kg	IV
Acute Inhalation LC_{50} - rat	00146507 and Acc.No. 257590	LC_{50} > 0.2 mg/L (200 mg/m^3) (nominal concentration)	II
Eye Irritation - rabbit	44209103	slight irritation resolved within 24 hours	IV
Dermal Irritation - rabbit	44209104	mild irritant; (irritation resolved within 7 days)	IV
Dermal Sensitization - guinea pig	44209105	non-sensitizing	NA
Acute Delayed Neurotoxicity - hens	00097144 00405106	not neurotoxic at 50, 100 or 110 mg/kg	NA

NA = Not Applicable

b. Occupational Exposure and Risk

1) Occupational Handler Exposure

Several chemical-specific handler exposure studies conducted and submitted by the technical registrants measured the exposures to professional pesticide applicators during application of chlorpyrifos products. These data include biological monitoring of urinary TCP, the primary metabolite of chlorpyrifos, and passive dosimetry data. In the absence of chemical-specific data, the Pesticide Handlers Exposure Database (PHED) Version 1.1 was used to assess potential exposures resulting from handling and applying chlorpyrifos. The exposure factors (e.g., body weight, amount treated per day, protection factors, etc.) are all standard values that are used by the Agency, and the PHED unit exposure values are the best available estimates of exposure. Nevertheless, it should be noted that some aspects of the included studies (e.g., duration, acres treated, pounds of active ingredient handled) may not accurately represent labeled uses in all cases. Further details on the data used for the assessments are discussed in the *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000, which is available in the public docket and on the internet at www.epa.gov/pesticides/op.

Anticipated use patterns and application methods, range of application rates, and daily amount treated were derived from current labeling and other available information. Application rates specified on chlorpyrifos labels range from 0.25 to 8 pounds of active ingredient per acre.

The Agency typically uses acres treated per day values that are thought to represent a typical work day for specific types of application equipment.

Occupational handler exposure assessments are conducted by the Agency using different levels of personal protective equipment (PPE). The Agency typically evaluates all exposures in a step-wise fashion, first assuming minimal protection and then incrementally adding protective measures until the target MOE is reached. For agricultural handlers, the estimated exposures considered PPE (a double layer of clothing and gloves and/or a dust/mist respirator), and engineering controls (closed mixing/loading systems and enclosed cabs/trucks).

The Agency identified 31 major occupational handler scenarios for which there were potential exposures during mixing, loading, and applying products containing chlorpyrifos to agricultural crops and ornamentals (22 scenarios) and to non-agricultural use sites (9 scenarios) such as sodfarms, golf courses and mosquito adulticide treatment. These scenarios reflect a broad range of application equipment, application methods and use sites. For agricultural uses, handler activities include open and closed mixing/loading, and aerial, tractor-drawn and handheld application. The application rates used in the assessment are intended to reflect the upper range of rates on the labels. In some instances, the rates also include values that registrants indicated were "typical" (e.g., a variety of sod farm rates, corn, citrus, greenhouse, and nursery rates).

The scenarios were classified as short-term (1 to 30 days) and intermediate-term (1 to 6 months). The handler scenarios for agricultural and golf course uses are expected to be of short-term duration only; the scenarios for mosquitocide use are short- and intermediate-term; and the scenario for pre-termiticide treatment is long-term (>6 months).

2) Occupational Handler Risk

Agricultural and Ornamental/Greenhouse Handler Risk

Combined dermal and inhalation margins of exposure for agricultural, ornamental and greenhouse handlers range from 8 to 10,890. The following exposure scenarios (by number as presented in Table 10) result in MOEs below 100 with engineering controls (or with PPE where engineering controls are not feasible) and thus are of concern:

(1a) Mixing/loading liquids for aerial/chemigation application at 1.5 lbs. ai/A
(1b) Mixing/loading liquids for groundboom application at 5 lbs. ai/A
(2a) Mixing wettable powder for aerial/chemigation application at 2 and 3.5 lbs. ai/A
(2b) Mixing wettable powder for groundboom application at 3 lbs. ai/A
(4a) Aerial application of spray in enclosed cockpit at 2 lbs. ai/A
(4b) Aerial application of granular in enclosed cockpit at 1.95 lbs. ai/A
(12) Application by backpack sprayer at 0.08 and 0.16 ai/gal, and at 3.5 lbs. ai/A

(14) Application by high-pressure handwand at 0.0033 and 0.0066 lbs. ai/gal
(15) Application by hydraulic hand-held sprayer for bark beetle treatment at 3.5 lbs. ai/A and at 0.08 lbs. ai/gal

Seed treatment, pre-plant peach dip and dry bulk fertilizer impregnation were not assessed due to a lack of appropriate data.

Table 10. Occupational Risk Estimates for Agricultural and Ornamental Uses of Chlorpyrifos

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre) (a)	Daily Acres Treated (b)	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
Mixer/Loader Exposure								
Mixing/Loading Liquids for Aerial/Chemigation Application (1a)	1.5 cranberries, corn	350	39	56	23	78	160	52
	3.5 citrus (c)	100	59	83	34	120	240	78
Mixing/Loading Liquids for Groundboom Application (1b)	1.5 predominant max	80	170	240	100	Target MOE reached at PPE		
	5.0 tobacco max (d)	80	51	73	30	100	210	69
	2 Sodfarm (includes tobacco/potatoes)	80	130	180	75	250	530	170
	4 Sodfarm (e)	80	64	91	38	130	260	86
	8.0 sodfarm fire ants	10	260	360	150	Target MOE reached at PPE		
Mixing/Loading Liquids for Airblast Application (1c)	2.0 predominant max such as Fruits & Nuts	40	260	360	150	Target MOE reached at PPE		
	6.0 citrus	20	170	240	100	Target MOE reached at PPE		
Mixing WP for Aerial/Chemigation Application (2a)	2.0 predominant max (orchards)	350	DAS is not supporting the open bag formulation for the WP			51	42	23
	3.5 citrus (c)	100				100	83	46
Mixing WP for Groundboom Application (2b)	1.0 predominant max (brassica)	80				450	360	200
	4.0 soil treatment ornamentals outdoors	10				890	730	400
	1.3 & 3.0 Sodfarm	80				340 / 150	280 / 120	150 / 67
	8.0 sodfarm fire ants (harvest only)	10				4500	3600	200

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre) (a)	Daily Acres Treated (b)	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
Mixing WP for Airblast Application (2c)	2.0 predominant max	40				450	360	200
	6.0 citrus	20				300	240	130
Loading Granulars for Aerial Application (3a)	1.95 maximum aerial rate (f)	350	150	30	25	3000	300	270
Loading Granulars for Ground Application (3b)	1.0 typical corn	80	1300	260	210	Target MOE reached at PPE		
	2.0 max corn	80	640	130	110	Target MOE reached at PPE		
	3.0 maximum ground rate (tobacco)	80	430	86	71	8600	860	780
Applicator Exposure								
Aerial (Spray) -- Enclosed Cockpit (4a)	2.0 orchards	350	No Open cockpit data available			100	150	60
	3.5 citrus (c)	100				200	290	120
Aerial (Granulars) -- Enclosed Cockpit (4b)	1.95 (f)	350	No Open cockpit data available			320	8	8
Groundboom Tractor (5)	1.5 predominant max	80	The biological monitoring results (Table A4) indicate that open cabs provide insufficient protection . Therefore, only the enclosed cab MOEs are presented.			580	1400	410
	5.0 tobacco max (d)	80				180	410	120
	4 Sodfarms (e)	80				220	510	150
	8.0 sodfarm fire ants	10				880	2000	610
Airblast Applicator (6)	2.0 predominant max	40	The biological monitoring results indicate that open cabs are insufficient.			230	190	110
	6.0 citrus	20				150	130	70
Tractor-Drawn Granular Spreader (7)	1.0 typical corn	80	1000	360	270	Target MOE reached at PPE		
	2.0 max corn	80	520	180	140	Target MOE reached at PPE		

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre) (a)	Daily Acres Treated (b)	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
	3.0 maximum ground rate (tobacco)	80	350	120	90	690	130	110
Seed Treatment (8)	No Data	No Data	No Data			No Data		
Dip Application (Preplant Peaches) (9)	No Data	No Data	No Data			No Data		
Flagger Exposure								
Spray Applications (10)	2.0 predominant max	350	50	140	37	2300	1400	880
	3.5 citrus (c)	100	100	290	74	4500	2900	1800
Granular Applications (11)	1.95	350	320	340	170	Target MOE reached at PPE		
Mixer/Loader/Applicator Exposure								
Backpack Sprayer (12)	0.0417 lb ai/gal predominant max / 0.08 lb ai/gal bark beetle treatment / 0.03 lb ai/gal stump treatment	40 gal/day	130 / 68 / 180	700 / 360 / 970	110 / 58 / 150	Target MOE reached at PPE, except for the higher concentration for the beetle bark treatment		
	3.5 citrus bark	1 A/day	63	330	53	Not feasible		
	0.039 lb ai/gal /750 ft2	1,000 ft2	4200	22000	3500	Target MOE reached at PPE		
Low Pressure Handwand (13)	0.0417 lb ai/gal predominant max / 0.08 lb ai/gal bark beetle treatment / 0.03 lb ai/gal stump treatment	40 gal/day	570 / 300 / 790	700 / 360 / 970	310 / 160 / 440	Target MOE reached at PPE		
	3.5 citrus bark	1 A/day	270	330	150	Target MOE reached at PPE		

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre) (a)	Daily Acres Treated (b)	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
	0.039 lb ai/gal/ 750 ft2 animal prem.	1,000 ft2	18,000	22,000	10,000	Target MOE reached at PPE		
High Pressure Handwand (greenhouse uses) (14)	Min. 0.0033 lb ai/gal	1,000 gal/day	66	88	38	Not feasible		
	Max. 0.0066 lb ai/gal		33	44	19	Not feasible		
Hydraulic Hand-held Sprayer for Bark Treatment (15)	3.5 citrus bark	10	16	100	14	Not feasible		
	0.08 lb ai/gal bark beetle treatment	1,000 gal/day	14 / 7	88 / 44	12 / 6	Not Feasible		
	0.039 lb ai/gal /750 ft2 animal prem	10,000 ft2	2,200	13,000	1,900	Target MOE reached at PPE		
Dry Bulk Fertilizer Impregnation	1.0 lb ai / 200 lb fertilizer / acre	No Data	No Data			No Data		

(a) Application rates are the maximum labeled rates found on EPA Reg. Nos. 62719-38, -221, -245, -34; -79, -72, -166, -220, 34704-66 (Clean Crop Chlorpyrifos 4E -- sodfarm fire ant rate), 499-367 (499-367 is the only greenhouse label identified), and 10350-22 for animal premise treatments. **"Predominant max"** in this table refers to the most **frequently identified maximum** application rate found on the labels for the specific formulation and equipment type. Typical rates are also included to characterize the chlorpyrifos uses. Not all application rates are included for all crops, instead, a cross-section of rates are used to represent the uses of chlorpyrifos.
(b) Daily acres treated are based on EPA's estimates of acreage (or gallonage) that would be reasonably expected to be treated in a single day for each exposure scenario of concern. The sodfarm fire ant rate is restricted on the label for harvest only, therefore, this rate is limited to the amount of sod that may be harvested in a reasonable time frame. Using the limited data available, 10 acres treated per day are assumed to be the upper range.
(c) The application rates on the Lorsban 4E (EPA Reg. No. 62719-220) and 50W (EPA Reg. No. 62719-39 discontinued as of 1995 and sold as -221) labels indicate that for citrus at the 6.0 lb ai/A rate it is necessary to use 100 to 2,400 gallons per acre dilute spray. Therefore, this rate is not expected to be feasible for an aerial applicator. The label language should be clarified so that the 6.0 lb ai/A rate is for ground only. Additionally, citrus orchards are believed to be relatively small plots and 100 acres per day is assumed in the assessment for aerial applications.
(d) The 5.0 lb ai/A rate for mixing/loading or applying liquids by groundboom application on tobacco has been canceled.
(e) The 4.0 lb ai/A rate for mixing/loading or applying liquids by groundboom application to sodfarms has been reduced to 3.0 lb ai/A.
(f) The 1.95 lb ai/A rate for aerial mixing/loading or applying granulars has been reduced to a maximum of 1.0 lb ai/A.

Non-Agricultural Occupational Handlers

The following exposure scenarios (by number as presented in Table 11) result in combined dermal and inhalation MOEs below 100 with label-recommended PPE, and thus are of concern.

(3) Short-term groundboom applicators of liquids on golf courses at 1 lb. ai/A wearing baseline PPE
(5) Short- and intermediate-term applicators of a dust product for control of fire ants
(9) Long-term mixer/loader/applicators of pre-construction termiticide treatments wearing baseline PPE
(13) Intermediate-term aerial applicators and mixer/loaders of mosquito adulticides using engineering controls at 0.023 lbs. ai/A

More detailed information on the non-agricultural occupational assessments can be found in the *Human Health Risk Assessment*, June 8, 2000, in the public docket and on the internet at www.epa.gov/pesticides/op.

Table 11. Risk Estimates for Non-Agricultural Occupational Handlers

Application Scenario	Clothing	Method of Evaluation	MOE			Risk Characterization/ Uncertainties
			Dermal	Inhalation	Total	
(3) Golf Course Use (Dursban Turf Insecticide; EPA Reg. 62719-35) (Short-term)						
Mixer/Loader (Liquid)	LS, LP, gloves	PHED V1.1	418	165	118	Central tendency estimate. Assumes handling product to treat 40 acres at lb ai/acre. The Agency has more confidence in the biomonitoring results than PHED.
Mixer/Loader (Wettable Powder in water soluble bags)	LS, LP, gloves	PHED V1.1	902	803	425	
Groundboom Applicator	LS, LP, no gloves	PHED V1.1	693	264	191	
		Biomonitoring (MRID 42974501)	69		69	
Mix/Load/Apply via Handgun (greens/tees) (Liquid)	LS, LP, gloves	PHED V1.1	209	594	155	Central tendency estimate. Assumes handling product to treat 5 acres at 1 lb ai/acre.
(5) Insecticidal Dust Product (Shaker Can or Bulbous Duster)						
Short-term	LS, LP, gloves	Scientific Literature Study	108 (7.9 g) 4.3 (198 g)	NE	108 (7.9 g) 4.3 (198 g)	Central-tendency short term risk assessments for 7.9 and 198 g ai; High-end intermediate-term risk estimates for 7.9 and 198 g ai (based on size of dust container); inhalation exposure not assessed due to an absence of data.

Application Scenario	Clothing	Method of Evaluation	MOE			Risk Characterization/ Uncertainties
			Dermal	Inhalation	Total	
Intermediate-term			22 (7.9 g) 0.9 (198 g)	NE	22 (7.9 g) 0.9 (198 g)	
(9) Pre-Construction Termiticide Treatment (0.5% chlorpyrifos as Dursban TC) (EPA Reg. 62719-47) (long-term)						
Mixer/Loader/ Applicator (3 hour average exposure)	label-specified PPE: single layer clothes and forearm-length chemically-resistant gloves (forearm length gloves not required by label)	Dosimetry and air monitoring from Registrant Study MRID No. 44589001	61	215	46	Low-end risk estimates for workers that wore double layer of clothing and forearm length gloves not required by the label; Central-tendency risk estimates for workers that wore a single layer of clothing and forearm length gloves; assumes 3 hour exposure, which could underestimate risks to workers exposed > 3 hrs/day, or that use 2% ai to treat utility poles or fences These MOEs have been adjusted to reflect the dilution rate of 0.5% ai for all termiticide products.
	double layer clothes (LS,LP, coveralls, rubber boots, and forearm-length gloves) (forearm-length gloves not required by label)		200	215	104	
(13) Mosquitocide Mixer/Loader/Applicator (PHED V1.1) (Short- and intermediate-term) (Mosquitomist One EPA Reg. 8329-24)						
Mixer/Loader--Aerial	PPE double layer clothes and gloves	PHED V1.1	132 (ST) 26 (IT)	58 (ST&IT)	40 (ST) 18 (IT)	High end risk estimates. Application rate of 0.023 lb ai/acre for 7500 acres
	Engineering Controls (enclosed cockpit) single layer clothes and gloves		260 (ST) 52 (IT)	833(ST&IT)	198 (ST) 49 (IT)	

Application Scenario	Clothing	Method of Evaluation	MOE			Risk Characterization/ Uncertainties
			Dermal	Inhalation	Total	
Mixer/Loader-- Ground-based fogger	PPE, single layer clothes and gloves		1111 (ST) 220 (IT)	663 (ST&IT)	415 (ST) 165 (IT)	High end risk estimates. Application rates of 0.005 and 0.01 lb ai//acre for 3000 acres. Surrogate ground-based fogger exposure data are not available, and therefore, it was necessary to extrapolate from airblast exposure data
	engineering controls (enclosed cab) and single layer clothes and gloves		297 (IT)	4760 (IT)	280 (IT)	
Aerial Applicator	engineering controls (enclosed cockpit) and single layer clothes and no gloves		440 (ST) 89 (IT)	2100 (ST&IT)	364 (ST) 85 (IT)	High end risk estimates. Application rate of 0.023/acre for 7500 acres
Ground-based fogger Applicator	engineering controls (enclosed cab) and single layer clothes and no gloves		671-1353 (ST)	1820-3640 (ST)	490-986 (ST)	High end risk estimates. Application rates of 0.005 and 0.01 lb ai/acre for 3000 acres. Surrogate ground-based fogger exposure data are not available, and therefore, it was necessary to extrapolate from airblast exposure data
			132-275 (IT)	1820-3640 (IT)	123-256 (IT)	

LS=Long sleeves; LP = Long pants; SS = short sleeves; SP = short pants
H20 = water; ST = short-term (1- 30 days); IT = intermediate term (30 days to 6 months) LT = long term (> 6 months)
NE = Not evaluated

3) Occupational Postapplication Exposure

Occupational postapplication exposure occurs when workers enter treated sites. In the agricultural setting, this includes scouts, pruners and harvesters, and may be of short- or intermediate-term duration. In the recreational setting, this includes golf course maintenance workers. Although a golf course maintenance worker may work up to 12 months per year, chlorpyrifos levels on turf will decline fairly rapidly, and so exposures are expected to be of short-term duration only. Postapplication activities are categorized as having low, medium and high potential for dermal contact.

Several chemical-specific postapplication exposure studies were conducted by the technical registrants and submitted to the Agency. These studies included biological monitoring, passive dosimetry and dislodgeable foliar residue (DFR) data. Data were submitted for sugar beets, cotton, sweet corn, almonds, pecans, apples, citrus, cauliflower, and tomatoes.

Specific transfer coefficients were also monitored and submitted for citrus harvesting, citrus tree pruning, cauliflower scouting, and tomato scouting. Transfer coefficients for other crops/activities have been submitted by the Agricultural Reentry Task Force (ARTF). In those scenarios where data have not been submitted, the Agency's standard values for transfer coefficients are used to estimate potential reentry exposure.

Chemical-specific DFR data are not available for many crops that are treated with chlorpyrifos. Therefore, the assessment of exposures for those crops is based on typical postapplication activities associated with representative crops, grouped according to their potential for dermal contact. Table 12 summarizes the crops and activities in terms of potential for dermal contact. Chemical-specific data are available for citrus, cauliflower, tree nuts and tree fruits, and these crops are assessed separately.

4) Occupational Postapplication Risk

For a detailed explanation of the preliminary occupational postapplication risk, refer to the *Agricultural and Occupational Exposure Assessment and Recommendations for the Reregistration Eligibility Decision Document for Chlorpyrifos*, dated June 19, 2000, which is available in the public document. In that preliminary risk assessment, restricted entry intervals (REIs) were calculated using default assumptions for transfer coefficients (Tc). Since that time, new exposure data for some activities have been submitted by the ARTF. The REIs have been recalculated using the new data for particular activities and are shown below in Table 12.

Table 12. Restricted Entry Intervals Based on Data Submitted by ARTF

Crop	Current REI	Proposed REI	Activity	PHI	MOE
Citrus Trees	5 days	5 days	Pruning during wet conditions	21 days	220
Fruit Trees	4 days	4 days	Thinning	28 days	280
Cauliflower	10 days	3 days	Using Tc for scouting, weeding, irrigating or hoeing	21 days	150
Nut Trees	2 days	24 hours	New Tc for pruning or thinning	14 days	270
Potatoes	2 days	24 hours	New Tc for irrigation or scouting	7 days	750
All Other Crops	24 hours	24 hours	Scouting, harvesting	7 days	110

Postapplication risks to golf course workers during mow/maintenance activities are presented in Table 13. The short-term MOEs are above 100 (MOE 110 to 210) and therefore are not of concern. These risk estimates assume contact with golf course turf on the day of treatment.

Table 13. Short-term Postapplication Risks to Workers in Mow/Maintenance Activities after Chlorpyrifos Treatment at 4 lbs. ai/A

Transfer Coefficient	DAT	Short-term MOE
500 cm2/hour	0	210
1000 cm2/hour	0	110

Postapplication risks to greenhouse/nursery workers were not assessed due to a lack of data. Information is needed concerning the timing of the applications in relation to the postapplication activities and a lack of residue data (foliar and bark treatments) to assess the REIs for the ornamental/greenhouse uses. These risks are of concern for activities such as pruning, transplanting and burlap/balling. The National Agricultural Pesticide Impact Assessment Program (NAPIAP 1996) reports chlorpyrifos is widely used for a broad range of insect applications including wood-boring, foliage feeding, sucking and soil-borne pests. NAPIAP (1996) also reports that although chlorpyrifos use represents only 5% of the total lbs. ai used in greenhouse/nursery operations, it is used by 35% of the survey respondents. It is obvious that chlorpyrifos is an important chemical for the industry, especially as a tool for resistance management. With such reliance by an industry, it is important to collect additional use information, greenhouse DFR data, and biological monitoring data to develop transfer coefficients for various greenhouse/nursery activities.

c. Residential Exposure and Risk

1) Residential Handler Exposure and Risk

Containerized baits in child-resistant packaging is the only residential use which may be applied by the homeowner. This use is not expected to result in exposures of concern. For further details, refer to the *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000, which is available in the public docket and on the internet at www.epa.gov/pesticides/op.

2) Residential Postapplication Exposure

Residential postapplication exposure occurs when people enter a treated golf course or following an application for mosquito control by a public agency. Residential postapplication exposures are expected to be of short-term duration (one day to one month).

Environmental concentrations of chlorpyrifos in homes may also result from spray drift, track-in, or from redistribution of residues brought home on the clothing of farm workers or pesticide applicators. The Agency is currently developing standard methodologies and guidance to evaluate these exposures. Modifications to EPA's assessment will be incorporated as that guidance becomes available.

3) Residential Postapplication Risk

No residential postapplication exposures pose risks of concern. A summary of the risk estimates, method of evaluation, and risk characterization/uncertainties is presented in Table 14. For residential postapplication risk, the target MOE is 1000. For golfers on a course treated at a rate of 1 lb. ai/A, MOEs are1500-2400. Following aerial and ground-based fogger mosquito adulticide use, MOEs are 17,000 and 29,000 for children and adults, respectively.

Table 14. Postapplication Risk Estimates to Residents/Recreational Users

Reentry Scenario	Method of Evaluation	Central-tendency MOE		Risk Characterization/ Uncertainties
		Adult	Child	
(8) Golf Course Treatment (Dursban Turf Insecticide; EPA Reg 62719-35) (1 lb ai/acre) (Short-term)				
Adolescent Golfer (12 yrs; 44kg)	Residential SOPs and surrogate residue data from flurprimidol study the day of treatment	1500 (1 lb ai/acre)		High-end risk estimates. Assumes exclusively dermal exposure the day of turf treatment Assumes a 4 hour exposure for an 18-hole round of golf.
Adult Golfer		2400 (1 lb ai/acre)		
(9) Aerial and Ground-Based Fogger Mosquitocide Application (Mosquitomist One, EPA Reg. 8329-24) (0.01 lb ai/acre) (Short-term)				
Dermal	Literature studies, the AgDrift Model and the updated Residential SOPs	42,000	26,000	High-end risk estimates based on the updated Residential SOPs. Assumes long-term inhalation exposure is negligible based on low application rate and infinite dilution.
Oral (hand to mouth)		NE	13,000	
Oral (Turfgrass Ingestion)		NE	54,000	
Oral (Soil Ingestion)		NE	20,000,000	
Total Exposure		42,000	15,000	

4) Incidents

Prior to implementation of the mitigation established in June 2000, chlorpyrifos was one of the most widely used insecticides in the home both by consumers and PCOs or exterminators. In a 1990 EPA-sponsored survey of pesticide use in households, chlorpyrifos was the fourth most commonly used insecticide, present in 18% of all households. A 1993 EPA survey of PCOs found it was the number one insecticide in use and accounted for a quarter of the poundage used in residential settings. Consequently, there have been many reports of human exposure and poisonings due to the widespread use of chlorpyrifos. The Agency estimates that approximately 98% of chlorpyrifos exposures discussed in the incident reports were associated with products removed as a result of the mitigation contained in the June 8, 2000 agreement. Human and pet poisoning incidents associated with chlorpyrifos exposure are discussed in greater detail in the *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000, which is available in the public docket and on the internet at www.epa.gov/pesticides/op.

4. Aggregate Risk

An aggregate risk assessment combines risk from dietary exposure (food and drinking water routes) and residential exposure (homeowner handler and postapplication exposures, including incidental oral exposure for toddlers who put grass in their mouths following mosquito adulticide use and exposure to treated golf course turf). As noted previously, this aggregate assessment reflects the mitigation that reduced potential chlorpyrifos exposures from food (elimination of use on tomatoes and limitations on the apple and grape uses) and in the residential/recreational environment. Acute, short-term and chronic aggregate assessments were conducted. For this assessment, the target MOE is 1000. Results of the aggregate risk assessment are summarized in here, and are discussed extensively in the *Human Health Risk Assessment for Chlorpyrifos*, June 8, 2000.

a. Acute Aggregate Risk

The acute aggregate risk assessment for chlorpyrifos addresses exposure from food and drinking water. For the highly refined acute probabilistic dietary exposure analysis, PDP, FDA and NFS monitoring data were used to the greatest extent possible, along with field trial data, and cooking and processing factors to assess dietary exposures. This aggregate assessment incorporates the mitigation measures agreed to in June 2000 (i.e., reduction of apple tolerance to 0.01 ppm to reflect dormant application, reduction of grape tolerance to 0.01 ppm based on domestic use pattern, cancellation of use on tomatoes and revocation of the tolerance on tomatoes).

With the apple, grape and tomato mitigation measures in place, the acute dietary risk estimates range from 4.1% to 82% of the aPAD, with children 1-6 years old being the most highly exposed population subgroup. Thus, the mitigated acute dietary (food only) risk estimate for chlorpyrifos exposure is not of concern. Acute estimated concentrations of chlorpyrifos in groundwater, derived from a conservative screening-level model, range from 0.007 to 0.103 ppb.

The acute surface water EECs, taken from monitoring data, range from 0.026 to 0.4 ppb. As indicated in Table 15 below, the EECs are below the DWLOCs for all populations. Thus acute food and drinking water exposures (except possible well contamination) are not of concern. It should be noted that neither the SCI-GROW model nor the monitoring data reflect concentrations after dilution (from source to treatment to tap) or drinking water treatment.

Table 15. Acute Aggregate Risk from Chlorpyrifos Including Risk Mitigation[a]

Population Subgroup (b)	Acute PAD (μg/kg/day)	Food Exposure 99.9th (μg/kg/day) (c)	Max. Water Exposure (μg/kg/day) (d)	Surface Water EEC (ppb)	Ground Water EEC (ppb)	Acute DWLOC (ppb) (e,f, g)
U.S. Population	5	0.237	4.76	0.026-0.4	0.007-0.103	166
All Infants (< 1 Year)	0.5	0.258	0.242			2.4
Children (1-6 years)	0.5	0.410	0.09			0.9
Females (13-50 years)	0.5	0.201	0.299			9

(a) Reflects mitigation implemented in June 2000 eliminating use on tomatoes and limiting use on grapes and apples.
(b) In addition to the U.S. population (all seasons), the most highly exposed subgroup within each of the infants, children, female groups is listed.
(c) 99.9th percentile exposure. Values are from Table 3 in Human Health Risk Assessment for Chlorpyrifos, June 8, 2000 (and rounded).
(d) Maximum Water Exposure (μg/kg/day) = Acute PAD (μg/kg/day) - [Acute Food Exposure (μg/kg/day)].
(e) DWLOC (μg/L) = Maximum water exposure (μg/kg/day) x body wt (kg) ÷ water consumed daily (L/day)]
(f) Default body weights are: general U.S. population, 70 kg; adult females, 60 kg; and infants/children, 10 kg.
(g) Default daily drinking water rates are 2 L/day for adults and 1 L/day for children.

b. Short-Term Aggregate Risk

The short-term aggregate risk estimate includes chronic dietary (food and water) exposure and short-term non-occupational (i.e., residential/recreational uses) exposures from chlorpyrifos use. As noted previously, this aggregate assessment reflects the mitigation that reduced potential chlorpyrifos exposures from food (apples, grapes and tomatoes) and in the residential/ recreational environment. This assessment evaluates potential exposures to treated golf courses and as a result of mosquitocide treatment by public agencies.

Table 16 presents the aggregate exposure estimates for chlorpyrifos from dietary and residential/non-occupational uses (golfing and mosquito abatement). Children 1-6 years old were assumed to be exposed to residues on turf following ground-based fogger applications of a mosquitocide and food residues. Children 7-12 years were assumed to be dermally exposed to chlorpyrifos residues while playing golf on the day of treatment, and to ingest food residues. Female residents were assumed to be concurrently exposed to turf following mosquito abatement, golfing (dermal contact with turf on the day of treatment), and food residues.

As shown in Table 16, aggregate MOEs are greater than 1000 for all subpopulations and are not of concern. Therefore, short-term DWLOCs were estimated to account for potential drinking water exposures.

Table 16. Short-Term Aggregate Exposure
[Chronic Dietary (Excluding Water) and Short-Term Residential Use]
Including Risk Mitigation[a]

Population Subgroup	Chronic Dietary Exposure	Short-Term Residential/Recreational Exposure (μg/kg/day)/ MOE Including Risk Mitigation			Total Aggregate MOE (c)
		Mosquitocide Exposure		Golf Course Exposure	Dietary & Residential Exposure
	Food (μg/kg BW/day) (b) / MOE	Oral (μg/kg BW/day) / MOE	Dermal (μg/kg BW/day) / MOE	Dermal (μg/kg BW/day) / MOE	Oral and Dermal MOE
Children (1-6 years)	0.008 MOE = 62,500	0.013 MOE = 38,500	0.19 MOE = 26,000	NE	12,000
Children (7-12 years)	0.015 MOE = 33,000	NE	NE	3.4 MOE = 1,500	1,400
Females 13-50	0.006 MOE = 83,000	NE	0.14 (d) MOE= 36,000	2.45 (d) MOE = 2,000	1,900

(a) Reflects mitigation implemented in June 2000 eliminating use on tomatoes and limiting use on grapes and apples.
(b) MOE calculated based on acute oral NOAEL of 500 μg/kg/day, and short-term dermal NOAEL of 5000 μg/kg/day.
(c) Oral and dermal exposures were combined because the oral and dermal endpoints are both based on plasma and RBC ChE inhibition.
(d) Adjusted from 70 kg to 60 kg for aggregate exposure.
NE = Not evaluated.

The short-term DWLOC values are presented in Table 17. The EECs for chronic exposures are below the DWLOCs for all populations. Thus, potential short-term aggregate exposure to chlorpyrifos resulting from food, water, golf course and mosquito abatement exposures are not of concern. This analysis is conservative because the Agency assumed that there could be concurrent residential and recreational exposures to chlorpyrifos (i.e., golfing and mosquito abatement on the same day). In addition, neither SCI-GROW nor the monitoring data reflect concentrations after dilution (from source to treatment to tap) or drinking water treatment.

Table 17. Short-term Aggregate Exposure DWLOCs (Chronic Dietary and Short-Term Residential Use) Including Risk Mitigation[a]

Population Subgroup (b)	Acute Oral NOAEL (μg/kg/day)	ST Food and Residential MOE (b)	Water MOE (c)	Max. Water Exposure (μg/kg/ day) (d)	Surface Water (ppb)	Ground Water (ppb)	ST DWLOC (ppb) (e,f,g)
Children (1-6 years)	500	12,000	1,090	0.4587	0.026	0.007-0.103	4.5
Children (7-12 years)		1,400	3,450	0.14			1.4
Females (13-50 years)		1,900	2,100	0.238			7.1

(a) Reflects mitigation implemented in June 2000 eliminating use on tomatoes and limiting use on grapes and apples.
(b) Values are from Table 16.
(c) MOE_{WATER} = 1 / [(1/MOE_{AGG} - [1/MOE_{FOOD} + 1/MOE_{DERMAL} + 1/MOE_{ORAL}]), where MOE_{AGG} is 1000.
(d) Maximum Water Exposure (μg/kg/day) = Acute NOAEL of 500 (μg/kg/day)÷ MOE_{WATER}
(e) DWLOC (ppb) = Maximum water exposure (μg/kg/day) x body wt (kg) ÷ water consumed daily (L/day)]
(f) EPA default body weights are: adult females, 60 kg; and infants/children, 10 kg.
(g) EPA default daily drinking water rates are 2 L/day for adults and 1 L/day for children.
ST = short-term

c. Intermediate-Term Aggregate Risk

No residential/recreational uses result in exclusively intermediate-term exposures (i.e., greater than 30 days but less than 6 months). Therefore, an intermediate-term aggregate risk assessment was not conducted.

d. Chronic Aggregate Risk

The chronic aggregate risk assessment for chlorpyrifos addresses exposures from food and drinking water. For the highly refined chronic dietary exposure analysis, PDP, FDA and NFS monitoring data were used to the greatest extent possible, along with field trial data, and cooking and processing factors. This aggregate assessment incorporates the mitigation agreed to in June 2000 (limitation of use the use on apples and grapes and deletion of use on tomatoes), and assumes there are no chronic exposures from termiticide treatments, since these uses are being phased down.

The chlorpyrifos chronic dietary (food only) risk estimates range from 2.5 to 51% of the cPAD, with children 1-6 years old being the most highly exposed population subgroup. Thus, the chronic dietary (food) risk from chlorpyrifos exposure is not of concern.

Chronic groundwater EECs, derived from SCI-GROW, range from 0.007 to 0.103 ppb. Chronic surface water EECs, based on monitoring data, are estimated at 0.026 ppb. The chronic

DWLOC values are shown below in Table 18. For all subpopulations, surface and groundwater EECs are below the DWLOCs and therefore are not of concern. These estimates are conservative because neither the SCIGROW model nor the monitoring data reflect actual drinking water concentrations after dilution (from source to tap) or drinking water treatment.

Table 18. Chronic Aggregate Exposure DWLOCs Including Mitigation [a]

Population Subgroup (b)	Chronic PAD (μg/kg/day)	Chronic Food Exposure (μg/kg/day)(c)	Max. Water Exposure (μg/kg/day) (d)	Surface Water (ppb)	Ground Water (ppb)	Chronic DWLOC (ppb) (e,f,g)
U.S. Population	0.3	0.008	0.292	0.026	0.007 to 0.103	10
All Infants (< 1 Year)	0.03	0.01	0.02			0.2
Children (1-6 years)	0.03	0.015	0.015			0.15
Females (13-50 years)	0.03	0.006	0.024			0.72

(a) Reflects mitigation implemented in June 2000 eliminating use on tomatoes and limiting use on grapes and apples.
(b) In addition to the U.S. population (all seasons), the most highly exposed subgroup within each of the infants, children, female groups is listed.
(c) Values are from Table 4 from the Human Health Risk Assessment, June 8, 2000 (and rounded).
(d) Maximum Water Exposure (μg/kg/day) = Chronic PAD (μg/kg/day) - [Chronic Food Exposure + Chronic Residential Exposure (μg/kg/day) (if applicable)]. Chronic residential uses were not considered based on mitigation options.
(e) DWLOC (ppb) = Maximum water exposure (μg/kg/day) x body wt (kg) ÷ water consumed daily(L/day)]
(f) HED default body weights are: general U.S. population, 70 kg; adult females, 60 kg; and infants/children, 10 kg.
(g) HED default daily drinking water rates are 2 L/day for adults and 1 L/day for children.

B. Environmental Risk Assessment

A summary of the Agency's environmental risk assessment is presented below. For detailed discussions of all aspects of the environmental risk assessment, see the *Fate and Environmental Risk Assessment*, dated October 1999 and revised March and June 2000, available in the public docket and on the internet at www.epa.gov/pesticides/op.

1. Environmental Fate and Transport

The environmental fate database for chlorpyrifos is largely complete. The major route of dissipation appears to be aerobic and anaerobic metabolism. Abiotic hydrolysis, photodegradation and volatilization do not seem to play significant roles in the dissipation process. Based on available data, chlorpyrifos appears to degrade slowly in soil under both aerobic and anaerobic conditions. Information on leaching and adsorption/desorption indicate that parent chlorpyrifos is largely immobile. The environmental fate of the major chlorpyrifos degradate, TCP, indicates that it is mobile in soils and persistent in soils when not exposed to

light. Available field data indicate that chlorpyrifos has a half-life in the field of less than 60 days, with little or no leaching observed. Because of its low water solubility and high soil binding capacity, there is potential for chlorpyrifos sorbed to soil to run off into surface water via erosion. Chlorpyrifos has been detected in fish tissues. Chlorpyrifos residues in aquatic species may result in dietary exposure for aquatic birds and mammals feeding on aquatic organisms. Chlorpyrifos rapidly depurates from fish when aquatic chlorpyrifos exposures cease.

The degradate TCP appears to be more persistent than chlorpyrifos (substantial amounts remain 365 days after application) and it exhibits much lower soil/water partitioning than chlorpyrifos. Consequently, substantial amounts of TCP are probably available for runoff for longer periods than chlorpyrifos. The relatively low soil/water partitioning of TCP indicates that its concentrations in sediment and water are probably comparable, and that runoff occurs primarily by dissolution in runoff water rather than by adsorption to eroding soil. The low soil/water partitioning of TCP suggests that its bioaccumulation potential is probably low.

Chlorpyrifos can contaminate surface water via spray drift at the time of application or as runoff up to several months after application. Available data indicate that most chlorpyrifos runoff is generally via adsorption to eroding soil rather than by dissolution in runoff water. However, under some conditions, dissolution in runoff water may be significant.

2. Ecological Risks

Risk characterization integrates the results of the exposure and ecotoxicity data to evaluate the likelihood of adverse ecological effects. The means of integrating the results of exposure and ecotoxicity data is called the quotient method. For this method, risk quotients (RQs) are calculated by dividing exposure estimates by ecotoxicity values, both acute and chronic.

RQ = Exposure/Toxicity

RQs are then compared to EPA's levels of concern (LOCs). The LOCs are criteria used by OPP to indicate potential risk to nontarget organisms. The criteria indicate that a pesticide used as directed has the potential to cause adverse effects on nontarget organisms.

Ecotoxicity endpoints derived from the results of short-term laboratory studies that assess acute effects are: (1) LC_{50} (fish and birds) (2) LD_{50} (birds and mammals) (3) EC_{50} (aquatic plants and aquatic invertebrates) and (4) EC_{25} (terrestrial plants). Endpoints derived from the results of long-term laboratory studies that assess chronic effects are NOAEL and LOAEL for birds and mammals and NOAEC and LOAEC for fish and aquatic invertebrates.

Risk presumptions along with the corresponding RQs and LOCs are shown below in Table 19.

Table 19. Risk Presumptions for Non-target Organisms

Terrestrial Animals		
Risk Presumption	**RQ**	**LOC**
Acute High Risk	EEC/LC_{50} or LD_{50}/sqft^2 or LD_{50}/day^3	0.5
Acute Restricted Use	EEC/LC_{50} or LD_{50}/sqft^2 or LD_{50}/day (or LD_{50} < 50 mg/kg)	0.2
Acute Endangered Species	EEC/LC_{50} or LD_{50}/sqft^2 or LD_{50}/day	0.1
Chronic Risk	EEC/NOAEL	1
Aquatic Animals		
Acute High Risk	EEC/LC_{50} or EC_{50}	0.5
Acute Restricted Use	EEC/LC_{50} or EC_{50}	0.1
Acute Endangered Species	EEC/LC_{50} or EC_{50}	0.05
Chronic Risk	EEC/NOAEC	1
Terrestrial and Semi-Aquatic Plants		
Acute High Risk	EEC/EC_{25}	1
Acute Endangered Species	EEC/EC_{50} or NOAEC	1
Aquatic Plants		
Acute High Risk	EEC/EC_{50}	1
Acute Endangered Species	EEC/EC_{50} or NOAEC	1

Calculated risk quotients represent a screening level assessment. Risk characterization provides further information on the likelihood of adverse effects occurring by considering the fate of the chemical in the environment, geographic patterns of chemical usage, communities and species potentially at risk, their spatial and temporal distributions and the nature of the effects observed in the studies.

a. Exposure Assumptions

Three types of terrestrial wildlife risk assessments were conducted. For non-granular pesticides, acute and chronic dietary exposures were assessed by comparing estimated environmental concentrations on food items to LC_{50} values. To assess risks from granular products, acute exposures are expressed as LD_{50} per square foot. Acute risk quotients for

granular formulations were calculated by dividing the maximum milligrams of chlorpyrifos exposed on the soil surface per square foot by LD_{50} values of various wildlife species times the animal's body weight.

For non-granular (liquid and dust) pesticides, the estimated environmental concentrations (EECs) were compared with LC_{50} values to assess risk. Maximum EECs were used to derive a conservative estimate of risk to wildlife that may feed on foods with higher than average residues. This risk assessment estimated risks to birds and mammals feeding on short grass or foliage and fruits, seeds, and large and small insects, which provides a range of risk quotients depending on the particular dietary needs of a wildlife species. The assessment assumes that animals would consume only chlorpyrifos- treated food items. Measured residue levels reported in three field studies on corn, citrus and golf courses sprayed with chlorpyrifos support the use of maximum residue levels for risk assessment. In case of soil incorporation following spray applications, it is assumed that soil incorporation reduces the amount of treated vegetation and seeds available to wildlife on the surface, but soil incorporation does not reduce the pesticide concentration on these food items. Soil incorporation reduces the amount of pesticide available for runoff.

Estimated environmental concentrations in aquatic systems were modeled using GENEEC and PRZM-EXAMS to reflect use on corn, citrus, peanuts, cotton and tobacco. Use patterns for these sites reflect the range of application rates, frequency of application, maximum seasonal limits and application methods for chlorpyrifos. Estimated concentrations derived from the models were used to assess acute and chronic risks to freshwater and estuarine organisms in ponds and estuarine areas, respectively. Concentrations reported in NAWQA and California monitoring data were used to assess risks for some typical flowing waters. Acute risks were assessed using peak EECs. Chronic risk quotients were calculated using an exposure period ranging from 96 hours to 21 days. For greater detail on exposure assumptions, see the *Fate and Environmental Risk Assessment*, revised June 2000.

b. Toxicity

Extensive acute and chronic toxicity data are available for chlorpyrifos. A summary of toxicity values used in terrestrial risk assessments is shown below in Table 20.

Table 20. Summary of Terrestrial Toxicity Values Used In Risk Assessment for Chlorpyrifos

Toxicity Category	Most Sensitive Species	Toxicity Value	Derived Toxicity Values	
			Herbivores and Insectivores	Granivores
Mammalian Acute LD_{50}	Rat	97 mg/kg	15 gr. 102 ppm 35 gr. 147 ppm 1000 gr. 647 ppm	15 gr. 462 ppm 35 gr. 647 ppm 1000 gr. 3233 ppm
Mammalian Dietary LC_{50}	Rat	1330 ppm	N/A	
Mammalian Reproduction NOAEL	Rat	10 ppm	N/A	
Avian Acute LD_{50}	House Sparrow	10 mg/kg	N/A	
Avian Dietary LC_{50}	Mallard Duck	136 ppm	N/A	
Avian Reproductive NOAEL	Mallard Duck	25 ppm	N/A	

Aquatic toxicity studies indicate that chlorpyrifos is moderately to very highly toxic to both fish and aquatic invertebrates. TCP was found to be much less toxic than chlorpyrifos. Aquatic toxicity values for chlorpyrifos are shown below in Table 21.

Table 21. Summary of Aquatic Toxicity Values

Toxicity Category		**Toxicity Value**
Freshwater Fish	Acute LC_{50}	1.8 ppb (bluegill sunfish)
		595 ppb (mosquitofish)
	Reproductive NOAEC	0.57 ppb (fathead minnow)
Estuarine Fish	Acute LC_{50}	0.96 ppb
	Reproductive NOAEC	0.28 ppb (Atlantic silverside)
Freshwater Invertebrate	Acute LC_{50}	0.1 ppb (*Daphnia magna*)

Toxicity Category		Toxicity Value
		50 ppb (stonefly *P. californica*)
	Reproductive NOAEC	0.04 ppb (*Daphnia magna*)
Estuarine Invertebrate	Acute LC_{50}	0.035 ppb (Mysid shrimp)
		2000 ppb (Oyster embryo-larvae)
	Reproductive NOAEC	<0.0046 ppb (Mysid shrimp)
Estuarine Algae	Acute LC_{50}	140-300 ppb (*S. costatum*)

c. Summary of Risks to Nontarget Organisms

The Agency calculated risk quotients for most agricultural and some non-crop uses such as golf courses and perimeter treatments for termites. Risk quotients have been estimated based on maximum use rates and maximum seasonal poundage permitted by the label for both acute and chronic exposures. In addition, typical use rates were assessed for selected major crops. The chronic exposure values for assessing risks to avian and mammalian reproduction have been modified since completion of the *Fate and Environmental Risk Assessment*, June 2000, to reflect mean residue levels on grasses, foliage, seeds and insects. Risk quotients for major use sites are presented in this document. For detailed discussion of these and risk quotients for other uses, see the *Fate and Environmental Risk Assessment*, June 2000, which is available in the public docket and on the internet at www.epa/gov/pesticides/op.

Risk quotients indicate that a single application of chlorpyrifos may pose high risks to small mammals, birds, fish and aquatic invertebrate species for nearly all registered outdoor uses. For multiple applications, EPA assumes that residues are additive and has used minimum retreatment intervals along with calculated half-lives, half-lives for soils, foliage and water. Multiple applications increase the risks to wildlife and prolong exposures to toxic concentrations. In most cases, acute risk quotients exceed 1 for the most sensitive small mammals and birds. All aquatic acute and reproductive risk quotients exceed 1; many aquatic risk quotients exceed 10 and 100; several risk quotients for estuarine invertebrates exceed 1,000. In a few cases at maximum application rates, chlorpyrifos may bioconcentrate in the tissues of fish and aquatic invertebrates to levels that exceed acute LC_{50} values for sensitive bird species and reproductive NOAELs for birds and small mammalian species. Hence bioconcentration of chlorpyrifos in ponds and estuarine areas may pose acute and/or reproductive risks to aquatic birds and mammals feeding adjacent to treated areas.

For aquatic risk assessments, the Agency used the screening-level model GENEEC to predict concentrations of chlorpyrifos in water following a single application. To estimate concentrations on a single site over multiple years, PRZM-EXAMS was used. Peak EECs range from 1 to 37 ppb. These EECs may be considered highly conservative because 1) the EECs generated by both models reflect agricultural uses with the highest application rates of chlorpyrifos, and 2) the EECs represent one in ten-year concentrations in a one-hectare, 2-meter

deep farm pond or other water body with no outlet draining 10 hectares, 100% of which is treated with chlorpyrifos. The aquatic risk quotients derived from these EECs are therefore conservative. In addition, the RQs for estuarine organisms are likely to be even more conservative than those for freshwater organisms. Concentrations in estuarine environments could be expected to be much lower than in a contained pond because of flushing and dispersion as a result of tidal fluctuations. RQs derived from GENEEC may also overestimate aquatic risks for crops with ground cover such as pome fruits and tree nuts.

Endangered species LOCs are exceeded for small mammals, birds, freshwater fish and invertebrates, and estuarine fish and invertebrates for most chlorpyrifos uses. The Fish and Wildlife Service has reviewed the use of 4 EC, 15 G, 50 W and Dursban 10 CR on numerous crops and as a mosquito larvicide. In several opinions, the most recent in 1993, FWS found jeopardy for a few bird and amphibian species, a snake, and many species of fish and aquatic invertebrates, under the conditions of use at the time of the opinion.

The Agency has consulted several times with the Fish and Wildlife Service (FWS) on the potential effects of chlorpyrifos for various uses on endangered and threatened species. To date, the FWS has issued five Biological Opinions. In these Opinions, the FWS found jeopardy for 35 fish species, 33 aquatic invertebrate species, 7 avian species, 4 amphibian species and 13 insect species. An additional 18 fish species, 2 aquatic invertebrate species, 1 avian species and 1 amphibian species were expected to be affected, but not jeopardized. These consultations and the findings expressed in the Opinions, however, are based on old labels and application methods, less refined risk assessment procedures, and an older approach to consultation which is currently being revised through interagency collaboration.

EPA's current assessment of ecological risks uses both more refined methods to define ecological risks of pesticides and new data, such as that for spray drift. Therefore, the Reasonable and Prudent Measures (RPMs) in the Biological Opinion(s) may need to be reassessed and modified based on these new approaches.

The Agency is currently engaged in a Proactive Conservation Review with FWS and the National Marine Fisheries Service under section 7(a)(1) of the Endangered Species Act. The objective of this review is to clarify and develop consistent processes for endangered species risk assessments and consultations. Subsequent to the completion of this process, the Agency will reassess the potential effects of the remaining chlorpyrifos uses to federally listed threatened and endangered species. At that time, the Agency will also consider any regulatory changes recommended in this IRED that are being implemented. Until such time as this analysis is completed, the overall environmental effects mitigation strategy articulated in this document and the County Specific Pamphlets described below, will serve as interim protection measures to reduce the likelihood that endangered and threatened species may be exposed to chlorpyrifos at levels of concern.

1) Risks to Terrestrial Mammals

Risk quotients for both maximum and typical use rates exceed the levels of concern for small mammalian herbivores and insectivores for most crop and non-crop uses of chlorpyrifos. The high risk LOC (0.5) for the mammalian acute oral LD_{50} values is usually exceeded for 15 gram mammals, frequently exceeded for 35 gram mammals and occasionally exceeded for 1000 gram mammals. The high risk LOC (0.5) for mammalian subacute dietary LC_{50} is rarely exceeded, but the restricted use LOC (0.2) is exceeded frequently. The LOC for reproductive effects (1.0) is usually exceeded.

2) Risks to Terrestrial Birds and Reptiles

Risk quotients for both maximum and typical application rates for spray uses usually exceed the levels of concern for high risks (0.5) for subacute LC_{50}s and (1.0) for reproduction NOAEL for avian species. Risk quotients for both maximum and typical application rates for granulars usually exceed the LOC for high acute risk. Several incidents with robins and other bird species reported for lawn and residential perimeter treatments for termites support these risk quotients for birds and reptiles.

Sensitivity of reptiles to pesticides is assumed to be similar or less than for birds, hence the avian risk quotients apply to reptiles as well. Some snake carcasses tested positive for chlorpyrifos in two of the three field studies. The presence of chlorpyrifos in snake carcasses suggests the possibility of secondary toxicity, that is, effects caused by a chemical present in the carcass of an animal eaten by a predator.

3) Risks to Bees and Beneficial Insects

Chlorpyrifos is highly acutely toxic to honey bees and applications would be expected to pose a risk to bees and beneficial insects present in the treated area during application. At present, there is no accepted method to determine risk quotients based on the bee acute contact toxicity data. Results from some field studies confirm predicted risks to bees, which are killed if present during application and for as long as 24 hours after treatment.

4) Risks to Fish and Amphibians

Risk quotients exceed the LOC for high acute (0.5) and chronic (1.0) effects for freshwater and estuarine fish for all uses. Reproductive risks to fish populations are indicated by risk quotients which are greater than 21-day EECs for all uses. Freshwater fish reproductive effects seen in the fathead minnow include reduced survival at 1.09 ppb; for estuarine fish, reproductive effects include reduced survival and body weight at 0.28 ppb. Fish reproductive effects are likely to be greater than indicated by RQ values presented in risk quotient tables for all chlorpyrifos uses. The fathead minnow tested in the full life-cycle study is less sensitive on an acute basis than other species, such as bluegill and trout. Thus the RQs for more sensitive fish would be expected to be greater than for the fathead minnow.

5) Risks to Aquatic Invertebrates

Risk quotients for all uses exceed the acute and chronic LOCs for freshwater and estuarine invertebrates. For 14 major crop uses, eight of the fourteen peak EECs exceed the EC_{50}/LC_{50} values for three of the four freshwater species. In the estuarine/marine invertebrate life cycle toxicity study using mysid shrimp, reproductive effects were seen at 0.0046 ppb, the lowest dose tested. Effects observed were a reduced number of young and reduced mean number of young per female.

6) Risks to Freshwater Organisms in Field Monitoring Studies

In an Iowa corn field study, chlorpyrifos was applied as an emulsifiable concentrate to four fields (4 applications per field, 1.5-3 lbs. ai/A) and as a granular formulation to four fields (3 applications per field, 1-2.6 lbs. ai/A). Chlorpyrifos levels were measured in aquatic areas adjacent to the treated fields. The mean residue level of 66.9 ppb exceeds all predicted EECs. After granular treatment to corn at 2 lbs. ai/A, one water sample had residue level of 1.80 ppb seven days after the tassel broadcast treatment. This concentration is below predicted EECs ranging from 5.5 to 8.6 ppb.

In a California citrus field study, two orange groves were sprayed by airblast, and chlorpyrifos concentrations measured in soil, crop and non-crop foliage, invertebrates and water adjacent to the groves. Modeled EECs were generally comparable to measured concentrations. Measured chlorpyrifos levels in water ranged from 1.041 to 486 ppb, depending upon the application scenario. More detailed information can be found in the *Environmental Fate and Effects Assessment*, June 2000. Dead fish and other aquatic vertebrates were found in ponds adjacent to treated groves on several occasions.

A field study in Florida measured chlorpyrifos levels after two applications to golf course turf at 4 lbs. ai/A, with a 21-day interval between applications. Applications were made using both granular and liquid sprays. For areas treated with the liquid formulation, measured initial mean concentrations in water were <1.0 ppb (non-detect). The predicted Tier I EEC was 14.75 ppb, and the Tier II EEC was 29.03 ppb. For the granular formulation, the measured initial mean concentrations were <1.0 ppb (non-detect) and 0.905 ppb. The predicted Tier I EECs were 13.28 ppb; the Tier II EEC was 25.31 ppb. Thus, measured chlorpyrifos concentrations were below modeled estimates.

Monitoring results from the early 1990s indicate widespread and persistent occurrence of chlorpyrifos in aquatic areas throughout the nation. In a national fish monitoring study approximately 23 percent of the fish nationwide had measurable levels of chlorpyrifos residues (EPA 1992). Chlorpyrifos was detected at levels up to 59 ppb in mussels in coastal California, and in concentrations of 245 ppb in sediments in Massachusetts (NOAA, 1992). The Agency's Storet database reports measurable chlorpyrifos levels in biota in 12 states and in one water sample. It is uncertain whether the chlorpyrifos levels in aquatic organism tissues are sufficient to adversely affect exposed organisms.

Chlorpyrifos was detected in storm water runoff in the San Francisco Bay area in 1994-1995 at levels that exceed the California Department of Fish and Game water quality criterion of 15 ng/L (pptr). Approximately 80 percent of the samples collected from Sacramento and Stockton exceeded the water quality criterion. In the San Francisco Bay area, approximately 75 percent of the samples collected exceeded the water quality criterion. Rainfall samples also collected in the San Francisco area contained chlorpyrifos at levels toxic to *Ceriodaphnia*.

7) Risks to Piscivorous Birds and Mammals from Bioconcentration of Chlorpyrifos in the Food Chain

At high application rates, chlorpyrifos levels in fish and aquatic invertebrates could exceed the avian subacute dietary toxicity value (136 ppm) and reproductive NOAELs for birds (25 ppm) and mammals (10 ppm).

8) Risks to Nontarget Plants

Plant toxicity studies are not currently required for insecticides. However, chlorpyrifos toxicity data are available for one out of five recommended aquatic plant species. Based on toxicity values for three estuarine algal species (only one recommended species), risk quotients for the highest exposures do not exceed any level of concern. However, the EC_{50} for all three algal species were exceeded by measured chlorpyrifos levels in some water samples found in the citrus field study.

3. Risk Characterization of TCP

A full set of acute studies has been submitted using TCP as the test substance. Studies indicate that TCP's acute toxicity ranges from moderately toxic to practically non-toxic. TCP is less acutely toxic than chlorpyrifos, hence risks to fish and wildlife would appear to be reduced as chlorpyrifos degrades.

4. Risk Quotients for Major Use Sites

a. Corn

Corn is the largest use site for chlorpyrifos in terms of pounds of active ingredient applied per year. The Agency estimates that for the years 1987-1999, an average of approximately 5.5 million lbs. ai per year were applied to corn. Based on that usage data, chlorpyrifos was applied to approximately 7% of corn grown in the U.S. A typical application on corn is an at-plant granular treatment at 1.1 lbs. ai/A.

Wildlife utilization of corn fields is high with a broad diversity of avian and mammalian species. Wildlife reported to feed in corn fields include quail, grouse, partridge, pheasant, prairie chicken, ducks, doves, songbirds, red fox, muskrat, opossum, raccoon and deer. Bobwhite quail, pheasant and rabbits also nest and brood young in corn fields.

Applications of spray and granular formulations to corn result in risk quotients which indicate acute risks to small terrestrial mammals, birds and aquatic organisms, except estuarine algae. In a field study evaluating use on corn, forty-four carcasses collected in and around the treated site. Seven carcasses were analyzed for chlorpyrifos and three carcasses were found to contain residues of chlorpyrifos. The field study did not monitor for aquatic effects, but measured chlorpyrifos residues at a mean level of 66.9 ppb adjacent to treated fields.

A comparison of risk quotients for various application scenarios in Table 22 indicates that risks are lowest with the ground application. Approximately 98% of chlorpyrifos use on corn is by ground application. Risk quotients for aquatic species from a ground application are about 28% lower than for a single aerial application at the same application rate. Aquatic risks in shallow ponds (2 meters deep) will be greater than in deeper ponds (3 meters deep); risks are higher in standing waters, marshes and swamps than they are in shallow ponds.

Granular treatments to corn at pre-plant, at plant, at cultivation, whorl and tassel stages indicate high risks to many species from all four treatment scenarios. Risk quotients exceed the high risk LOCs for all wildlife categories, except mammals weighing 1,000 grams.

Table 22. Ranges of Risk Quotients for Chlorpyrifos Use on Corn

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuar-ine Fish	Estuarine Inverts.
Ground spray, preplant, 1 app.@ 3 lbs. ai/A, 2" soil incorporation	Acute	0.014-7.1	--	1.5	28	2.9	79
	Subacute	0.03-0.54	0.33 – 5.3	--	--	--	--
	Reproduction NOAEL/NOAEC	4.5-26	1.8-19	2.2-3.8	32-54	4.6-7.8	>280 - >470
Ground spray, postemergence/ foliar, 1 app. @ 1.5 lbs. ai/A	Acute	0.007-3.5	--	3.1	55	5.7	160
	Subacute	0.02-0.27	0.17-2.6	--	--	--	--
	Reproduction NOAEL/NOAEC	2.3-13	0.92-5	4.7-8.4	68-120	9.6-17	>590->1000
Aerial spray, postemergence/foliar, 1 app. @ 1.5 lbs. ai/A	Acute	0.007 - 3.5	--	4.3	77	8	220
	Subacute	0.017 - 0.27	0.17 - 2.6	--	--	--	–
	Reproduction NOAEL/NOAEC	2.3 - 36	0.92 - 14	6.7 - 12	95 - 170	14 - 24	> 830 > 1500
Ground spray, postemergence/ foliar, 3 apps. @ 1.5 lbs. ai/A, 14-day intervals	Acute	0.009-4.6	--	13	240	25	690
	Subacute	0.02-0.35	0.22-3.5	--	--	--	--
	Reproduction NOAEL/NOAEC	3-17	1.2-6.7	21-38	290-540	42-77	>2500->4700

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuar-ine Fish	Estuarine Inverts.
Aerial spray, postemergence/ foliar, 11 apps. @ 1 lb. ai/A, 3-day intervals	Acute	0.017-8.8	--	19	340	35	970
	Subacute	0.04-0.68	0.41-6.6	--	--	--	--
	Reproduction NOAEL/NOAEC	5.6-90	2.2-36	42-49	590 - 700	85-100	>5200 >6100
Granular, ground broadcast, preplant, 1 app. @ 1.1 lbs. ai/A, 4" soil incorporation (typical rate, modeled on Iowa soil)	Acute	0.018 - 1.1	6.1	0.54	9.8	1.0	28
	Reproduction NOAEL/NOAEC	--	–	0.77 - 1.4	11 - 19	1.6 - 2.8	>95 >167
Granular, ground broadcast, preplant, 1 app. @ 1.1 lbs. ai/A, 4" soil incorporation (typical rate, modeled on Mississippi soil)	Acute	0.018 - 1.1	6.1	1.5	27	2.8	77
	Reproduction NOAEL/NOAEC	--	–	2.3 - 3.9	32 - 55	4.6 - 7.9	>280 >480
Granular, ground broadcast, preplant, 1 app. @ 2 lbs. ai/A, 4" soil incorporation	Acute	0.032-2.1	11	0.92	17	1.7	47
	Reproduction NOAEL/NOAEC	NA[2]	--	1.4-2.5	20-36	2.9-5.1	>180 >310
Granular, at-plant, 7" band or T-band, 1 app. @ 1.8 oz/1000 row feet, 1" soil incorporation	Acute	0.13-8.5	46	3.7	66	6.9	190
	Reproduction NOAEL/NOAEC	--	–	5.9-10	84-140	12-21	>730 >1300
Granular, postemergence aerial broadcast, 2 apps. @ 0.975 ai/A, 14-day intervals, 50% interception by plant	Acute	0.05-3.3	18	3.5	64	6.6	180
	Reproduction NOAEL/NOAEC	--	–	5.4-9.6	78-140	11-20	>670 >1200

b. Cover Crops

Risk quotients for alfalfa, clover and grass grown for seed, mint and wheat are summarized in Table 23. Chlorpyrifos applications to these crops are largely limited to liquid formulations. Runoff from foliar applications to cover crops is expected to be lower than to crops grown on plowed or bare ground. The GENEEC and PRZM3-EXAMS Models estimate EECs for row crops, but data on runoff are unavailable to model EECs for vegetative ground cover. The degree to which ground cover reduces runoff and yields lower EECs is unknown.

Hence, the aquatic risk quotients in the following tables for these cover crops are higher than would actually be anticipated

Alfalfa is the major use site in this group. Alfalfa fields are heavily utilized by a diversity of avian and mammalian species. Ring-necked pheasants, grouses, partridges, quail, sandhill crane, ducks, geese, mourning dove, songbirds, rabbits, groundhogs, muskrats, deer and elk feed in alfalfa fields to a moderate to high degree. Many of the avian species also nest in alfalfa fields.

Table 23. Ranges of Risk Quotients for Chlorpyrifos Use on Cover Crops (Alfalfa, Clover and Grass Grown for Seed, Mint, Wheat)

Crop and Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuar-ine Fish	Estuarine Inverts.
Alfalfa, granular, at-plant, in-furrow, 1 app. @ 1 lb. ai/A, 4" soil incorporation	Acute	0.016-1.1	5.7	3.5	8.3	0.86	24
	Reproduction NOAEL/NOAEC	–	--	0.7-1.3	10-18	1.4-2.6	>87 >160
Alfalfa, aerial spray, postemergent/ foliar, 4 apps. @ 1 lb. ai/A, 42-day interval	Acute	0.005-2.4	--	10	180	19	510
	Subacute	0.011-0.18	0.11-1.8	--	--	--	--
	Reproduction NOAEL/NOAEC	1.5-8.5	0.6-3.4	15-28	220-400	31-57	>1900 >3500
Alfalfa, aerial spray, postemergence/ foliar, 1 app. @ 0.7 lbs. ai/A	Acute	0.003-1.6	--	2	36	3.7	100
	Subacute	0.008 - 0.13	0.08-1.2	--	--	--	--
	Reproduction NOAEL/NOAEC	1.1-6	0.42-2.4	3-5.5	52-78	6.1-11	>370 >680
Clover grown for seed, ground spray, preplant and foliar, 2 apps. @ 2 lbs. ai/A, 14-day interval	Acute	0.012-5.9	--	8.3	150	16	430
	Subacute	0.25 - 0.45	2.5-4.4	--	--	--	--
	Reproduction NOAEL/NOAEC	8.8 - 21	3.6 - 8.5	13-23	180-320	26-46	>1600 >2800
Grass grown for seed, aerial spray, foliar, 3 apps. @ 1 lb. ai/A, 7-day intervals	Acute	0.008-4.1	--	9.4	170	18	490
	Subacute	0.18-0.32	1.7-3.1	--	--	--	--
	Reproduction NOAEL/NOAEC	6.2 - 15	2.4 - 6	14-26	200-380	29-54	>1700 >3300

Crop and Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuar-ine Fish	Estuarine Inverts.
Mint, ground spray, foliar, 1 app. @ 2 lbs. ai/A	Acute	0.009-4.7	--	4.1	74	7.7	210
	Subacute	0.023-0.36	0.22-3.5	--	--	--	--
	Reproduction NOAEL/NOAEC	3-17	1.2-6.7	6.5 -11	93-160	13-23	>810 >1400
Wheat, aerial spray, foliar, 2 apps. @ 0.5 lb. ai/A, 7-day interval	Acute	0.004-1.8	--	3.1	55	5.7	160
	Subacute	0.01-0.14	0.096-1.3	--	--	--	--
	Reproduction NOAEL/NOAEC	1.3-6.4	0.52-2.6	4.6-8.6	65-120	9.3-18	>570 >1100
Winter wheat, aerial spray, foliar, 1 app. @ 0.47 lb. ai/A (typical)	Acute	0.002-1.1	--	1.3	24	2.5	69
	Subacute	0.005 - 0.085	0.05-0.83	--	--	--	--
	Reproduction NOAEL/NOAEC	0.18 - 3.9	0.07 - 1.6	2-3.7	28-53	4-7.6	>240 >460

c. Peanuts

Risk quotients for use on peanuts are shown in Table 24. About 1.5 percent of total chlorpyrifos poundage is used on peanuts and is applied to 10-15 percent of the approximately 1,600,000 acres of peanuts in the U.S. The granular formulation is the primary treatment on peanuts. The Agency estimates that the typical use rate is 1.1 granular applications at an average of 1.8 lbs ai/A on approximately 160,000 to 240,000 acres. The leading states using chlorpyrifos in decreasing order of poundage are Georgia, North Carolina, Virginia and Alabama. The registrant has agreed to eliminate the granular aerial spraying of peanuts. Therefore, the risk to wildlife from the aerial spraying of granulars will be eliminated.

Wildlife utilization of peanut fields is relatively high with a fair diversity of avian and mammalian species. Wildlife reported to feed with moderate to high frequency in peanuts fields include bobwhite quail, doves, songbirds, waterfowl, wild turkey, rabbits, squirrels, raccoons, opossum, and deer. Bobwhite quail is the only species specifically listed as nesting in peanut fields.

Table 24. Range of Risk Quotients for Chlorpyrifos Use on Peanuts

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuar-ine Fish	Estuarine Inverts.
Ground spray, preplant, 1 app. @ 2 lbs. ai/A, 4" soil incorporation	Acute	0.009-4.7	--	1.4	24	2.5	70
	Subacute	0.023-0.36	0.22-3.5	--	--	--	--
	Reproduction NOAEL/NOAEC	3-17	1.2-6.7	2.2-3.8	31-54	4.4-7.8	>270 >470
Granular, 6" band, at-plant, 1 app. @ 2.25 oz ai/1000 ft, 4" soil incorp. (typical)	Acute	0.2-13	68	1.4	25	2.6	71
	Reproduction NOAEL/NOAEC	–	–	2.2-3.8	32-54	4.5-7.8	>270 >470
Granular, aerial broadcast, early pegging, 1 app. @ 1.95 lbs ai/A	Acute	0.21-13	71	0.92	17	1.7	47
	Reproduction NOAEL/NOAEC	–	–	1.5-2.5	21-36	3-5.1	>180 >320
Spray (preplant, 4" incorporation) followed by granular (early pegging, aerial broadcast), 2 apps. @ 2 lbs. ai/A, 40-day interval	Acute	NA[1]	NA	5.2	94	9.8	270
	Reproduction NOAEL/NOAEC	NA	NA	7.5-13	110-180	15-26	>930 >1600

[1] The Agency currently has no methodology for assessing risks from a combination of spray and granular formulations for terrestrial organisms. Therefore, only aquatic risks were assessed for this scenario.

d. Cotton

Risk quotients for use on cotton are shown in Table 25. The major chlorpyrifos use pattern on cotton is six foliar spray applications per season. The Agency estimates that about 3.2 percent of the total chlorpyrifos use is applied to up to 6 percent of the approximately 12,400,000 acres of cotton in the U.S. The typical average chlorpyrifos usage on cotton is 1.7 applications at 0.6 lbs ai/A on approximately 640,000 to 800,000 acres. The leading states using about 84 percent of the chlorpyrifos applied to cotton in decreasing order of poundage are Arizona, Mississippi, and California, Texas, and Louisiana.

Wildlife utilization of cotton fields is low to moderate. Wildlife that feed in cotton fields include quail, pheasant, doves, songbirds, rabbits, raccoon, and deer with a low to high degree of use. Bobwhite quail, pheasant (brood-rearing), and rabbits also nest and brood young in cotton fields.

Table 25. Range of Risk Quotients for Chlorpyrifos Use on Cotton

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuarine Fish	Estuarine Inverts.
Aerial spray, foliar, 6 apps. @ 1 lb. ai/A, 3-day intervals	Acute	0.015-7.6	--	15	270	28	780
	Subacute	0.036-0.58	0.36-5.7	--	--	--	--
	Reproduction NOAEL/NOAEC	4.9-28	1.9-11	30-40	340-570	62-82	>3800 >5000
Aerial spray, foliar, 1 app. @ 0.6 lb. ai/A	Acute	0.002-1.2	--	0.77	14	1.5	40
	Subacute	0.007 - 0.09	0.055-0.89	--	--	--	--
	Reproduction NOAEL/NOAEC	0.75-4.2	0.3-1.7	1.1-1.9	15-28	2.1-3.9	>130 >240

e. Citrus

Risk quotients for use on citrus are shown in Table 26. Citrus use represents about 3 percent of the total chlorpyrifos poundage. Chlorpyrifos is applied to oranges on about 60 percent of the total US acreage; grapefruit on about 12-16 percent or approximately 23,000 to 32,000 acres; lemons on about 30-43 percent or approximately 19,000 to 27,000 acres; and other citrus (including kumquats, limes, tangelos and tangerines) on about 16-32 percent of the total US acreage or about 8,000 to 16,000 acres. Maximum and typical risks for chlorpyrifos on citrus are assessed only for applications to oranges, because oranges represent the highest use rate and largest acreage of any citrus crop.

Wildlife utilization of citrus groves ranges from low to high for a diversity of avian and mammalian species (Gusey and Maturgo 1973). Mammals reported to feed moderately in citrus groves include raccoons and deer. Mourning doves, pheasants and 13 species of birds are listed as nesting in citrus groves. During the California orange field study in which two airblast applications were made, between 188 to 561 birds were observed in orange groves. Wildlife carcasses with chlorpyrifos residues found in the field study included a mockingbird, ground squirrel, pocket gopher and a western rattlesnake.

Table 26. Range of Risk Quotients for Chlorpyrifos Use on Citrus

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuarine Fish	Estuarine Inverts.
Airblast spray, foliar, 2 apps. @3.5 lbs. ai/A, 30-day interval, 5% spray drift	Acute	0.017-8.7	--	21	370	39	1100
	Subacute	0.041-0.66	0.4-6.5	--	--	--	--
	Reproduction NOAEL/NOAEC	5.5-88	2.2-35	33-54	470-770	67-110	>4100 >6700
Ground spray or sprinkler irrigation, 10 apps. @ 1 lb ai/A, 7-day interval	Acute	0.08-2.6	--	19	340	35	970
	Subacute	0.02-0.2	0.22-2	--	--	--	--
	Reproduction NOAEL/NOAEC	3-27	1.2-11	30-53	420-750	61-110	>3700 >6500
Airblast spray, foliar, 1 app. @ 6 lbs. ai/A, 5% spray drift	Acute	0.028-14	--	17	310	32	880
	Subacute	--	0.66-11	--	--	--	--
	Reproduction NOAEL/NOAEC	0-140	3.6-58	27-48	390-690	56-99	>3400 >6000

f. Golf Course Turf

Risk quotients for use on golf course turf are shown in Table 27. The volume of chlorpyrifos applied nationally on golf course turf and typical use rates have not been reported. Comparison of risk quotients for spray and granular applications on golf course turf at the same use rates suggest that the granular formulation is more acutely toxic to birds, mammals and other terrestrial species, while the spray formulation is only slightly more toxic to aquatic species. It is important to note that the risk quotients shown in Table 27 are based on application at the rate of 4 lbs. ai/A. Mitigation agreed to in June 2000 reduced the maximum application rate on golf course turf to 1 lb. ai/A. Therefore, actual RQs will be considerably lower than those shown below.

Table 27. Range of Risk Quotients for Chlorpyrifos Use on Golf Course Turf(a)

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuarine Fish	Estuarine Inverts.
Ground spray, 2 apps. @ 4 lbs. ai/A, 30-day interval	Acute	0.097-9.9	--	16	290	30	830
	Subacute	0.43-0.76	4.2-7.4	--	--	--	--
	Reproduction NOAEL/NOAEC	57-100	23-58	26-456	370-640	52-91	>3200 >5500

Application Method	Exposure Scenario	Mammals	Birds	Fresh-water Fish	Aquatic Inverts.	Estuarine Fish	Estuarine Inverts.
Granular, soil broadcast, 2 apps. @ 4 lbs. ai/A, 30-day interval	Acute	0.43-28	--	14	250	26	720
	Subacute	--	150	--	--	--	--
	Reproduction NOAEL/NOAEC	NA	--	22-39	320-550	46-79	>2800 >4800

(a) Mitigation agreed to in June, 2000, reduced the maximum application rate to golf course turf to 1 lb. ai/A. Therefore, actual RQs will be considerably lower than those shown.

Risk quotients for use on other, minor crops can be found in the *Environmental Fate and Effects Assessment*, June 8, 2000, located in the public docket and on the internet at www.epa.gov/pesticides/op.

5. Incidents

Bird kills involving mallard ducklings, geese, other waterfowl, robins and a bluebird have been reported for chlorpyrifos, most of which occurred following golf course and lawn treatments. These incidents were reported between 1974 and 1992. In some cases, carcass analysis detected more than one pesticide per carcass. Determination of the presence of chlorpyrifos in an animal or carcass only indicates that the animal was exposed.

Aquatic mortality incidents have also been reported, most of which were related to perimeter applications around residences. Incidents were reported between 1975 and 1992.

The preceding assessment indicates potential risks of concern to nontarget species. However, it should be noted that some mitigation measures implemented as a result of the June 2000 agreement are not reflected in the assessment. For example, all outdoor residential uses and most outdoor non-residential uses have been eliminated. The few remaining outdoor uses, golf courses, road medians and industrial plant sites are now limited to 1 lb. ai/A (reduced from 4 lbs. ai/A). These measures are expected to result in significant reductions in the levels of chlorpyrifos in surface water, particularly in urban areas.

To address ecological risk from the agricultural uses of chlorpyrifos, additional measures including rate reductions, aquatic buffer zones, seasonal limits and increased intervals between applications will be needed. These are outlined in the following section.

IV. Interim Risk Management and Reregistration Decision

A. Determination of Interim Reregistration Eligibility

Section 4(g)(2)(A) of FIFRA calls for the Agency to determine, after submissions of relevant data concerning an active ingredient, whether products containing the active ingredient are eligible for reregistration. The Agency has previously identified and required the submission of the generic (i.e., active ingredient specific) data required to support reregistration of products containing the active ingredient chlorpyrifos.

The Agency has completed its assessment of the occupational and ecological risks associated with the use of chlorpyrifos, as well as a chlorpyrifos-specific dietary risk assessment that has not considered the cumulative effects of organophosphates as a class. Based on a review of these data and public comments on the Agency's assessments for the active ingredient chlorpyrifos, EPA has sufficient information on the human health and ecological effects of chlorpyrifos to make interim decisions as part of the tolerance reassessment process under FFDCA and reregistration under FIFRA, as amended by FQPA. Taking into account both risks and benefits, the Agency has determined that, with the exception of open-pour dust formulations for fire ant control, products containing chlorpyrifos uses are eligible for reregistration provided that: (i) current data gaps and additional data needs are addressed; (ii) the risk reduction measures outlined in this document as well as those in the Memorandum of Agreement of June 2000 are adopted, and label amendments are made to reflect these measures; and (iii) cumulative risks considered the organophosphates support a final reregistration eligibility decision. Label changes are described in Section IV. Appendix B identifies the generic data requirements that the Agency reviewed as part of its interim determination of reregistration eligibility of chlorpyrifos products, and lists the submitted studies that the Agency found acceptable.

Although the Agency has not yet considered cumulative risks of the organophosphates, the Agency is issuing this interim assessment now in order to identify risk reduction measures that are necessary to support the continued use of chlorpyrifos. Based on its current evaluation of chlorpyrifos alone, the Agency has determined that chlorpyrifos products, unless labeled and used as specified in this document, would present risks inconsistent with FIFRA. Accordingly, should a registrant fail to implement appropriate risk mitigation measures, the Agency will take regulatory action to address the risk concerns from use of chlorpyrifos.

At the time that a cumulative assessment is conducted, the Agency will address any outstanding risk concerns. For chlorpyrifos, if all changes outlined in this document are incorporated into the labels, risks will be mitigated to acceptable levels taking into account the benefits of chlorpyrifos use where appropriate. But, because this is an interim RED, the Agency may take further actions, if warranted, to finalize the reregistration eligibility decision for chlorpyrifos products after assessing the cumulative risk of the organophosphate class. Such an incremental approach to the reregistration process is consistent with the Agency's goal of improving the transparency of the reregistration and tolerance reassessment processes. By

evaluating each organophosphate in turn and identifying appropriate risk reduction measures, the Agency is addressing the risks from the organophosphates in as timely a manner as possible.

Because the Agency has not yet considered cumulative risks for the organophosphates, this reregistration eligibility decision does not fully satisfy the reassessment of the existing chlorpyrifos food residue tolerances as called for by FQPA. When the Agency has considered cumulative risks, chlorpyrifos tolerances will be reassessed in that light. At that time, the Agency will reassess chlorpyrifos along with the other organophosphate pesticides to complete the FQPA requirements and make a final reregistration eligibility determination. By publishing this interim decision on reregistration eligibility and requesting mitigation measures now for the individual chemical chlorpyrifos, the Agency is not deferring or postponing FQPA requirements; rather, EPA is taking steps to assure that uses which EPA has already determined exceed FIFRA's unreasonable risk standard do not remain on the label, pending completion of assessment required under the FQPA. This decision does not preclude the Agency from making further FQPA determinations and tolerance-related rulemakings that may be required on this pesticide or any other in the future.

If the Agency determines, before finalization of the RED, that any of the determinations described in this interim RED are no longer appropriate, the Agency will pursue appropriate action, including but not limited to, reconsideration of any portion of this interim RED.

B. Regulatory Position

1. FQPA Assessment

a. "Risk Cup" Determination

As part of the FQPA tolerance reassessment process, EPA assessed the risks associated with this organophosphate. The assessment is for this individual organophosphate, and does not attempt to fully reassess these tolerances as required under FQPA. FQPA requires the Agency to evaluate food tolerances on the basis of cumulative risk from substances sharing a common mechanism of toxicity, such as the toxicity expressed by the organophosphates through a common biochemical interaction with the cholinesterase enzyme. The Agency will evaluate the cumulative risk posed by the entire class of organophosphates once the methodology is developed and the policy concerning cumulative assessments is resolved.

EPA has determined that risk from exposure to chlorpyrifos is within its own "risk cup." In other words, if chlorpyrifos did not share a common mechanism of toxicity with other chemicals, EPA would be able to conclude today that the tolerances for chlorpyrifos meet the FQPA safety standards. In reaching this determination EPA has considered the available information on the special sensitivity of infants and children, as well as the chronic and acute food exposure. An aggregate assessment was conducted for exposures through food, residential uses and drinking water. Results of this aggregate assessment indicate that the human health risks from these combined exposures are considered to be within acceptable levels; that is,

combined risks from all exposures to chlorpyrifos "fit" within the individual risk cup. Therefore, except for tolerances that will be revoked as indicated in Tables 28 and 29, the chlorpyrifos tolerances remain in effect and unchanged until cumulative risks from all organophosphates are considered.

b. Tolerance Summary

In the individual assessment, established tolerances for residues of chlorpyrifos in/on raw agricultural, animal, and processed food/feed commodities [40 CFR §180.241] are presently expressed in terms of either the combined residues of chlorpyrifos and its metabolite 3,5,6-trichloro-2-pyridinol (TCP) or as chlorpyrifos *per se*. The Agency has determined that residues of TCP are not of concern for dietary risk and can therefore be excluded from the tolerance expression. The tolerance levels should be amended to reflect residues of chlorpyrifos *per se*. Based on the Agency's decision to change the tolerance expression, the tolerances listed in 40 CFR need to be reorganized as shown in Table 28. A summary of the tolerances is included in Table 29.

Table 28. Reorganization of Tolerances for Chlorpyrifos

	Current Tolerance	Tolerance Reassessment*	
40 CFR	**Expression [Restrictions]**	**40 CFR**	**Tolerance Expression [Restrictions]**
§180.342 (a)(1)	Chlorpyrifos and TCP.	§180.342 (a)(1)	Chlorpyrifos per se.
§180.342 (a)(2)	Chlorpyrifos per se.	§180.342 (a)(1)	Transfer all tolerances under this section to §180.342 (a)(1) at their respective proposed levels.
§180.342(a)(3)	[Provisions on safe use of chlorpyrifos on food-handling establishments].	§180.342(a)(2)	Conditions for safe use of chlorpyrifos on food-handling establishments. Redesignate as §180.342(a)(2).
§180.342(a)(4)	Chlorpyrifos per se (tolerances established in food items [other than those already covered by a higher tolerance as a result of use on growing crops] in food-service establishments, as result of the application of microencapsulated form.	§180.342(a)(3)	Chlorpyrifos per se. Redesignate as §180.342(a)(3).
§180.342 (c)(1)	Chlorpyrifos and TCP [For regional registrations].	§180.342 (c)	Chlorpyrifos per se [For regional registrations].
§180.342 (c)(2)	Chlorpyrifos per se [For regional registrations].		Delete §180.342 (c)(2) section since all tolerances under this section are to be revoked (no registered uses).

* The term "reassessed" here is not meant to imply that the tolerance has been reassessed as required by FQPA, since this tolerance may be reassessed only upon completion of the cumulative risk assessment of all organophosphates, as required by this law. Rather, it provides a tolerance level for this single chemical, if no cumulative assessment was required, that is supported by all of the submitted residue data.

Table 29. Tolerance Summary for Chlorpyrifos.

Commodity	Current Tolerance (ppm)	Tolerance Reassessment* (ppm)	[*Correct Commodity Definition*]/ Comments
Tolerances Listed Under 40 CFR §180.342(a)(1)			
Alfalfa, forage	3	3	
Alfalfa, hay	13	13	
Almonds	0.2	0.2	[*Almond*].
Almonds, hulls	12.0	12.0	[*Almond, hulls*].
Apple, pomace, wet	None	0.02	[*Apple, wet pomace*] Proposed tolerance (0.01 ppm) and average concentration factor (2.1).
Apples	1.5	0.01	[*Apple*].The registrant has submitted a rebuttal to the modification of this tolerance. This rebuttal is under review.
Aspirated grain fractions	None	TBD	[*Grain, aspirated grain fractions*]. A 0.5 ppm tolerance was recommended for corn aspirated grain fractions based on a concentration factor of ~10x in the <420 μ dust fraction (see CBRS No. 11372, D188151, S. Knizner, 8/26/93). Additional data are required for sorghum, soybean, and wheat aspirated grain fractions before a tolerance for aspirated grain fractions can be established (see "Aspirated Grain Fractions (Grain Dust): A Tolerance Perspective", E.Saito and E.Zager, 6/7/94.
Bananas, whole	0.1	0.1	
Bananas, pulp with peel removed	0.01	0.01	
Bean, forage	0.7	Revoke	Not a feed item Table 1 (OPPTS 860.1000)
Beans, lima	0.05	Reassign	Covered by legume vegetables group.
Beans, lima, forage	1.0	Revoke	Not a food/feed item.
Beans, snap	0.05	Reassign	Covered by legume vegetables group.
Beans, snap, forage	1.0	Revoke	Not a food/feed item.
Beets, sugar, molasses	15.0	15.0	[*Beet, sugar, molasses*].
Beets, sugar, pulp (dried)	5.0	5.0	[*Beet, sugar, dried pulp*].
Beets, sugar, roots	1.0	1.0	[*Beet, sugar, roots*].
Beets, sugar, tops	8.0	8.0	[*Beet, sugar, tops*].
Blueberries	2 (1) [a]	Revoke	No registered uses exist.
Broccoli	1	Reassign	Covered by *Vegetable, Brassica, leafy, group.*
Brussels sprouts	1	Reassign	Covered by *Vegetable, Brassica, leafy, group.*
Cabbage	1	Reassign	Covered by *Vegetable, Brassica, leafy, group.*
Caneberries	1.0	Revoke	No registered uses exist.
Cattle, fat	0.3	0.3	
Cattle, meat and meat byproducts	0.05	0.05	[*Cattle, meat*]
	0.05	0.05	[*Cattle, meat byproducts*]

Commodity	Current Tolerance (ppm)	Tolerance Reassessment* (ppm)	[*Correct Commodity Definition*]/ Comments
Cauliflower	1	Reassign	Covered by *Vegetable, Brassica, leafy, group.*
Cherries	1	TBD	[*Cherries, sweet*] Additional data and/or label revisions are required.
		TBD	[*Cherries, tart*] Additional data and/or label revisions are required.
Chinese cabbage	1	Reassign	Covered by *Vegetable, Brassica, leafy, group.*
Citrus fruits	1.0	1.0	[*Fruit, citrus, group*].
Citrus oil	25.0	20	
Citrus pulp, dried	5.0	5.0	[*Citrus, dried pulp*].
Clover, forage	None	TBD	
Clover, hay	None	TBD	
Corn, fresh (inc. sweet K-CWHR)	0.1	0.05	[*Corn, sweet , kernel plus cob with husks removed*].
Corn, field, grain	0.05	0.05	
Corn, forage	8	8	[*Corn, field, forage*]
	8	8	[*Corn, sweet, forage*]
Corn, fodder	8	8	[*Corn, field, stover*]
	8	8	[*Corn, sweet, stover*]
Corn oil	3.0	0.25	[*Corn, field, refined oil*]/ Recommended tolerance based on a average concentration factor of 3.3x (see CBRS No. 11372, D188151, S. Knizner, 8/26/93).
Cotton, gin byproducts	None	TBD	
Cottonseed	0.2	0.2	[*Cotton, undelinted seed*]
Cranberries	1.0	1.0	[*Cranberry*]
Cucumbers	0.05	0.05	[*Cucumber*]
Eggs	0.01	0.01	[*Egg*]
Figs	0.01	0.01	[*Fig*]
Filbert	None	0.2	[*Filbert*] Use previously covered under tree nuts.
Goats, fat	0.2	0.2	[*Goat, fat*]
Goats, meat and meat byproducts	0.05	0.05	[*Goat, meat*]
	0.05	0.05	[*Goat, meat byproducts*]
Grass, forage	None	TBD	
Grass, hay	None	TBD	
Grass, seed screenings	None	TBD	
Hogs, fat	0.2	0.2	[Hog, fat]
Hogs, meat	0.05	0.05	[Hog, meat]
	0.05	0.05	[Hog, meat byproducts]

Commodity	Current Tolerance (ppm)	Tolerance Reassessment* (ppm)	[*Correct Commodity Definition*]/ Comments
Horses, fat	0.25	0.25	[Horse, fat]
Horses, meat	0.25	0.25	[Horse, meat]
	0.25	0.25	[Horse, meat byproducts]
Kiwifruit	2.0	2.0	
Legume vegetables, succulent or dried (except soybeans)	0.05	0.05	[*Vegetable, legume, group*]
Lettuce	None	1	Recommended tolerance from PP#4F03132.
Macadamia nut	None	0.2	Use previously covered under tree nuts.
Milk, fat	0.25	0.25	[*Milk fat (reflecting 0.01 ppm in whole milk)*]/ Recommended tolerance from PP#3F2884.
Milk, whole	0.01	Reassign	Covered by tolerance from milk fat (reflecting 0.01 ppm in whole milk).
Mint, hay	0.8	0.8	[*Peppermint, tops*]
		0.8	[*Spearmint, tops*]
Mushrooms	0.1	Revoke	No registered uses exist.
Nectarines	0.05	Revoke	[*Nectarine*]
Onions (dry bulb)	0.5	0.5	[*Onion, dry bulb)*].
Pea forage	0.7	Revoke	Not a feed item (Table 1, OPPTS 860.1000)
Peaches	0.05	0.05	[*Peach*]
Peanuts	0.2	0.2	[*Peanut, nutmeat]*.
Pears	0.05	0.05	
Plums (fresh prunes)	0.05	0.05	[*Plums*]
Pecan	None	0.2	Use previously covered under tree nuts.
Peppers	1.0	1.0	[*Pepper*] Chlorpyrifos labels from foreign countries that import peppers to the U.S. are required.
Poultry, meat, fat, and meat byproducts (inc. turkeys)	0.1	0.1	[*Poultry,fat*]
		0.1	[*Poultry, meat*]
		0.1	[*Poultry, meat byproducts*]
Pumpkins	0.05	0.05	[*Pumpkin*]
Radishes	2	2	[*Radish*]
Rutabagas	0.5	0.5	[*Rutabaga, root*]
Seed and pod vegetables	0.1	Revoke	Uses of chlorpyrifos on dill and okra, for which this obsolete crop group was supposed to cover, have been deleted.
Sheep, fat	0.2	0.2	
Sheep, meat and meat byproducts	0.05	0.05	[*Sheep, meat*]
		0.05	[*Sheep, meat byproducts*]
Soybean grain	0.3	0.3	[*Soybean, seed*].

Commodity	Current Tolerance (ppm)	Tolerance Reassessment* (ppm)	[*Correct Commodity Definition*]/ Comments
Soybean forage	0.7	Revoke	Feeding may be restricted on the label.
Sorghum, fodder	6.0	2.0	[*Sorghum, grain, stover*]. Recommended tolerance from PP#4F3008/FAP#1H5295.
Sorghum, forage	1.5	0.5	[*Sorghum, grain, forage*].
Sorghum, grain	0.75	0.5	[*Sorghum, grain, grain*].
Sorghum milling fractions	1.5	Revoke	According to Table 1, OPPTS Test Guidelines 860, August 1996, sorghum flour is used exclusively in the US as a component for drywall, not as either a human or animal feed item.
Strawberries	0.2	0.2	[*Strawberry*].
Sugarcane	0.01	Revoke	No registered uses exist.
Sunflower, seeds	0.25	0.1	[*Sunflower, seed*]. Recommended tolerance from PP#4F3008/FAP#1H5295.
Sweet potatoes	0.05	0.05	[*Sweet potato, root*].
Tomatoes	0.5	Revoke	The registrant has submitted a rebuttal to the modification of this tolerance. This rebuttal is under review.
Tree nuts	0.2	Reassign	Individual tolerances exist for almond and walnut, and are being established for filbert, pecan, and macadamia nut.
Turnip greens	0.3	0.3	[*Turnip, tops*].
Turnips	1	1	[*Turnip, root*].
Vegetables, leafy, *Brassica* (cole)	2.0 (1.0) [a]	1.0	[*Vegetable, Brassica, leafy, group*].
Walnuts	0.2	0.2	[*Walnut*].
Wheat, forage	3	3	
Wheat, grain	0.5	0.5	
Wheat, hay	None	TBD	
Wheat, straw	6	6	
Tolerances Listed Under 40 CFR §180.342(a)(2)			
Milling fractions (except flour) of wheat	1.5	Reassign	Wheat tolerance for wheat (0.5 ppm) will cover processed milling fractions under the revised procedures for the determination of need for food additive tolerances.
Mint oil	8	8	[Peppermint, oil]
		8	[Spearmint, oil]
Peanut oil	0.4	0.2	[*Peanut, refined oil*] Revised procedures for calculating food additive tolerance values. (HAFT (0.11) x average processing factor (1.7)).
Tolerances Listed Under 40 CFR §180.342(c)(1)			
Asparagus	5.0	5.0	

Commodity	Current Tolerance (ppm)	Tolerance Reassessment* (ppm)	[*Correct Commodity Definition*]/ Comments
Dates	0.5 (0.3) [a]	Revoke	[*Date*] No registered uses exist.
Grapes	0.5	0.01	[*Grape*] Tolerance based on currently registered US use pattern. The registrant has submitted a rebuttal to the modification of this tolerance. This rebuttal is under review.
Leeks	0.5 (0.2) [a]	Revoke	[*Leek*] No registered uses exist.
Tolerances Listed Under 40 CFR §180.342(c)(2)			
Cherimoya	0.05	Revoke	No registered uses exist.
Feijoa (pineapple guava)	0.05	Revoke	No registered uses exist.
Sapote	0.05	Revoke	No registered uses exist.

* The term "reassessed" here is not meant to imply that the tolerance has been reassessed as required by FQPA, since this tolerance may be reassessed only upon completion of the cumulative risk assessment of all organophosphates, as required by this law. Rather, it provides a tolerance level for this single chemical, if no cumulative assessment was required, that is supported by all of the submitted residue data.

The Agency will commence proceedings to modify the existing tolerances, and correct commodity definitions. The revocation of a tolerance, establishment of a new tolerance, or the raising or lowering of tolerances will be deferred until submitted data are reviewed.

c. Codex Harmonization

Residue data used to establish U.S. tolerances were examined to determine if U.S. tolerance levels could be adjusted to harmonize with Codex Maximum Residue Limits (MRLs). Whenever possible, tolerance levels were changed to achieve harmonization.

Several maximum residue limits (MRLs) for chlorpyrifos have been established by Codex in various commodities as shown below in Table 30. The Codex MRLs (expressed in terms of chlorpyrifos *per se*) and the U.S. tolerance expression will be compatible when TCP is deleted from the U.S. tolerance expressions.

Compatibility between the U.S. tolerances and Codex MRLs exists for cabbage, Chinese; kale [Brassica (cole) leafy vegetables group]; kiwifruits; milks; and poultry meat. Further harmonization of U.S. tolerances and Codex MRLs on other commodities are not feasible at this time. U.S. tolerances are based on domestic use patterns supported by domestic field trial data. Codex MRLs may differ from U.S. tolerances because of different use patterns in foreign countries.

Table 30. Codex MRLs and Applicable U.S. Tolerances

Commodity	MRL (mg/kg) [a]	U.S. Tolerance (ppm) [b]	Recommendation/ Comments
Apple	1	0.01	--
Cabbages, head	0.05 [c]	1	--
Carrot	0.5	None	--
Cattle meat	2 (fat)	0.05	--
Cauliflower	0.05 [c]	1	--
Celery	0.05 [c]	None	--
Chicken meat	0.1 (fat)	0.1	Compatibility exists.
Chinese cabbage, type "Pe-tsai"	1	1	Compatibility exists.
Citrus fruits	0.3	1.0	--
Common bean (pods and/or immature seeds)	0.2	0.05 (Legume vegetables group, except soybeans)	--
Cottonseed	0.05 [c]	0.2	--
Cotton seed oil, crude	0.05 [c]	None	--
Dried grapes	2	0.5	Recommend increase to 1.0.
Eggplant	0.2	None	--
Eggs	0.05 [c]	0.01	--
Grapes	1	0.01	
Kale	1	1 (Brassica (cole) leafy vegetables group)	Compatibility exists.
Kiwifruit	2	2.0	Compatibility exists.
Lettuce, head	0.1	1 (proposed)	--
Milk	0.01 [c]	0.01	Compatibility exists.
Mushrooms	0.05 [c]	Revoke	No registered US use.
Onion, bulb	0.05 [c]	0.5	--
Pear	0.5	0.05	--
Peppers	0.5	1.0	--
Potato	0.05 [c]	None	--
Raspberries, red, black	0.2	1.0 (caneberries)	--
Rice	0.1	None	--
Sheep meat	0.2 (fat)	0.05	--
Tomato	0.5	Revoke	under review
Turkey meat	0.2 (fat)	0.1 (poultry meat, including turkeys)	--

[a] All chlorpyrifos MRLs are final (CXL).
[b] Based on chlorpyrifos *per se*.
[c] At or about the limit of detection.

d. Endocrine Disruptor Effects

EPA is required under the FFDCA, as amended by FQPA, to develop a screening program to determine whether certain substances (including all pesticide active and other ingredients) "may have an effect in humans that is similar to an effect produced by a naturally occurring estrogen, or other such endocrine effects as the Administrator may designate." Following the recommendations of its Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC), EPA determined that there were scientific bases for including, as part of the program, the androgen and thyroid hormone systems, in addition to the estrogen hormone system. EPA also adopted EDSTAC's recommendation that the Program include evaluations of potential effects in wildlife. For pesticide chemicals, EPA will use FIFRA and, to the extent that effects in wildlife may help determine whether a substance may have an effect in humans, FFDCA authority to require the wildlife evaluations. As the science develops and resources allow, screening of additional hormone systems may be added to the Endocrine Disruptor Screening Program (EDSP).

When the appropriate screening and/or testing protocols being considered under the Agency's EDSP have been developed, chlorpyrifos may be subjected to additional screening and/or testing to better characterize effects related to endocrine disruption.

e. Labels

Provided the following risk mitigation measures are incorporated in their entirety into labels for chlorpyrifos-containing products, the Agency finds that, with the exception of the dust formulation for fire ant control, all currently registered uses of chlorpyrifos are eligible for reregistration, pending consideration of cumulative risks of the organophosphates. The regulatory rationale for each of the mitigation measures outlined below is discussed immediately after this list of mitigation measures.

Dietary Risk

Neither acute nor chronic dietary (food and drinking water) risks are of concern. This conclusion reflects measures agreed to in the Memorandum of Agreement of June 2000 eliminating use on tomatoes and limiting use on grapes and apples. No further mitigation is necessary at this time.

Occupational Risk

In order for chlorpyrifos products (except for the dust formulation for fire ant control) to be eligible for reregistration, a combination of reduced application rates and seasonal maximum limits, increased retreatment intervals, increased PPE and/or use of engineering controls to address occupational handler risks are needed. In addition, increased REIs for a number of crops will address postapplication risks to workers. Taking into account all feasible mitigation, several worker scenarios are still below the target MOE of 100. In such cases, and in accordance with

PR Notice 2000-9, EPA further characterizes the risk by looking at the strengths and weaknesses of the data and assumptions used in the risk assessment and evaluates the benefits of a chemical's use. The worker scenarios are discussed further below.

Residential Risk

No mitigation is necessary at this time. All products for homeowner use except ant and roach baits in child-resistant packaging have been canceled. Professional termiticide treatment products are being phased out, with all use for termite control prohibited by December 31, 2005.

Ecological Risk

Risks to terrestrial and aquatic organisms are of concern for all outdoor uses of chlorpyrifos. To address these risks, reductions in application rates, the number of applications per season and the maximum amount that may be applied per acre per season and increased intervals between applications will be needed. In addition, no-spray buffer zones will be applied to protect water bodies, further mitigating aquatic risks. Taking into account mitigation, some aquatic risk quotients still exceed levels of concern, particularly for estuarine invertebrates. EPA has considered benefits of chlorpyrifos use on the major crops contributing to aquatic risk concerns. The Agency will also require submission of water monitoring data to confirm the reduction of chlorpyrifos levels in surface water.

C. Regulatory Rationale

The following is a summary of the rationale for managing risks associated with the current use of chlorpyrifos products. Where labeling revisions are warranted, specific language is set forth in the summary tables of Section V of this document.

1. Benefits

The Agency has considered the benefits of chlorpyrifos use in its determination of eligibility for reregistration as well as appropriate reduction of remaining risks. Since corn, cotton, citrus and alfalfa represent approximately 70% - 80% of the use of chlorpyrifos and thus are the greatest contributors to ecological risk, the Agency has considered the benefits of chlorpyrifos use on these sites.

Corn

Chlorpyrifos use on corn (an estimated 5 ½ to 7 million pounds) accounts for more than half of the total annual use of chlorpyrifos in agriculture. Chlorpyrifos is applied to corn primarily to control corn rootworm (larvae and adults), cutworm and European corn borer. Corn growers considered chlorpyrifos critical for control of these damaging pests. The granular product is primarily incorporated in the soil at the time corn is planted for control of rootworm

larvae. This type of application represents the largest use of chlorpyrifos with approximately 4 to 5 ½ million pounds applied annually. Granular applications have the additional benefit of protecting the corn from cutworm. Foliar applications of granular chlorpyrifos by air are targeted at European corn borer. This method represents a relatively small portion of chlorpyrifos use–approximately 100,000 pounds of active ingredient per year. Approximately 500,000 pounds of the liquid formulation of chlorpyrifos are applied to corn per year. The liquid formulation is generally used as a foliar application, with some at-plant use as well.

The principal alternatives to chlorpyrifos on corn are terbufos (which is currently undergoing reregistration), tefluthrin, fipronil, and a combination product of tebupirimphos and cyfluthrin. The most effective non-chemical alternative for management of corn rootworm is crop rotation, which is practiced on the majority of corn acreage.

Citrus

Approximately 600,000 pounds of chlorpyrifos are applied annually to citrus primarily in California and to a lesser extent in Florida. Chlorpyrifos is the most effective product available for the control of California red scale (CRS). Other insecticides used to control CRS include methidathion, carbaryl, and oil. Chlorpyrifos is preferred due to its effectiveness against CRS and its relatively short residual activity compared to the other available insecticides. Chlorpyrifos' short residual minimizes the impact on beneficial insects such as the *Aphytis* wasp, which is important for late season biological control of CRS populations. The majority of California citrus is grown for the fresh market and for export. Although CRS damage is primarily cosmetic, there is a low threshold for CRS damaged fruit in these markets.

In Florida, Chlorpyrifos is used as an alternative chemical control for managing scale and thrips, and it is used to manage nuisance pests such as fire ants and termites in the grove. The majority of the chlorpyrifos use in Florida is for the control of fire ants. There are currently no alternatives labeled for this use. Fire ant control is critical to allow workers the opportunity to complete orchard production activities, such as harvesting, without the threat of attack by the fire ants.

Cotton

Approximately 700,000 pounds of chlorpyrifos are applied annually to cotton. Liquid chlorpyrifos is used on cotton primarily to control plant bugs in the Mississippi delta area, cotton aphid in Texas and California, silverleaf whitefly in Arizona, pink bollworm in Arizona and beet armyworms in all cotton growing areas. It is considered to be important in resistance management programs for cotton aphid. Alternatives to chlorpyrifos for aphid control include profenofos and carbofuran. Imidacloprid provides early season aphid and plant bug control. Two relatively new insect growth regulators (IGR), pyriproxyfen and buprofizen, have shown good control of silverleaf whitefly.

Alfalfa

Approximately 500,000 lbs. ai of chlorpyrifos are applied annually to alfalfa by both ground (Midwest to Northeast) and air (West) equipment. A single application per year is typical. Alfalfa weevil, Egyptian alfalfa weevil, armyworms (beet and Western yellowstriped) and aphids are the key pests. The principal alternatives to chlorpyrifos are carbofuran, methyl parathion and dimethoate. Pyrethroids are also registered for alfalfa pest management, but do not suppress and control aphids, as well as chlorpyrifos, carbofuran and methyl parathion.

Since corn, cotton, citrus and alfalfa represent 70% - 80% of the chlorpyrifos use, the Agency has considered the benefits of chlorpyrifos use on these sites. Additional benefits information on these and other uses can be found in the public docket and is discussed under specific worker scenarios below in the Occupational Risk Mitigation section. Usage information can also be found at http://pestdata.ncsu.edu/cropprofiles/cropprofiles.cfm.

2. Human Health Risk Mitigation

a. Dietary Mitigation

1) Acute Dietary (Food)

Based on use patterns established before the June 2000 mitigation agreement, acute dietary risk from food alone at the 99.9th percentile for the most highly exposed subpopulation, children 1-6 years old was 355% of the aPAD. The mitigation agreement addressed this risk by reducing or canceling use on three commodities frequently consumed by children: apples, grapes and tomatoes. Post-bloom use on apples was removed from product labels effective December 31, 2000 and the tolerance will lowered to 0.01 ppm. Production of products for use on tomatoes was prohibited effective September 2000, and use of existing products was stopped as of December 31, 2000. The tolerances for tomatoes will be revoked. The tolerance for grapes will be lowered to 0.01 ppm to reflect domestic use patterns. The Agency is coordinating with the FDA to implement these tolerance reductions/revocations. The registrant has submitted a rebuttal to the modification of the tolerances. This rebuttal is under review.

With implementation of these reductions, acute dietary risk from food alone is at 82% of the aPAD for children 1-6 years old, and thus is not of concern. No further mitigation of acute dietary risk is needed at this time.

2) Chronic Dietary (Food)

Prior to implementation of the mitigation for apples, grapes and tomatoes, chronic dietary risk from food alone occupied 81% of the cPAD for children 1-6 years old, the most highly exposed population subgroup, and thus was not of concern. The mitigation further reduced risks

to a range of 2.5% to 51% of the cPAD. No additional mitigation of chronic dietary risk is needed at this time.

3) Drinking Water

Neither acute nor chronic risks from drinking water are of concern for any population subgroup, except in the event of well contamination following termiticide use. Incidents of these types have occurred in the past as a result of the high concentrations required for termiticide use, treatments being applied when wells were in or near the building foundation, and/or when well casings were cracked. Since issuance of PR 96-7 instituting risk reduction measures for termiticides, the number of reported incidents has dropped significantly. For example, the frequency of incidents in 1997 (before PR 96-7) was 28.2 per 100,000 homes; in 1998 (after the notice) the frequency was 8.3 per 100,000 homes.

To address these remaining risks, termiticide products were reclassified to "restricted use." In addition, the application rate for all termiticide products was limited to 0.5% solution effective December 1, 2000. Use and sale of termiticide products will be phased out as follows: formulation of products for post-construction treatment stopped on December 1, 2000, and all sales of whole-house and spot/local treatment products will stop effective December 31, 2001, and December 31, 2002, respectively. Production of products for pre-construction treatment will stop as of December 31, 2004; these products may not be used after December 31, 2005. A provision of the June 2000 agreement allows the technical registrants to submit exposure data by June 2004. If acceptable data demonstrate that pre-construction use does not pose risks of concern to residents, that use may be allowed to continue.

b. Occupational Risk Mitigation

1) Agricultural and Ornamental/Greenhouse Handler Risks

Since the chlorpyrifos occupational assessment was completed, some refinements in methodology have been identified. In calculating occupational handler risks for the preliminary *Human Health Risk Assessment* completed in June 2000, the potential dermal and inhalation doses used to calculate exposures were those identified in the Agency's Series 875 Group A (previously known as Subdivision U).

However, for dermal calculations, the ratio of the body surface area to the body weight has been found to overestimate risk by a factor of 1.1. The ratio is not physiological matched in that the surface area is for an average male, while the body weight is the median for both male and female. Therefore, dermal MOEs from the June 2000 assessment have been adjusted with a reduction factor of 1.1 and are presented in the following table.

In addition, to calculate inhalation risks for handlers, the Agency used a standard breathing rate of 29 L/min for all exposure scenarios. Since that time, the Agency has adopted the breathing rates recommended by NAFTA. The NAFTA inhalation rates and the

corresponding exposure reduction factors are: 8.3 L/min. for sedentary activities (e.g., driving a tractor); exposure reduction factor 3.5; 16.7 L/min. for light activities (e.g., flaggers and mixer/loaders using <50 lb. containers); exposure reduction factor 1.7; and 26.7 L/min. for moderate activities (e.g., loading >50 lb. containers or using handheld equipment in hilly areas); exposure reduction factor 1.1.

Table 31 presents the MOEs for occupational risk taking into account the revised dermal surface area and breathing rate factors.

Table 31. Occupational Risk Estimates for Agricultural Uses of Chlorpyrifos

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre)	Daily Acres Treated	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
Mixer/Loader Exposure								
Mixing/Loading Liquids for Aerial/Chemigation Application (1a)	1.5 cranberries, corn	350	43	95	30	86	272	66
	3.5 citrus	100	65	141	44	132	408	100
Mixing/Loading Liquids for Groundboom Application (1b)	1.5 predominant max	80	187	408	128	Target MOE reached at PPE		
	2 Sodfarm (includes tobacco/ potatoes)	80	143	306	97	275	901	211
	3 Sodfarm	80	88	193	60	278	861	210
	8.0 sodfarm fire ants	10	286	612	195	Target MOE reached at PPE		
Mixing/Loading Liquids for Airblast Application (1c)	2.0 predominant max such as Fruits & Nuts	40	286	612	195	Target MOE reached at PPE		
	6.0 citrus	20	187	408	128	Target MOE reached at PPE		
Mixing WP for Aerial/Chemigation Application (2a)	2.0 predominant max (orchards)	350	DAS is not supporting the open bag formulation for the WP			56	71	31
	3.5 citrus (d)	100				110	141	62
Mixing WP for Groundboom Application (2b)	1.0 predominant max (brassica)	80				495	612	274
	4.0 soil treatment ornamentals outdoors	10				979	1241	547
	1.3 & 3.0 Sodfarm	80				374 / 165	476 / 204	209 / 91
	8.0 sodfarm fire ants (harvest only)	10				495	360	200
Mixing WP for Airblast Application (2c)	2.0 predominant max	40				495	612	274
	6.0 citrus	20				330	408	182
Loading Granulars for Aerial Application (3a)	1.0 maximum aerial rate for corn	350	321	99	75	3300	510	442

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre)	Daily Acres Treated	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
Loading Granulars for Ground Application (3b)	1.0 typical corn	80	1430	442	338	Target MOE reached at PPE		
	2.0 max corn	80	704	221	168	Target MOE reached at PPE		
	3.0 maximum ground rate (tobacco)	80	473	146	112	Target MOE reached at PPE		
Applicator Exposure								
Aerial (Spray) -- Enclosed Cockpit (4a)	2.0 orchards	350	No Open cockpit data available			110	525	91
	3.5 citrus	100				220	1015	181
Aerial (Granulars) -- Enclosed Cockpit (4b)	1.0	350	No Open cockpit data available			686	55	51
Groundboom Tractor (5)	1.5 predominant max	80	The biological monitoring results (Table A4) indicate that open cabs provide insufficient protection. Therefore, only the enclosed cab MOEs are presented.			638	4900	564
	3 Sodfarms	80				302	2231	270
	8.0 sodfarm fire ants	10				968	7000	850
Airblast Applicator (6)	2.0 predominant max	40	The biological monitoring results indicate that open cabs are insufficient.			253	665	183
	6.0 citrus	20				165	455	121
Tractor-Drawn Granular Spreader (7)	1.0 typical corn	80	1100	1260	587	Target MOE reached at PPE		
	2.0 max corn	80	572	630	300	Target MOE reached at PPE		
	3.0 maximum ground rate (tobacco)	80	385	420	201	Target MOE reached at PPE		
Seed Treatment (8)	No Data	No Data	No Data			No Data		
Dip Application (Preplant Peaches) (9)	No Data	No Data	No Data			No Data		
Flagger Exposure								
Spray Applications (10)	2.0 predominant max	350	55	490	49	2530	1540	957
	3.5 citrus (d)	100	110	319	82	4950	3190	1940
Granular Applications (11)	1.95	350	352	374	181	Target MOE reached at PPE		
Mixer/Loader/Applicator Exposure								

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre)	Daily Acres Treated	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
Backpack Sprayer/Bark and Pine Seedling Treatment (12)	0.0417 lb ai/gal predominant max	40 gal/day	143	770	121	Target MOE reached at PPE,		
	0.08 lb ai/gal bark beetle treatment	40 gal/day	75	396	63	Not feasible		
	0.03 lb ai/gal stump treatment	40 gal/day	198	1067	167	Target MOE reached at PPE,		
	0.16 lb ai/gal pine seedling treatment	40 gal/day	37	198	31	Not feasible		
	3.5 citrus bark	1 A/day	69	363	58	Not feasible		
	0.039 lb ai/gal /750 ft2	1000 ft2	4620	24,200	3,879	Target MOE reached at PPE		
Low Pressure Handwand (13)	0.0417 lb ai/gal predominant max	40 gal/day	627	770	346	Target MOE reached at PPE		
	0.08 lb ai/gal bark beetle treatment	40 gal/day	330	396	180	Target MOE reached at PPE		
	0.03 lb ai/gal stump treatment	40 gal/day	869	1067	479	Target MOE reached at PPE		
	3.5 citrus bark	1 A/day	297	363	163	Target MOE reached at PPE		
	0.039 lb ai/gal/ 750 ft2 animal prem.	1000 ft2	19,800	24,200	10,890	Target MOE reached at PPE		
High Pressure Handwand (greenhouse uses) (14)	Min. 0.0033 lb ai/gal	1000 gal/day	73	97	41	Not feasible		
	Max. 0.0066 lb ai/gal		36	48	21	Not feasible		
Hydraulic Hand-held Sprayer for Bark Treatment (15)	3.5 citrus bark	10	18	110	15	Not feasible		
	0.08 lb ai/gal bark beetle treatment	1,000 gal/day	15	97	13	Not Feasible		
	0.039 lb ai/gal /750 ft2 animal prem	10000 ft2	2420	14,300	2070	Target MOE reached at PPE		

Exposure Scenario (Scenario#)	Application Rates (lb ai/acre)	Daily Acres Treated	Short-Term PPE MOEs			Short-Term Eng. Control MOEs		
			Dermal	Inhalation	Total	Dermal	Inhalation	Total
Dry Bulk Fertilizer Impregnation	1.0 lb ai / 200 lb fertilizer / acre	No Data	No Data			No Data		

The following scenarios are not of concern, i.e., MOEs are greater than 100, with PPE consisting of double layers, chemical resistant gloves, chemical resistant shoes plus socks, chemical resistant headgear for overhead exposure, chemical resistant apron when cleaning and mixing or loading and a dust/mist respirator:

(1b) Mixing/loading liquids for groundboom application (except at 3 lbs. ai/A sodfarm use)
(1c) Mixing/loading liquids for airblast application
(3b) Loading granulars for ground application
(7) Tractor drawn granular spreader
(13) Low pressure handwand

The following scenarios have MOEs greater than 100 with appropriate engineering controls:

(2b) Mixing wettable powder for groundboom application (water soluble packaging)
(2c) Mixing wettable powder for airblast application (water soluble packaging)
(4a) Aerial application of spray (enclosed cockpit)

The following occupational risk scenarios are still below the target MOE of 100, even with all feasible PPE or engineering controls.

Mixing/Loading Liquids for Aerial/Chemigation Application

The MOEs for mixing/loading liquids for aerial application (scenario 1a) are 66 and 100 depending on the application rate and the acres treated. The dermal route is driving the total MOE in this scenario (dermal MOEs range from 86 to 132 and the inhalation MOEs range from 272 to 408). Mixer/loaders for aerial application must use mechanical transfer systems for any container greater than 2.5 gallons for transfer of material from container to chemical holding tank. The registrant has agreed to reduce the rate on corn from 1.5 to 1 lb ai/A.

Aerial application is critical to large field crops such as cotton, wheat and sorghum. Ground application is not economically feasible. Approximately 200,000 lbs. ai of chlorpyrifos are applied per year to sorghum for control of greenbugs. Chlorpyrifos is the primary insecticide for foliar applications to wheat and is important for control of Russian wheat aphid, pale western cutworm and grasshoppers. Approximately100,000-150,000 lbs ai per year are applied to wheat.

For chemigation the MOEs will be higher than aerial application because the typical use rates are lower (0.5 to 1 lb ai/A) and the acres treated would typically average 40 to 80 acres. The combination of these lower rates and acres will increase the MOEs above 100.

Mixing/Loading Liquids for Groundboom Application to Sodfarms at 3 lbs. ai/A

The MOE for mixing/loading liquids for groundboom application to sodfarms at the 3 lbs. ai/A rate (scenario 1b) is 60. Currently enclosed mixing/loading is not required for the groundboom application to sodfarms. Dermal exposure contributes the most to the total MOE in this scenario (dermal MOE is 88 and the inhalation MOE is 193). The 3 lb. ai/A rate is used to control mole crickets and is mainly used as a patch application. Therefore, the 80 acres applied in a day is an overestimate for this particular use. The 2 lbs. ai/A rate is critical for the control of chinch bugs and lepidopterus (sod webworms, cutworms and army worms). Current PPE consists of double-layer clothing, chemical resistant gloves, chemical resistant shoes plus socks, chemical resistant headgear for overhead exposure, chemical resistant apron when cleaning and mixing or loading and a dust/mist respirator. Usage data are being required to confirm the acres treated per day for the 3 lbs. ai/A rate on sodfarms to control mole crickets, and will be used to refine risk estimates.

Mixing Wettable Powders for Aerial/Chemigation Application

The MOEs for mixing wettable powders in water soluble packaging (WSP) for aerial or chemigation application (scenario 2a) are 31 and 62, depending on the application rate the worker uses and the acres treated. EPA acknowledges the uncertainties associated with the risk assessment for WSP for aerial or chemigation application. Current WSP data in PHED are of low quality due to a limited number of replicates.

EPA believes the actual exposure from water soluble packaging in aerial/chemigation operations is less than predicted by the limited data in PHED. Confirmatory data will be required for the WSP formulation. These data may be developed in conjunction with the Agricultural Handler Task Force which has been formed between EPA and the industry to generate data to update PHED.

Loading Granulars for Aerial Application

The MOE for loading granulars for aerial application is 75 (scenario 3a). The inhalation route is driving the total MOE in this scenario (dermal MOE is 321 and the inhalation MOE is 99). Currently enclosed loading systems are not required for loading chlorpyrifos granulars for aerial application.

Because of new technology to reduce the dust and exposure from granular pesticides, EPA believes the actual exposure from loading granulars for aerial application is less than predicted by the limited data in PHED. Confirmatory data will be required for loading granulars. These data may be developed in conjunction with the Agricultural Handler Task Force which has been formed between EPA and the registrants to generate data to update PHED.

Aerially Applying Granulars

The MOE for aerially applying granulars is 51 (scenario 4b). The inhalation route is driving the total MOE in this scenario (dermal MOE is 686 and the inhalation MOE is 51). The inhalation data in PHED for this scenario is of low confidence because it lacks the sufficient replicates. The data in PHED for applying granulars is based on smaller acreage being treated. The pilot entered and left the plane after every 17-acre application. For chlorpyrifos where up to 350 acres are treated per day this would result in an overestimate because the pilot would not be entering and leaving the plane after every 17 acres. Information from aerial applicators indicate that entering and leaving the plane 3-4 times during the day is typical

EPA believes the actual exposure from applying granulars for aerial application is less than predicted by the limited data in PHED. Confirmatory data will be required for applying granulars. These data may be developed in conjunction with the Agricultural Handler Task Force which has been formed between EPA and the registrants to generate data to update PHED.

Airblast/Groundboom Application

The MOEs for airblast/groundboom application range from 121 to 850 depending on the application rate and acres treated and with the engineering control of an enclosed cab (scenario 5 and 6). A label statement is needed indicating that airblast applicators must wear double-layer clothing and a dust-mist respirator.

The available biological monitoring data for groundboom application was conducted with baseline PPE (one-layer of clothing) and are of minimal quality due to a low number of replicates. A label statement is needed indicating that groundboom applicators must wear double-layer clothing.

Confirmatory data will be required for groundboom application. These data may be developed in conjunction with the Agricultural Handler Task Force which has been formed between EPA and the registrants to generate data to update PHED.

Backpack Sprayer

Risks to mixer/loader/applicators using a backpack sprayer for bark beetle and pine seedling treatment (scenario 12) are of concern. For bark beetle treatment using 3.5 lbs. ai/A (for citrus bark), the MOE is 58; for other crops at 0.08 lbs. ai/gal, the MOE is 63; and for pine seedling treatment, the MOE is 31. These risk estimates are of low confidence because the data available lacked sufficient replicates to meet Agency guideline requirements.

Dermal exposure contributes most to the total MOE in this scenario. Dermal MOEs range from 37 to 75 while the inhalation MOEs range from 198 to 396. Confirmatory backpack exposure data are required and are being developed by the Forest Service (USDA) to refine

current risk estimates. The Agency has reviewed the study protocol and the study will be initiated in Spring of 2002.

The Forest Service has stated that chlorpyrifos is important in the control of bark beetles or borers and that no suitable alternative exists. Documentation from the Forest Service indicates that 40 gallons per day (as assumed in EPA's assessment) would rarely if ever be used for pine seedlings.

Since the *Human Health Risk Assessment* was conducted, product labels for this use were amended to add protection including double layers, chemical-resistant gloves, footwear and apron (for mixers and loaders). These protective measures will be required unless or until exposure data for this scenario are submitted and demonstrate otherwise.

High Pressure Handwand

Mixer/loader/applicator risks for use of the high-pressure handwand (scenario 14) are of concern, with MOEs of 41 and 21 depending on the application rate. These risk estimates are based on biological monitoring data but are of low confidence due to a lack of information on the types of sprayers and volumes used in the studies. In addition, the data lacked sufficient replicates to meet Agency guideline requirements. Comments from the American Nursery and Landscape Association indicate the EPA's assumption of 1,000 gallons per day of use are extremely unrealistic. Chlorpyrifos is used as a rotational tool to treat small blocks or areas of plant material–only to areas of the greenhouse that have infestation problems. Actual use is likely to be 100 gallons per day or less, and use is intermittent. Usage data are being required to confirm the current use per day. Additional information is required concerning the types of sprayers used. This information will be used to refine risk estimates.

Since the *Human Health Risk Assessment* was conducted, product labels for this use were amended to add protection including double layers, chemical-resistant gloves, footwear and apron (for mixers and loaders). These protective measures will be maintained unless or until exposure data for this scenario are submitted and demonstrate otherwise.

Hydraulic Handheld Sprayer

Risks to mixer/loader/applicators using a hydraulic handheld sprayer (scenario 15) are of concern. For application to citrus bark at 3.5 lbs./gal, the MOE is 15; for other crops at 0.08 lbs./gal, the MOE is 13. These risk estimates are of low confidence because the data lacked sufficient replicates. The driving factor in this assessment is the volume of spray estimated to be applied. Usage data are being required to confirm the actual amount of chlorpyrifos used on a daily and seasonal basis. Preliminary industry estimates report a high end usage of about 500 gallons a day, half of EPA's estimate assumed. Additional information is required concerning the types of sprayers used since EPA's assessment assumed a rights-of-way type sprayer. This information will be used to refine risk estimates. The Forest Service has stated that chlorpyrifos is important in the control of bark beetles or borers and that no suitable alternative exists.

Since the *Human Health Risk Assessment* was conducted, product labels for this use were amended to add protection including double layers, chemical-resistant gloves, footwear and apron (for mixers and loaders). A dust-mist respirator will also be necessary.

Dry Bulk Fertilizer Impregnation

Risks to mixer/loader/applicators for dry bulk fertilizer impregnation could not be assessed due to a lack of exposure data. This use is for the control of fire ants on orchard floors. For this use, dry fertilizer is placed in a closed rotary drum mixer equipped with suitable spraying equipment. Spray nozzles are positioned to provide uniform spray coverage of the tumbling fertilizer with chlorpyrifos.

This use is similar to mixing/loading liquids for groundboom application at the 1 pound rate (scenario 1b) and applying with a tractor drawn granular spreader (scenario 7). The MOEs are above 100 for both of these scenarios. Thus, EPA assumes that PPE for this use should be similar, i.e., double-layer clothing.

Seed Treatment

The Agency has no data at this time to assess the exposure for mixer/loaders and applicators for seed treatment. Seed treatment labels currently specify single-layer clothing, chemical-resistant footwear over socks, chemical-resistant gloves and respirators. The Agency does not anticipate that the exposures for this use with the prescribed PPE will be any greater than for mixer/loaders of wettable powders for groundboom application with engineering controls (MOEs 200-400), and the amount of ai handled per day is likely to be less. Therefore, this use is eligible for reregistration and confirmatory data are required. This protective equipment must be maintained on the labels until/unless exposure data indicate that less PPE is appropriate.

Preplant Peach Dip

The Agency has no specific data at this time to assess the exposure for mixer/loaders and applicators for the preplant peach dip. Labels for the preplant peach dip currently require double-layer clothing, chemical-resistant gloves, chemical-resistant shoes plus socks, protective eyewear, chemical-resistant headgear for overhead exposure, chemical-resistant apron when cleaning equipment and mixing or loading and a respirator. The Agency does not anticipate that exposures for this use will be any greater than for mixer/loaders of liquids for citrus and fruit ground applications (MOEs 100-150) and the amount of ai handled per day is likely to be less. Confirmatory data are required. Therefore, this use is eligible for reregistration and confirmatory data are required. This protective equipment must be maintained on the label until/unless exposure data indicate that less PPE is appropriate.

Flaggers

Risks to flaggers involved in spray applications (scenarios 10 and 11) are of concern with use of PPE, with MOEs of 49 and 82. Information from USDA indicates that human flagging is no longer necessary in modern agriculture. Therefore, a prohibition against human flagging will mitigate these risks with minimum impact on current production practices.

Taking into account the strengths and weaknesses of the risk assessment and the benefits of chlorpyrifos use, EPA has determined that the uses listed above are eligible for reregistration with the designated mitigation and confirmatory data.

2) Agricultural and Ornamental/Greenhouse Postapplication Risks

The results of the short- and intermediate-term postapplication assessments indicate that REIs need to be established. The REIs range from 24 hours for most crops to 5 days for citrus trees. REIs and pre-harvest intervals (PHIs) are needed to ensure that risks are not of concern are shown below in Table 32.

Table 32. Restricted Entry Intervals and Preharvest Intervals

Crop	REI	MOEs	PHI
Cauliflower	3 days	150	21-30 days
Nut trees	24 hours	270	14 days
Potatoes	24 hours	750	7 days
Citrus trees	5 days	220	21 days
Fruit trees	4 days	280	21 days
Sweet corn	24 hours	83	7 days
All other crops	24 hours	110	7 days

In addition to the foliar chlorpyrifos treatments, there are many soil incorporated/directed treatments to field crops and citrus. At this time, there are insufficient exposure and soil residue data to assess the potential risk from soil incorporated/directed uses of chlorpyrifos. However, these treatments are expected to result in less postapplication exposure than the foliar treatments. Confirmatory data for soil directed/incorporated uses are required.

Postapplication risks to greenhouse/nursery workers were not assessed due to a lack of data. Information is needed concerning the timing of the applications in relation to the

postapplication activities and a lack of residue data (foliar and bark treatments) to assess the REIs for the ornamental/greenhouse uses. These risks are of concern for activities such as pruning, transplanting and burlap/balling. The National Agricultural Pesticide Impact Assessment Program (NAPIAP 1996) reports chlorpyrifos is widely used for a broad range of insect applications including wood-boring, foliage feeding, sucking and soil-borne pests. NAPIAP also reports that although chlorpyrifos use represents only 5% of the total lbs. ai used in greenhouse/nursery operations, it is used by 35% of their survey respondents. Chlorpyrifos is an important chemical for the industry, especially as a tool for resistance management. Additional use information, i.e., timing of application relative to postapplication activities, greenhouse DFR data, and biological monitoring data to develop transfer coefficients for various greenhouse/nursery activities are required.

The current REI of 24 hours was established by the MOA of June 2000 and remains in effect until acceptable data indicate that it should be changed.

3) Non-Agricultural Occupational Handler Risks

Risk estimates for the application of a dust product for fire ant control are of concern. With PPE, the short-term MOEs are 4.3 to 108; intermediate-term MOEs are 0.9 to 22. These MOEs are based on one literature study, which did not include inhalation exposure data; therefore, the MOEs are likely to underestimate actual risk. This use is ineligible for reregistration at this time. Since this product is used to control fire ants and may have public health benefits, registrants and other interested parties may provide benefits and usage information and mitigation suggestions during the comment period.

Application by groundboom to golf course turf is of concern. Using baseline PPE, the short-term MOE is 60. A label statement is needed indicating that groundboom applicators must be in fully enclosed cabs or, if not in fully enclosed cabs, applicators must wear double-layer clothing, chemical-resistant footwear and socks, and a dust-mist respirator.

4) Non-Agricultural Occupational Postapplication Risks

Occupational postapplication exposures by commercial operators in the residential setting (termiticide and mosquito adulticide uses) are not expected to occur. For golf course workers, postapplication exposures are not of concern.

c. Residential Risk Mitigation

1) Residential Handler Risk

The only products that can be applied by a resident are the containerized baits in child-resistant packaging. This is not expected to result in exposures of concern. All other residential uses have been canceled.

2) Residential Postapplication Risk

Residential postapplication exposures may occur after termiticide use in residential structures. To mitigate risks from this use, the technical registrants agreed in June 2000 to limit termiticide treatments to 0.5% solution, and cancel all postconstruction uses. Pre-construction use will remain until 2005, unless acceptable exposure data are submitted that show that residential postapplication risks from this use are not a concern.

Chlorpyrifos treatments to processed wood products was maintained in the Memorandum of Agreement of June, 2000. Since that time, it has come to the Agency's attention that some wood products such as window frames and floor joists that are treated are eventually used in homes. Exposure data are required to confirm that this use is not a concern.

3. Environmental Risk Mitigation

The technical registrants have agreed to the following label amendments to address environmental risk concerns. The amendments include the use of buffer zones to protect water quality, fish and wildlife, reductions in application rates, number of applications per season, seasonal maximum amounts applied, and increases in the minimum intervals for retreatment.

The mitigation measures prescribed in this IRED along with mitigation that is already being implemented as a result of the June, 2000, Memorandum of Agreement, will reduce risk to both terrestrial and aquatic species. For example, many of the reported incidents of wildlife mortality associated with chlorpyrifos use were related to residential lawn and termite uses and use on golf courses. The residential uses have been eliminated, the termiticide use is being phased out, and the application rate on golf courses has been reduced from 4 to 1 lb/ai/A. Additionally, no-spray buffers around surface water bodies, as well as rate reductions for agricultural uses will be implemented as a result of this IRED and will further reduce the environmental burden of chlorpyrifos.

Although the magnitude of the risk reduction cannot be precisely quantified, EPA's recalculation of risk quotients, taking into account new use restrictions, indicates that the potential risk to invertebrates, particularly estuarine invertebrates may still be of concern. Risk quotients represent a screening level assessment and are inadequate to predict whether the levels of chlorpyrifos entering estuarine areas are sufficient to affect invertebrate populations or

populations of the larger species that depend on them as a food source. Monitoring for chlorpyrifos in waters that feed into estuaries would provide useful information on the magnitude and frequency of actual residues.

Taking into account the extensive mitigation already underway, additional mitigation to be adopted as a result of this IRED, as well as the benefits of chlorpyrifos use, EPA finds the remaining risk to non-target species is not unreasonable. Because the use of chlorpyrifos will be declining over the next few years as existing stocks of canceled products are exhausted, EPA expects that levels of chlorpyrifos in the environment will also be reduced. In order to confirm that levels of chlorpyrifos in the aquatic environment are declining, EPA is requiring updated usage information and collection of water monitoring data for the areas of greatest remaining chlorpyrifos use.

The following crop-specific mitigation will be needed to address environmental risk concerns:

Alfalfa (liquid formulations)

The maximum number of applications per season will be reduced from 8 to 4.

Citrus (liquid formulations)

The maximum number of applications per season will be limited to 2; the maximum application rate of 6 lbs. ai/A will be limited to five counties in California (Fresno, Tulare, Kern, Kings, and Madera); the minimum interval for retreatment will be 30 days. The 6 lbs. ai/A rate is for ground application only. Sprays must be directed toward the canopy.

Citrus orchard floors (granular formulations)

The maximum number of applications per season will be reduced from 10 to 3; the maximum amount applied per season will be reduced from 10 lbs. ai/A to 3 lbs. ai/A.

Corn, field, sweet and seed (liquid formulations)

The maximum number of applications per season will be limited to 3; the maximum amount applied per season will be reduced from 7.5 lbs. ai/A to 3 lbs. ai/A.

Corn, field, sweet and seed (granular formulations)

The maximum number of applications per season will be limited to 2; the maximum amount applied per season will be limited to 2 lbs. ai/A.

Cotton (liquid formulations)

The maximum number of applications per season will be reduced from 6 to 3; the maximum amount applied per season will be reduced from 6 lbs. ai/A to 3 lbs. ai/A.

Peanuts (granular formulations)

Aerial application will be eliminated.

Sorghum (liquid formulations)

The maximum number of applications per season will be limited to 3; it was previously unspecified.

Soybeans (liquid formulations)

The maximum number of applications per season will be limited to 3; it was previously unspecified.

Sugar beets (liquid formulations)

The maximum number of applications per season will be reduced from 4 to 3; the maximum amount applied per season will be reduced from 4 lbs. ai/A to 3 lbs. ai/A.

Sugar beets (granular formulations)

The maximum number of applications per season, previously unspecified, will be limited to 3; the maximum amount applied per season will be reduced from 13.5 lbs. ai/A to 3 lbs. ai/A.

Sunflowers (liquid formulations)

The maximum number of applications per season, previously unspecified, will be limited to 3; the maximum amount applied per season will be reduced from 4.5 lbs. ai/A to 3 lbs. ai/A.

Tobacco (liquid formulations)

The maximum number of applications per year will be limited to 1; the application rate of 5 lbs. ai/A for root-knot nematodes in North Carolina, South Carolina, and Virginia will be eliminated; the maximum amount applied per season will be reduced from 1.5 lbs. ai/A to 1 lb. ai/A.

Tree nuts (liquid formulations)

The maximum amount applied per season will be reduced from 8 lbs. ai/A to 4 lbs. ai/A.

Walnut and almond orchard floors (liquid formulations):

The maximum amount applied per season will be reduced from 8 lbs. ai/A to 4 lbs. ai/A; the maximum number of applications per season, previously unspecified, will be limited to 2.

All crops

Spray drift warnings and no-spray zones will be included on labels, as shown in Table 33. These no-spray zones will apply to rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries and commercial fish ponds. For more information on spray drift management language, please see section 4. Other Labeling, subsection b. Spray Drift Management.

Table 33. Proposed No-Spray Buffer Zones around Water Bodies

Application Method	Required Setback (No-spray Zone)
Ground Boom	25 feet
Chemigation	25 feet
Orchard Airblast	50 feet
Aerial (fixed-wing or helicopter)	150 feet

Table 34 summarizes the range of risk quotients for major use sites taking into account the mitigation measures outlined above.

Table 34. Risk Quotients for Corn, Citrus, Cotton and Tobacco With Proposed Risk Mitigation

Species	Range of Risk Quotients
Freshwater Fish Acute LC_{50}	2.8 - 11
Fish Reproduction NOAEC	8.9 -36 [1] 5.4 - 46 [2]
Aquatic Invertebrate Acute LC_{50}	51 - 210
Freshwater Invert. Reproduction NOAEC	130 520 [1] 65 - 230 [2]
Estuarine Fish Acute LC_{50}	5.3 - 22
Estuarine Fish Reproduction NOAEC	11 - 74 [1] 9.3 - 20 [2]
Estuarine Invertebrate Acute LC_{50}	110 - 590

Estuarine Invert. Reproduction NOAEC	>1100 [1]
Estuarine Algae EC_{50}	0.036 - 0.15

[1] Peak EECs in 2-meter deep pond or estuarine water
[2] 21-day EECs in 2-meter deep pond or estuarine water

4. Other Labeling

In order to remain eligible for reregistration, other use and safety information needs to be placed on the labeling of all end-use products containing chlorpyrifos. For the specific labeling statements, refer to Section V of this document

a. Endangered Species Statement

The Agency has developed the Endangered Species Protection Program to identify pesticides whose use may cause adverse impacts on endangered and threatened species, and to implement mitigation measures that address these impacts. The Endangered Species Act requires federal agencies to ensure that their actions are not likely to jeopardize listed species or adversely modify designated critical habitat. To analyze the potential of registered pesticide uses to affect any particular species, EPA puts basic toxicity and exposure data developed for REDs into context for individual listed species and their locations by evaluating important ecological parameters, pesticide use information, the geographic relationship between specific pesticides uses and species locations, and biological requirements and behavioral aspects of the particular species. This analysis will take into consideration any regulatory changes recommended in this RED that are being implemented at that time. A determination that there is a likelihood of potential impact to a listed species may result in limitations on use of the pesticide, other measures to mitigate any potential impact, or consultations with the Fish and Wildlife Service and/or the National Marine Fisheries Service as necessary.

The Endangered Species Protection Program as described in a Federal Register notice (54 FR 27984-28008, July 3, 1989) is currently being implemented on an interim basis As part of the interim program, the Agency has developed County Specific Pamphlets that articulate many of the specific measures outlined in the Biological Opinions issued to date. These Pamphlets are available for voluntary use by pesticide applicators, on EPA's web site at www.epa.gov/espp. A final Endangered Species Protection Program, which may be altered from the interim program, is scheduled to be proposed for public comment in the Federal Register before the end of 2001.

b. Spray Drift Management

The Agency is in the process of developing more appropriate label statements for spray and dust drift control to ensure that public health and the environment are protected from unreasonable adverse effects. In August 2001, EPA published draft guidance for label statements in a pesticide registration (PR) notice ("Draft PR Notice 2001-X"

http://www.epa.gov/PR Notices/#2001). A Federal Register notice was published on August 22, 2001 (http://www.epa.gov/fedrgstr) Announcing the availability of this draft guidance for a 90-day public comment period. After receipt and review of the comments, the Agency will publish final guidance in a PR notice for registrants to use when labeling their products.

Until EPA decides upon and publishes the final label guidance for spray and dust drift, registrants (and applicants) may choose to use the statements proposed in the draft PR notice. Registrants should refer to and read the draft PR notice to obtain a full understanding of the proposed guidance and its intended applicability, exemptions for certain products, and the Agency's willingness to consider other versions of the statements.

For purposes of complying with the deadlines for label submission outlined in this document, registrants (and applicants) may elect to adopt the appropriate sections of the proposed language below, or a version that is equally protective, for their end-use product labeling.

For products as liquids:

"Do not allow spray to drift from the application site and contact people, structures people occupy at any time and the associated property, parks and recreation areas, nontarget crops, aquatic and wetland areas, woodlands, pastures, rangelands or animals."

"For ground boom applications, apply with nozzle height no more than 4 feet above the ground or crop canopy, and when wind speed is 10 mph or less at the application site as measured by an anemometer. Use _____ (registrant to fill in blank with spray quality, e.g. fine or medium) or coarser spray according to ASAE 572 definition for standard nozzles or VMD for spinning atomizer nozzles."

"For orchard and vineyard airblast applications, do not direct spray above trees and vines, and turn off outward pointing nozzles at row ends and outer rows. Apply only when wind speed is 3 -10 mph at the application site as measured by an anemometer outside of the orchard or vineyard on the upwind side."

"For aerial applications, the boom width must not exceed 75% of the wingspan or 90% of the rotary blade. Use upwind swath displacement, and apply only when wind speed is 3 - 10 mph as measured by an anemometer. Use _____ (registrant to fill in blank with spray quality, e.g. fine or medium) or coarser spray according to ASAE 572 definition for standard nozzles or VMD for spinning atomizer nozzles. If application includes a no-spray zone, do not release spray at a height greater than 10 feet above the ground or the crop canopy."

For hand-applied products, to be applied as sprays:

"Do not allow spray to drift from the application site, and contact people, structures people occupy at any time, and the associated property, parks and recreation areas, nontarget crops, aquatic and wetland areas, woodlands, pastures, rangelands, or animals. Apply only when wind speed is not more than 10 mph. For sprays, apply largest size droplets possible."

Alternatively, registrants may elect to use the following language, which is the current Agency policy on drift labeling. For products that are applied outdoors in liquid sprays (except mosquito adulticides), regardless of application method:

"Do not allow this product to drift."

The Agency recognizes that the above option does not address other application types. Registrants may therefore wish to adapt some variation of the old, and proposed new language for their particular products, depending on their application methods.

V. What Registrants Need to Do

In order to be eligible for reregistration, registrants need to implement the risk mitigation measures outlined in Section IV and V, which include, among other things, submission of the following:

For chlorpyrifos technical grade active ingredient products, registrants need to submit the following items.

Within 90 days from receipt of the generic data call-in (DCI):

(1) completed response forms to the generic DCI (i.e., DCI response form and requirements status and registrant's response form); and

(2) submit any time extension and/or waiver requests with a full written justification.

Within the time limit specified in the generic DCI:

(1) Cite any existing generic data which address data requirements or submit new generic data responding to the DCI.

Please contact Tom Myers at 703/308-8589 with questions regarding generic reregistration and/or the DCI. All materials submitted in response to the generic DCI should be addressed:

By US mail:
Document Processing Desk (DCI/SRRD)
Chemical Review Manager's Name
US EPA (7508C)
1200 Pennsylvania Ave., NW
Washington, DC 20460

By express or courier service:
Document Processing Desk (DCI/SRRD)
Chemical Review Manager's Name
Office of Pesticide Programs (7508C)
Room 266A, Crystal Mall 2
1921 Jefferson Davis Highway
Arlington, VA 22202

For products containing the active ingredient chlorpyrifos, registrants need to submit the following items for each product.

Within 90 days from the receipt of the product-specific data call-in (PDCI):

(1) Complete response forms to the PDCI (i.e., PDCI response form and requirements status and registrant's response form); and

(2) Submit any time extension or waiver requests with a full written justification.

For all products that have agricultural uses, items 1 through 5, listed below, are required to be submitted to the Agency within 45 days of receipt of the PDCI. Item number 6, the product specific data, is required within eight months from the receipt of the PDCI.

Within eight months from the receipt of the PDCI:

(1) Two copies of the confidential statement of formula (EPA Form 8570-4);

(2) A completed original application for reregistration (EPA Form 8570-1). Indicate on the form that it is an "application for reregistration";

(3) Five copies of the draft label incorporating all label amendments outlined in Table 35 of this document;

(4) A completed form certifying compliance with data compensation requirements (EPA Form 8570-34);

(5) If applicable, a completed form certifying compliance with cost share offer requirements (EPA Form 8570-32); and

(6) The product-specific data responding to the PDCI.

Please contact Venus Eagle at (703)308-8045 with questions regarding product reregistration and/or the PDCI. All materials submitted in response to the PDCI should be addressed:

By US mail:
Document Processing Desk (PDCI/PRB)
Chemical Review Manager's Name
US EPA (7508C)
1200 Pennsylvania Ave., NW
Washington, DC 20460

By express or courier service only:
Document Processing Desk (PDCI/PRB)
Chemical Review Manager's Name
Office of Pesticide Programs (7508C)
Room 266A, Crystal Mall 2
1921 Jefferson Davis Highway
Arlington, VA 22202

A. Manufacturing Use Products

1. Additional Generic Data Requirements

The generic data base supporting the reregistration of chlorpyrifos for the above eligible uses has been reviewed and determined to be substantially complete. The following data gaps remain:

Product Chemistry Data requirements for the TGAI and Manufacturing-Use Products.

830.1550 (formerly 61-1)	Product Identity and Disclosure of Ingredients
830.1600 (formerly 61-2a)	Starting Materials and Manufacturing Process
830.1670 (formerly 61-2b)	Discussion of Formation of Impurities
830.1700 (formerly 62-1)	Preliminary Analysis
830.1750 (formerly 62-2)	Certification of Limits
830.1800 (formerly 62-3)	Analytical Method
830.6302 (formerly 63-2)	Color
830.6303 (formerly 63-3)	Physical State
830.6304 (formerly 63-4)	Odor
830.7200 (formerly 63-5)	Melting Point
830.7300 (formerly 63-7)	Density, Bulk Density or Specific Gravity
830.7840 and 830.7860 (formerly 63-8)	Solubility
830.7950 (formerly 63-9)	Vapor Pressure
830.7550 (formerly 63-11)	Octanol/Water Partition Coefficient
830.6313 (formerly 63-13)	Stability
830.6316 (formerly 63-16)	Explodability
830.6317 (formerly 63-17)	Storage Stability
830.6320 (formerly 63-20)	Corrosion Characteristics

Residue chemistry data requirements.

860.1500 (formerly 171-4k)	Magnitude of the residue in corn fodder and forage
860.1500 (formerly 171-4k)	Magnitude of the residue in cotton gin by-products
860.1500 (formerly 171-4k)	Magnitude of the residue in clover and grasses

860.1500 (formerly 171-4k)	Magnitude of the residue in aspirated grain fractions of sorghum, soybeans and wheat
860.1500 (formerly 171-4k)	Magnitude of the residue in cherries

Other data requirements:

875.1100 and 875.1300	Exposure data for seed treatment uses.
875.1100 and 875.1300	Exposure data for dip applications (e.g., preplant peaches).
875.1100 and 875.1300	Exposure data for mixing wettable powders for aerial/chemigation application.
875.1100 and 875.1300	Exposure data for loading and applying granulars for aerial application.
875.1100 and 875.1300	Exposure data for groundboom application.
875.1100 and 875.1300	Exposure data for backpack spray application.
875.1100 and 875.1300	Exposure data for reentry into treated areas with soil incorporated/directed applications.
875.2100 (formerly 132-1a)	Dislodgeable foliar residues on ornamentals in greenhouses.
233 and 234	Risk Assessment data for treated wood in residential structures.
810.1000 (formerly 90-1)	Use pattern information for hydraulic handheld spray applications (amounts handled per day, per season; types of sprayers used).
810.1000 (formerly 90-1)	Use pattern information for high pressure hand-wand spray applications (amounts handled per day, per season; types of sprayers used).
810.1000 (formerly 90-1)	Use pattern information, i.e., timing of application relative to postapplication activities, greenhouse DFR data, and biological monitoring data to develop transfer coefficients for various greenhouse/nursery activities are required.
810.1000 (formerly 90-1)	Usage data to confirm the acres treated for the 3 lb/A on sodfarms for mole crickets.

Summarize water monitoring data to confirm reduction of residue levels in surface water.

Also, a Data Call-In Notice (DCI) was sent to registrants of organophosphate pesticides currently registered under FIFRA (August 6, 1999 64FR42945-42947, August 18 64FR44922-44923). DCI requirements included acute, subchronic, and developmental neurotoxicity studies.

2. Labeling for Manufacturing Use Products

To remain in compliance with FIFRA, manufacturing use product (MUP) labeling should be revised to comply with all current EPA regulations, PR Notices and applicable policies. The MP labeling should bear the labeling contained in Table 38 at the end of this section.

B. End-Use Products

1. Additional Product-Specific Data Requirements

Section 4(g)(2)(B) of FIFRA calls for the Agency to obtain any needed product-specific data regarding the pesticide after a determination of eligibility has been made. Registrants must review previous data submissions to ensure that they meet current EPA acceptance criteria and if not, commit to conduct new studies. If a registrant believes that previously submitted data meet current testing standards, then the study MRID numbers should be cited according to the instructions in the Requirement Status and Registrants Response Form provided for each product.

A product-specific data call-in, outlining specific data requirements, accompanies this interim RED.

2. Labeling for End-Use Products

Labeling changes are necessary to implement the mitigation measures outlined in Section IV above. Specific language for these changes is specified in the Table 35.

C. Existing Stocks

Registrants may generally distribute and sell products bearing old labels/labeling for 26 months from the date of the issuance of this Interim Reregistration Eligibility Decision document. Persons other than the technical registrants may generally distribute or sell such products for 50 months from the date of the issuance of this interim RED. However, existing stocks time frames will be established case-by-case, depending on the number of products involved, the number of label changes, and other factors. Refer to "Existing Stocks of Pesticide Products; Statement of Policy"; *Federal Register*, Volume 56, No. 123, June 26, 1991.

The Agency has determined that registrant may distribute and sell chlorpyrifos products bearing old labels/labeling for 26 months from the date of issuance of this interim RED. Persons other than the technical registrants may distribute or sell such products for 50 months from the date of the issuance of this interim RED. Registrants and persons other than the technical registrants remain obligated to meet pre-existing label requirements and existing stocks requirements applicable to products they sell or distribute.

D. Labeling Changes Summary Table

In order to be eligible for reregistration, amend all product labels to incorporate the risk mitigation measures outlined in Section IV. Table 35 describes how language on the labels should be amended.

Table 35. Summary of Labeling Changes for Chlorpyrifos

Description	Amended Labeling Language	Placement on Label
Manufacturing Use Products		
One of these statements may be added to a label to allow reformulation of the product for a specific use or all additional uses supported by a formulator or user group	"Only for formulation into an ***insecticide*** for the following use(s) [fill blank only with those uses that are being supported by MP registrant]."	Directions for Use
	"This product may be used to formulate products for specific use(s) not listed on the MP label if the formulator, user group, or grower has complied with U.S. EPA submission requirements regarding support of such use(s)." Or "This product may be used to formulate products for any additional use(s) not listed on the MP label if the formulator, user group, or grower has complied with U.S. EPA submission requirements regarding support of such use(s)."	Directions for Use
Environmental Hazards Statements Required by the RED and Agency Label Policies	This pesticide is toxic to birds and wildlife, and extremely toxic to fish and aquatic organisms. Do not discharge effluent containing this product into lakes, streams, ponds, estuaries, oceans or other waters unless in accordance with the requirements of a National Pollutant Discharge Elimination System (NPDES) permit and the permitting authority has been notified in writing prior to discharge. Do not discharge effluent containing this product to sewer systems without previously notifying the local sewage treatment plant authority. For guidance, contact your State Water Board or Regional Office of the EPA.	Directions for Use
End Use Products Intended for Occupational Use Products That Have Worker Protection Standard (WPS) Uses Only or Both WPS and Non WPS Uses on Same Label		
Handler PPE requirements (all formulations)	Note the following information when preparing labeling for all end use products: For **sole-active-ingredient** end-use products that contain chlorpyrifos, the product label must be revised to adopt the handler personal protective equipment (PPE)/engineering control requirements set forth in this section. Any conflicting PPE requirements on the current label must be removed.	

Description	Amended Labeling Language	Placement on Label
	For **multiple-active-ingredient** end-use products that contain chlorpyrifos, the handler PPE/engineering control requirements set forth in this section must be compared with the requirements on the current label, and the more protective language must be retained. For guidance on which requirements are considered to be more protective, see PR Notice 93-7. PPE that is established on the basis of Acute Toxicity testing with the end-use products must be compared with the active ingredient PPE specified below in this document. The more protective PPE must be placed in the product labeling. For example, the Handler PPE in this RED does not require protective eyewear which may be required by the Acute Toxicity testing for the end-use product. For guidance on which PPE is considered more protective, see PR Notice 93-7.	
Handler PPE requirements for liquid formulation packaged in containers holding more than 2.5 gallons.	"Personal Protective Equipment (PPE) Some materials that are chemical-resistant to this product are [registrant inserts correct material]. For more information, following instructions in Supplement Three of PR Notice 93-7. If you want more options, follow the instructions for category [insert A,B,C,D,E,F,G or H] on an EPA chemical-resistance category selection chart." "Mixers and loaders using a mechanical transfer loading system and applicators using aerial application equipment must wear: - long sleeved shirt and long pants; - socks and shoes. In addition to the above, mixers and loaders using a mechanical transfer loading system must wear: - chemical resistant gloves; - chemical resistant apron; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter. See engineering controls for additional requirements	Immediately following/below Precautionary Statements: Hazards to Humans and Domestic Animals

Description	Amended Labeling Language	Placement on Label
	All other mixers, loaders, applicators and handlers must wear: - coveralls over long-sleeved shirt and long pants; - chemical-resistant gloves; - chemical resistant apron when mixing or loading or exposed to the concentrate; - chemical-resistant footwear plus socks; - chemical-resistant headgear for overhead exposures; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter. *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	
Handler PPE requirements for liquid formulation packaged in containers holding 2.5 gallons or less.	"Personal Protective Equipment (PPE) Some materials that are chemical-resistant to this product are" [registrant inserts correct material]. "For more information, following instructions in Supplement Three of PR Notice 93-7. If you want more options, follow the instructions for category [insert A,B,C,D,E,F,G or H] on an EPA chemical-resistance category selection chart." All mixers, loaders, other applicators and other handlers must wear: - coveralls over long-sleeved shirt and long pants; - chemical-resistant gloves; - chemical resistant apron when mixing or loading or exposed to the concentrate; - chemical-resistant footwear plus socks; - chemical-resistant headgear for overhead exposures; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter. *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	

Description	Amended Labeling Language	Placement on Label
Handler PPE requirements for wettable powder formulations. (wettable powder formulations must be in water-soluble packaging to be eligible for reregistration)	"Personal Protective Equipment (PPE) Some materials that are chemical-resistant to this product are" [registrant inserts correct material]. " For more information, following instructions in Supplement Three of PR Notice 93-7. If you want more options, follow the instructions for category [insert A,B,C,D,E,F,G or H] on an EPA chemical-resistance category selection chart." "Mixers and loaders must wear: - long-sleeved shirt and long pants; - socks and shoes; - chemical resistant gloves; - chemical resistant apron. Applicators using aerial application equipment must wear: - long-sleeved shirt and long pants; - socks and shoes. See engineering controls for additional requirements. All other handlers must wear: - coveralls over long-sleeved shirt and long pants; - chemical-resistant gloves; - chemical resistant apron when mixing or loading; - chemical-resistant footwear plus socks; - chemical-resistant headgear for overhead exposures; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter. *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	Immediately following/below Precautionary Statements: Hazards to Humans and Domestic Animals

Description	Amended Labeling Language	Placement on Label
Handler PPE requirements for granular products	"Personal Protective Equipment (PPE) Some materials that are chemical-resistant to this product are" [registrant inserts correct material]. "For more information, following instructions in Supplement Three of PR Notice 93-7. If you want more options, follow the instructions for category [insert A,B,C,D,E,F,G or H] on an EPA chemical-resistance category selection chart." "Loaders, applicators and all other handlers must wear: - coveralls over long-sleeved shirt and long pants; - chemical-resistant gloves; - chemical-resistant footwear plus socks; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter.	Immediately following/below Precautionary Statements: Hazards to Humans and Domestic Animals
User Safety Requirements	"Follow manufacturer's instructions for cleaning/maintaining PPE. If no such instructions for washables exist, use detergent and hot water. Keep and wash PPE separately from other laundry." "Discard clothing and other absorbent materials that have been drenched or heavily contaminated with this product's concentrate. Do not reuse them." (*This second statement is not required for granular formulations*)	Precautionary Statements: Hazards to Humans and Domestic Animals immediately following the PPE requirements

Description	Amended Labeling Language	Placement on Label
Engineering Controls required for liquid formulations packaged in containers holding more than 2.5 gallons.	"Engineering Controls" "Mixers and loaders supporting aerial applications must use a mechanical transfer system that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides [40 CFR 170.240(d)(4)] for dermal protection, and must: -- wear the personal protective equipment required above for mixers/loaders, -- wear protective eyewear if the system operates under pressure, and -- be provided and have immediately available for use in an emergency, such as a broken package, spill, or equipment breakdown: coveralls, chemical resistant footwear and chemical resistant headgear if overhead exposure." "Pilots must use an enclosed cockpit in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides [40 CFR 170.240(d)(6)]." "Use of human flaggers is prohibited. Mechanical flagging equipment must be used." "When handlers use closed cab motorized ground application equipment in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides (40 CFR 170.240(d)(4-6), the handler PPE requirements may be reduced or modified as specified in the WPS."	Precautionary Statements: Hazards to Humans and Domestic Animals (Immediately following PPE and User Safety Requirements.)
Engineering Controls for liquid formulations packaged in containers less than 2.5 gallons.	"Engineering Controls" "When handlers use closed systems or closed cab motorized ground application equipment in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides (40 CFR 170.240(d)(4-6), the handler PPE requirements may be reduced or modified as specified in the WPS."	Precautionary Statements: Hazards to Humans and Domestic Animals (Immediately following PPE and User Safety Requirements.)

Description	Amended Labeling Language	Placement on Label
Engineering controls for wettable powder formulations	"Engineering Controls" "Water-soluble packets, when used correctly, qualify as a closed mixing/loading system under the Worker Protection Standard (WPS) for Agricultural Pesticides [40 CFR 170.240(d)(4)]. Mixers and loaders using water-soluble packets must wear the PPE required above for mixer/loaders, and have immediately available for use in emergency (such as a broken package, spill or equipment breakdown) additional PPE. These PPE include coveralls and chemical-resistant footwear and a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter." "Pilots must use an enclosed cockpit in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides [40 CFR 170.240(d)(6)]." "Use of human flaggers is prohibited. Mechanical flagging equipment must be used." "When applicators use closed cab motorized ground equipment in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides (40 CFR 170.240(d)(4-6), the handler PPE requirements may be reduced or modified as specified in the WPS." *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	Precautionary Statements: Hazards to Humans and Domestic Animals (Immediately following PPE and User Safety Requirements.)
Engineering controls for Granular formulations	"Engineering Controls" "Pilots must use an enclosed cockpit in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides [40 CFR 170.240(d)(6)]." "When applicators use closed cab equipment in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides (40 CFR 170.240(d)(4-6), the handler PPE requirements may be reduced or modified as specified in the WPS."	Precautionary Statements: Hazards to Humans and Domestic Animals (Immediately following PPE and User Safety Requirements.)

Description	Amended Labeling Language	Placement on Label
User Safety Recommendations	"User Safety Recommendations" "Users should wash hands before eating, drinking, chewing gum, using tobacco or using the toilet." "Users should remove clothing/PPE immediately if pesticide gets inside. Then wash thoroughly and put on clean clothing." "Users should remove PPE immediately after handling this product. Wash the outside of gloves before removing. As soon as possible, wash thoroughly and change into clean clothing."	Precautionary Statements immediately following the Engineering Controls
Environmental Hazards	"Environmental Hazards" "This pesticide is toxic to fish, aquatic invertebrates, small mammals and birds. Do not apply directly to water, or to areas where surface water is present or to intertidal areas below the mean high water mark. Drift and runoff may be hazardous to aquatic organisms in water adjacent to treated areas. Do not contaminate water when disposing of equipment wash water or rinsate. This product is highly toxic to bees exposed to direct treatment or residues on blooming crops or weeds. Do not apply this product or allow it to drift to blooming crops or weeds if bees are visiting the treatment area."	Precautionary Statements immediately following the User Safety Recommendations
Restricted-Entry Interval	"Do not enter or allow entry into treated areas during the restricted entry interval (REI). The REI for each crop is listed in the directions for use associated with each crop"	Directions for Use, Agricultural Use Requirements Box
WPS Restricted Entry Intervals (REI)	The Directions for Use must be amended to reflect the following REI: The REI for all crops except those listed below is 24 hours cauliflower: 3 days citrus trees: 5 days fruit trees: 4 days	Directions for Use Under Application Instructions for Each Crop

Description	Amended Labeling Language	Placement on Label
Early Re-entry Personal Protective Equipment established by the RED.	"PPE required for early entry into treated areas that is permitted under the Worker Protection Standard and involves contact with anything that has been treated, such as plants, soil, or water, is: Coveralls over short sleeved shirt and shirt pants; Chemical resistant gloves made out of any waterproof material; Chemical resistant footwear plus socks; Chemical Resistant headgear for over head exposures." "Notify workers of the application by warning them orally and by posting warning signs at entrances to treated areas."	Directions for Use, Agricultural Use Requirements Box
Entry Restrictions for products applied as sprays that have Non-WPS uses on the label	"Do not enter or allow others to enter until sprays have dried"	Directions for Use in the Non-Agricultural Use Requirements Box.
General Application Restrictions	"Do not apply this product in a way that will contact workers or other persons, either directly or through drift. Only protected handlers may be in the area during application." Labels must be amended to reflect the following application restrictions which supercede or are in addition to restrictions currently on labels: **Preharvest interval restrictions:** All crops 7 days except: cauliflower: 21-30 days nut trees: 14 days citrus trees: 21 days fruit trees: 21 days **Aerial application restrictions:** All formulations: "Aerial application to peanuts is prohibited." Granular formulations: "Do not apply by aircraft at a rate greater than 1 lb. ai/A."	Place in the Direction for Use

Description	Amended Labeling Language	Placement on Label
	Maximum application rates for a single application: - golf course turf : 1 lb. ai/A - citrus: 4 lbs. ai/A, except in Fresno, Tulare, Kern, Kings and Madera Counties, in California, where it may be applied at 6 lbs. ai/A for control of red scale by ground application. - tobacco (liquids): 2 lbs. ai/A - tobacco (granulars): 3 lbs. ai/A - corn 1.0 lb/A **Maximum number of applications per season:** - alfalfa (liquids): 4 - citrus orchard floors (granulars): 3 - corn (field, sweet, seed) (granulars): 2 - sorghum (liquids): 3 - sugar beets (liquids): 3 - sunflowers (liquids): 3 - walnut and almond orchard floors (liquids): 2 - citrus (liquids): 2 - corn (field, sweet, seed) (liquids): 3 - cotton (liquids): 3 - soybeans (liquids): 3 - sugar beets (granulars): 1 - tobacco (liquids): 1 **Maximum amount a.i to be applied per acre per season:** - citrus (granulars) use on orchard floors: 3 lbs. ai/A - corn (field, sweet, seed) (liquids): 3 lbs. ai/A - corn (field, sweet, seed) (granulars): 2 lbs. ai/A - cotton (liquids): 3 lbs. ai/A - sugar beets (liquids): 3 lbs ai/A - walnut and almond orchard floors (liquids): 4 lbs. ai/A - sugar beets (granulars): 2 lbs ai/A - tobacco (liquids): 2 lbs ai/A - tree nuts (liquids): 4 lbs. ai/A - sunflowers (liquids): 3 lbs. ai/A	

Description	Amended Labeling Language	Placement on Label
Spray drift restrictions for outdoor products applied as sprays.	"Do not allow spray to drift from the application site and contact people, structures people occupy at any time and the associated property, parks and recreation areas, nontarget crops, aquatic and wetland areas, woodlands, pastures, rangelands, or animals." "For ground boom applications, do not apply within 25 feet of rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries and commercial fish ponds. Apply with nozzle height no more than 4 feet above the ground or crop canopy and when wind speed is 10 mph or less at the application site as measured by an anemometer. Use (registrant to fill in blank with spray quality, e.g. fine or medium) or coarser spray according to ASAE 572 definition for standard nozzles or VMD for spinning atomizer nozzles." "For orchard/vineyard airblast applications, do not apply within 50 feet of rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries and commercial fish ponds. Direct spray above trees/vines and turn off outward pointing nozzles at row ends and outer rows. Apply only when wind speed is 3 –10 mph at the application site as measured by an anemometer outside of the orchard/vineyard on the upwind side." "For aerial applications, do not apply within 150 feet of rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries and commercial fish ponds. The boom width must not exceed 75% of the wingspan or 90% of the rotary blade. Use upwind swath displacement and apply only when wind speed is 3 -- 10 mph as measured by an anemometer. Use _____ (registrant to fill in blank with spray quality, e.g. fine or medium) or coarser spray according to ASAE 572 definition for standard nozzles or VMD for spinning atomizer nozzles. If application includes a no-spray zone, do not release spray at a height greater than 10 feet above the ground or the crop canopy." "For overhead chemigation, do not apply within 25 feet of rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries and commercial fish ponds. Apply only when wind speed is 10 mph or less." "The applicator also must use all other measures necessary to control drift."	Directions for Use in General Precautions and Restrictions

Description	Amended Labeling Language	Placement on Label
End Use Products Intended for Occupational Use Products That Have Only Non-Worker Protection Standard (Non-WPS) Uses on the Label		
Handler PPE requirements (all formulations)	Note the following information when preparing labeling for all end use products: For **sole-active-ingredient** end-use products that contain chlorpyrifos, the product label must be revised to adopt the handler personal protective equipment (PPE)/engineering control requirements set forth in this section. Any conflicting PPE requirements on the current label must be removed. For **multiple-active-ingredient** end-use products that contain chlorpyrifos, the handler PPE/engineering control requirements set forth in this section must be compared with the requirements on the current label, and the more protective language must be retained. For guidance on which requirements are considered to be more protective, see PR Notice 93-7. PPE that is established on the basis of Acute Toxicity testing with the end-use products must be compared with the active ingredient PPE specified below in this document. The more protective PPE must be placed in the product labeling. For example, the Handler PPE in this RED does not require protective eyewear which may be required by the Acute Toxicity testing for the end-use product. For guidance on which PPE is considered more protective, see PR Notice 93-7.	
Handler PPE requirements for liquid formulations [1]	"Personal Protective Equipment (PPE) All mixers, loaders, applicators and handlers must wear: - coveralls over long-sleeved shirt and long pants; - chemical-resistant gloves such as (insert glove type as per Supplement Three of PR Notice 93-7); - chemical resistant apron when mixing or loading or exposed to the concentrate; - chemical-resistant footwear plus socks; - chemical-resistant headgear for overhead exposures; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter." *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	Immediately following/below Precautionary Statements: Hazards to Humans and Domestic Animals

Description	Amended Labeling Language	Placement on Label
Handler PPE requirements for wettable powder formulations. (wettable powder formulations must be in water-soluble packaging to be eligible for reregistration)	"Personal Protective Equipment (PPE) Mixers and loaders must wear: - long-sleeved shirt and long pants; - socks and shoes; - chemical resistant gloves such as (Registrant inserts glove type as per Supplement Three of PR Notice 93-7); - chemical resistant apron. Applicators using motorized ground boom application equipment must wear: - long-sleeved shirt and long pants; - socks and shoes. See engineering controls for additional requirements. All other handlers must wear: - coveralls over long-sleeved shirt and long pants; - chemical-resistant gloves; - chemical resistant apron when mixing or loading; - chemical-resistant footwear plus socks; - chemical-resistant headgear for overhead exposures; - a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter." *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	Immediately following/below Precautionary Statements: Hazards to Humans and Domestic Animals

Description	Amended Labeling Language	Placement on Label
Handler PPE requirements for granular products[1]	"Personal Protective Equipment (PPE) "Loaders, applicators and all other handlers must wear: –long-sleeved shirt and long pants; –socks and shoes. In addition to the above, loaders must wear: –chemical-resistant gloves such as (registrant inserts glove type as per Supplement Three of PR Notice 93-7.); –chemical-resistant apron; –a NIOSH-approved dust mist filtering respirator with MSHA/NIOSH approval number prefix TC-21C or a NIOSH-approved respirator any N, R, P, or HE filter. *Note: The registrant must drop the N-series filter from the respirator statement if the pesticide product contains or is used with oil.*	Immediately following/below Precautionary Statements: Hazards to Humans and Domestic Animals
User Safety Requirements	"Follow manufacturer's instructions for cleaning/maintaining PPE. If no such instructions for washables exist, use detergent and hot water. Keep and wash PPE separately from other laundry." "Discard clothing and other absorbent materials that have been drenched or heavily contaminated with this product's concentrate. Do not reuse them." (*This second statement is not required for granular formulations*)	Precautionary Statements: Hazards to Humans and Domestic Animals immediately following the PPE requirements
Engineering Controls requirements for liquid formulations	"Engineering Controls" "When handlers use closed cab motorized ground application equipment in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides (40 CFR 170.240(d)(4-6), the handler PPE requirements may be reduced or modified as specified in the WPS."	

Description	Amended Labeling Language	Placement on Label
Engineering Controls requirements for wettable powder formulations for products in water-soluble packaging	"Engineering Controls" "Water-soluble packets, when used correctly, qualify as a closed mixing/loading system. Mixers and loaders using water-soluble packets must wear the PPE required above for mixer/loaders, and have immediately available for use in emergency (such as a broken package, spill or equipment breakdown) additional PPE. These PPE include coveralls and chemical-resistant footwear and a non-powered air purifying respirator equipped with an N-, R- or P-series filter." "When handlers use closed cab motorized ground application equipment in a manner that meets the requirements listed in the Worker Protection Standard (WPS) for agricultural pesticides (40 CFR 170.240(d)(4-6), the handler PPE requirements may be reduced or modified as specified in the WPS."	
User Safety Recommendations	"User Safety Recommendations" "Users should wash hands before eating, drinking, chewing gum, using tobacco or using the toilet." "Users should remove clothing/PPE immediately if pesticide gets inside. Then wash thoroughly and put on clean clothing." "Users should remove PPE immediately after handling this product. Wash the outside of gloves before removing. As soon as possible, wash thoroughly and change into clean clothing."	Placed in a box in the Precautionary Statements under Hazards to Humans and Domestic Animals immediately following Engineering Controls.
Entry Restrictions for products applied as sprays	"Do not enter or allow others to enter until sprays have dried"	Directions for Use under Application Restrictions.
Entry Restrictions for granular products	"Do not enter or allow others to enter until dusts have settled"	Directions for Use under Application Restrictions.

Description	Amended Labeling Language	Placement on Label
Application Restrictions (all applicable formulations)	"Do not apply this product in a way that will contact workers or other persons, either directly or through drift. Only protected handlers may be in the area during application." The following statement should be placed on labels of products used on either golf course turf or manhole covers: "The maximum application rate per application is 1 lb. ai/A." "Do not use this product on manhole covers in storm drain systems."	Directions For Use under General Precautions and Restrictions
Spray drift restrictions for outdoor products applied as sprays.	"Do not allow spray to drift from the application site and contact people, structures people occupy at any time and the associated property, parks and recreation areas, nontarget crops, aquatic and wetland areas, woodlands, pastures, rangelands, or animals. For ground boom applications, do not apply within 25 feet of rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries and commercial fish ponds. Apply with nozzle height no more than 4 feet above the ground or crop canopy and when wind speed is 10 mph or less at the application site as measured by an anemometer. Use (registrant to fill in blank with spray quality, e.g. fine or medium) or coarser spray according to ASAE 572 definition for standard nozzles or VMD for spinning atomizer nozzles. The applicator also must use all other measures necessary to control drift."	Directions for Use under Application Restrictions.

[1] PPE that is established on the basis of Acute Toxicity of the end-use product must be compared to the active ingredient PPE in this document. The more protective PPE must be placed in the product labeling. For guidance on which PPE is considered more protective, see PR Notice 93-7.

[2] If the product contains oil or bears instructions that will allow application with an oil-containing material, the "N" designation must be dropped.

Instructions in the Labeling Changes section of Table 35 appearing in quotations represent the exact language that should appear on the label.

Instructions in the Labeling Changes section of Table 35 not in quotes represents actions that the registrant should take to amend their labels or product registrations.

VI. Related Documents and How to Access Them

This interim Reregistration Eligibility Document is supported by documents that are presently maintained in the OPP docket. The OPP docket is located in Room 119, Crystal Mall #2, 1921 Jefferson Davis Highway, Arlington, VA. It is open Monday through Friday, excluding legal holidays from 8:30 am to 4 pm..

The docket initially contained preliminary risk assessments and related documents as of October 17, 1999. Sixty days later the first public comment period closed. The EPA then considered comments, revised the risk assessment, and added the formal “Response to Comments” document and the revised risk assessment to the docket on August 16, 2000.

All documents, in hard copy form, may be viewed in the OPP docket room or downloaded or viewed via the Internet at the following site: "http://www.epa.gov/pesticides/op."

VII. Appendices

Appendix A. Table of Chlorpyrifos Use Patterns Eligible for Reregistration

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Crop Uses					
Alfalfa					
Soil in-furrow treatment At planting Ground equipment	15% G	1 lb/A	1	Not Applicable (NA)	Use limited to MO. A 21-day PHI/PGI has been established.
Broadcast application Foliar or postemergence Ground, sprinkler irrigation, or aerial equipment	4 lb/gal EC	1 lb/A	1 (per cutting) 4 (per season)	10	A 7-day PHI (rates ≤0.25 lb ai/A), a 14-day PHI (rates ≤0.5 lb ai/A), and a 21-day PHI (rates >0.5 lb ai/A) have been established.
Broadcast application Foliar Ground or aerial equipment	2 lb/gal EC	0.5 lb/A	1 (per cutting) 4 (per season)	10	Use limited to AZ and CA. A 4-day PHI/PGI (rates 0.375-0.5 lb ai/A) has been established.
Almonds					
Spray application Dormant/delayed dormant Ground equipment	50% WP	2 lb/A or 2 lb/100 gal	1	NA	
Spray application Dormant/delayed dormant Ground equipment	1 lb/gal EC 4 lb/gal EC	0.5 lb/100 gal [200-600 gal finished spray/A, 1 lb/A - 3 lb/A]	1	NA	Application may be made alone or as a tank mix with petroleum spray oil. Grazing of meat or dairy animals in treated orchards is prohibited.
Spray application Foliar Ground or aerial equipment	50% WP 50% DF 1 lb/gal EC 4 lb/gal EC	2 lb/A or 2 lb/100 gal	3	--	A 14-day PHI has been established. Grazing of livestock in treated orchards is prohibited (Section 3 and CA940017).

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Almonds (cont.)					
Trunk spray (bark) application Ground equipment	4 lb/gal EC	2 lb/A	1	--	Use limited to CA (CA940013). Grazing of livestock in treated orchards is prohibited.
Soil broadcast application Orchard floor Ground equipment	4 lb/gal EC	4 lb/A	2	--	A 14-day PHI has been established. Grazing of livestock in treated orchards is prohibited.
Soil broadcast application Orchard floor Ground equipment	4 lb/gal EC	3 lb/100 gal with 1.5 gal spray/tree	2	--	Use limited to CA (CA940024). Grazing of livestock in treated orchards is prohibited.
Apples					
Spray application Dormant/delayed dormant Ground equipment	1 lb/gal EC 4 lb/gal EC	0.5 lb/100 gal [200-600 gal finished spray/A]	1	NA	Application may be made alone or as a tank mix with petroleum spray oil. Grazing of meat or dairy animals in treated orchards is prohibited.
Spray application - branches and trunk Dormant/delayed dormant	4 lb/gal EC	2.0 lb/A	1		Use restricted to CA (Section 24(c) CA940013)

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Asparagus					
Broadcast foliar application Preharvest Ground equipment	4 lb/gal EC	1 lb/A	1	NA	Use limited to AZ, CA, the Midwest, and Pacific Northwest. A 1-day PHI has been established.
Broadcast application Postharvest (fern stage) Ground equipment	4 lb/gal EC	1 lb/A	2	10	Use limited to AZ, CA, the Midwest, and Pacific Northwest.
Bananas					
Fruit bag (shroud) application	1% Impr	--	--	--	Shrouds are installed on the stem after all fruit bunches have formed and are removed at harvest.
Bean (field, green, kidney, lima, navy, snap, string and wax)					
Slurry seed treatment Preplant	50% WP	1 oz/cwt	(1)	--	Grazing/feeding of livestock on bean hay grown from treated seed is prohibited. Treated seeds may not be used for food, feed, or oil purposes.
Slurry seed treatment Stored seed	50% WP	19.3 oz/23.5 gal [3 fl.oz/cwt]	(1)	--	Use limited to TX. Treated seeds may not be used for food, feed, or oil purposes.
Broccoli					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	Maximum seasonal application rates of 2.25 lb ai/A (0.5-15% G and 4 lb/gal EC) and 2.6 lb ai/A (1 lb/gal EC) are in effect. A 30-day PHI has been established for the EC formulations.

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Broccoli (continued)					
Soil band treatment At planting Ground equipment	4 lb/gal EC	1.4 oz/1,000 ft. of row	2	10	Use limited to CA (CA940016). Maximum seasonal application rate of 2.25 lb ai/A is in effect. Application may be repeated at thinning time as a directed spray. A 30-day PHI has been established.
Soil injected sidedress application	4 lb/gal EC	1.3 oz/1,000 ft. of row	1	NA	A 30-day PHI has been established.
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	A 21-day PHI has been established. Application may be made alone or as a tank mix with other pesticides (AZ870006, AZ940003, CA860066, CA940001).
Broccoli Raab (rapini)					
Soil application At planting Ground equipment	4 lb/gal EC	2.25 lb/A	1	NA	Section 24(c) CA940015.
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	40-day PHI. Section 24(c) AZ870006, AZ940003, CA860066, CA940001

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Brussels sprouts					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."
Soil band treatment At planting Ground equipment	4 lb/gal EC	1.4 oz/1,000 ft. of row	2	10	See "Broccoli."
Broadcast application Foliar Ground or aerial equipment	4 lb/gal EC	1 lb/A	3	10	A 21-day PHI has been established.
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Cabbage					
Soil band treatment At planting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."
Soil band treatment At planting Ground equipment	4 lb/gal EC	1.4 oz/1,000 ft. of row	2	--	See "Broccoli."
Soil injected sidedress application	4 lb/gal EC	1.3 oz/1,000 ft. of row	1	NA	See "Broccoli."
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."
Carrot (grown for seed)					
Broadcast application Foliar, After Bolting Ground or aerial equipment	4 lb/gal EC	1 lb/A	--	--	Use limited to WA (WA940002). Feeding of treated carrot cuttings or seed screenings to livestock or grazing of livestock in treated areas is prohibited.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Cauliflower					
Soil band treatment At planting/transplanting Ground equipment	0.5% G 1% G 15% G	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."
Soil band treatment At planting Ground equipment or Directed spray application Post-transplant Ground equipment	1 lb/gal EC 4 lb/gal EC	1.2 oz/1,000 ft. of row	1	NA	Maximum seasonal application rate of 2 lb ai/A is in effect. A 30-day PHI has been established.
Soil band treatment At planting Ground equipment	4 lb/gal EC	1.2 oz/1,000 ft. of row or 2 lb/A	2	10	Use limited to CA (CA960016). Maximum seasonal application rate of 2 lb ai/A is in effect. A 30-day PHI has been established.
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."
Cherries					
Trunk spray (bark) application Foliar and postharvest and/or dormant/delayed dormant Ground equipment	1 lb/gal EC 4 lb/gal EC	3 lb/100 gal	3	10	Use limited to sweet cherries. One of the three permitted applications per season may be applied as a dormant spray tank mixed with petroleum spray oil at 0.5 lb ai/100 gal. A 6-day PHI has been established. Grazing of meat or dairy animals in treated orchards is prohibited.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Cherries (continued)					
Spray application Foliar Ground or aerial equipment	50% WP 1 lb/gal EC	1.5 lb/A or 1.5 lb/100 gal	8	10	Use limited to sour (tart) cherries. A 14-day PHI has been established. Grazing of livestock in treated orchards is prohibited.
Chinese broccoli (gai lon)					
Soil application At planting Ground equipment	4 lb/gal EC	2.25 lb/A	1	NA	See "Broccoli raab."
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/gal	3	10	See "Broccoli."
Chinese cabbage (bok choy, napa)					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."
Soil application At planting Ground equipment	4 lb/gal EC	2.25 lb/A	1	NA	See "Broccoli."

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Chinese cabbage (bok choy, napa) (continued)					
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli." (AZ870006, AZ940003, CA860066, CA940001)
Chinese mustard (gai choy)					
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli, raab."
Citrus					
Spray application Foliar Ground or aerial equipment	4 lb/gal EC	6 lb/A (rates above 4 lb/A are limited to 5 counties in California)	1	30	Maximum seasonal application rate of 7.5 lb ai/A is in effect. A 21-day PHI (rates ≤ 3.5 lb ai/A) and a 35-day PHI (rates > 3.5 lb ai/A) have been established. Grazing of livestock in treated areas is prohibited. Application may be made alone or as a tank mix with other pesticides.
Spray application Foliar Ground or aerial equipment	4 lb/gal EC	3.5 lb/A	2	30	Maximum seasonal application rate of 7.5 lb ai/A is in effect. A 21-day PHI (rates ≤ 3.5 lb ai/A) and a 35-day PHI (rates > 3.5 lb ai/A) have been established. Grazing of livestock in treated areas is prohibited. Application may be made alone or as a tank mix with other pesticides.
Spray application Foliar Ground equipment	4 lb/gal EC	0.5 lb/100 gal	2	30	Use limited to residential citrus. A 21-day PHI has been established.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Citrus (continued)					
Spray application Foliar Ground or aerial equipment	1 lb/gal EC	0.4 lb/100 gal	2	30	Maximum seasonal application rate of 2 lb ai/A is in effect. A 21-day PHI has been established.
Trunk spray application Foliar Ground equipment	4 lb/gal EC	0.625 lb/A	4	--	Use limited to CA. A 28-day PHI has been established.
Fiberglass band application Foliar Ground equipment	4 lb/gal EC	2.5 lb/A	4	--	
Soil broadcast application Postplant (grove floor) Ground or sprinkler irrigation equipment	15% G 4 lb/gal EC	1 lb/A	3	10	Maximum seasonal application rate of 3 lb ai/A is in effect. A 28-day PHI has been established. Grazing of livestock in treated areas is prohibited. For use in FL, a maximum seasonal rate of 3 lb ai/A (EC) is in effect.
Clover (grown for seed)					
Soil broadcast application Preplant Ground equipment or Broadcast application Foliar Ground equipment	4 lb/gal EC	2 lb/A	1	--	Use limited to OR (OR940031). Grazing or feeding of treated clover cuttings or seed screenings or using of hay for livestock is prohibited.[b]

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Collards					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."
Collards (continued)					
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli, Raab."
Corn: field or sweet or pop or grown for seed					
Soil incorporated treatment Ground equipment	15% G	2 lb/A	(1)	NA	Maximum seasonal application rate of 2 lb ai/A is in effect. A 35-day PHI (corn grain), a 14-day PGI (corn silage), and a 35-day PFI (corn fodder) have been established.
Soil treatment At planting Ground equipment	0.5% G 1% G 7.5% G 15% G	2.4 oz/1,000 ft. of row or 2 lb/A	(1)	NA	
Soil treatment or broadcast application Ground or aerial equipment	15% G	1.2 oz/1,000 ft. of row	(1)	NA	

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Corn: field or sweet or pop or grown for seed (continued)					
Soil incorporated treatment Preplant Ground equipment	4 lb/gal EC	3 lb/A	(1)	NA	Maximum seasonal application rate of 3 lb ai/A is in effect. A 35-day PHI (corn grain), a 14-day PGI (corn silage), and a 35-day PFI (corn fodder) have been established. Application may be made alone or as a tank mix with other pesticides.
Soil broadcast application Preplant, at planting, or preemergence Ground equipment	4 lb/gal EC	1 lb/A	(1)	NA	
Broadcast application Postemergence/foliar Ground, aerial, or sprinkler irrigation equipment	4 lb/gal EC	1.5 lb/A	(5)	10	
Corn: Sweet					
Broadcast application Foliar Ground, aerial, or sprinkler irrigation equipment	4 lb/gal EC	1 lb/A	3	10	Use limited to FL and GA. Maximum seasonal application rate of 3 lb ai/A is in effect. A 21-day PHI (corn ears), PGI, and PFI (corn silage, fodder, or grain) have been established.
Broadcast application Foliar Ground or aerial equipment	4 lb/gal EC	0.5 lb/A	3	10	Use limited to DE (DE930004), A 7-day PHI has been established. Grazing of livestock in treated areas and feeding treated corn silage, forage, or fodder to meat or dairy animals is prohibited.[b]

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Corn: Field and Sweet					
Slurry seed treatment Preplant	50% WP	1 oz/cwt	(1)	--	Treated seeds may not be used for food, feed, or oil purposes.
Slurry seed treatment Stored seed	50% WP	19.3 oz/23.5 gal [3 fl.oz/cwt]	(1)	--	See "Bean."
Cotton					
Broadcast application Foliar Ground, sprinkler irrigation, aerial equipment	4 lb/gal EC	1 lb/A	3	10	A 14-day PHI has been established. Grazing of livestock in treated areas and feeding of gin trash or treated forage to livestock is prohibited.[b]
Broadcast application Foliar Ground or aerial equipment	2 lb/gal EC	0.5 lb/A	3	10	Use limited to AZ and CA. A 40-day PHI has been established. Grazing of livestock in treated areas and feeding of gin trash or treated forage to livestock is prohibited.[b] Applications may be made undiluted at the same rate.
Slurry seed treatment Stored seed	50% WP	19.3 oz/23.5 gal [3 fl.oz/cwt]	(1)	--	See "Bean."
Gin trash treatment Ground equipment	4 lb/gal EC	1 lb per 20 tons of gin trash	--	--	Use limited to MS.
Cranberry					
Broadcast application Foliar Ground, aerial, or sprinkler irrigation equipment	4 lb/gal EC	1.5 lb/A	2	10	A 60-day PHI has been established. Application may not be made when bogs are flooded.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Cucumbers					
Slurry seed treatment Preplant	50% WP	1 oz/cwt	(1)	--	Treated seeds may not be used for food, feed, or oil purposes.
Figs					
Soil incorporated treatment Dormant Ground equipment	4 lb/gal EC	2 lb/A	1	NA	Use limited to CA. A 210-day PHI has been established.
Filberts					
Spray application Foliar Ground or aerial equipment	50% WP 1 lb/gal EC 4 lb/gal EC	2 lb/A or 2 lb/100 gal	3	--	A 14-day PHI has been established. Grazing of livestock in treated orchards is prohibited.
Grapefruit					
Spray application Foliar Ground or aerial equipment	4 lb/gal EC	6 lb/A	2	30	See "Citrus."
Spray application Foliar or transplant Ground or aerial equipment	4 lb/gal EC	3.5 lb/A	2	30	See "Citrus."
Spray application Foliar Ground equipment	4 lb/gal EC	0.5 lb/100 gal	2	30	See "Citrus."

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Grapefruit (continued)					
Spray application Foliar Ground or aerial equipment	1 lb/gal EC	0.4 lb/100 gal	2	30	See "Citrus."
Grapes					
Directed spray soil application Ground equipment	1 lb/gal EC 4 lb/gal EC	2.25 lb/100 gal [2 qt finished spray/15 sq. ft.]	1	NA	Use limited to states east of the Rocky Mountains. A 35-day PHI has been established.
Directed spray soil application Ground equipment	4 lb/gal EC	1.125 lb/100 gal [2 qt finished spray per 15 sq. ft.]	2	--	Use limited to TN (TN940001). A 35-day PHI has been established.
Directed spray soil application Ground equipment	4 lb/gal EC	1 lb/A	3	--	Use limited to CA (CA940018). A 76-day PHI has been established.
Spray/drench application Prebloom Ground equipment	4 lb/gal EC	1 lb/A	1	NA	Use limited to MI and MO (MI940001 and MO940001).
Broadcast foliar application **Nonbearing** Ground or aerial equipment	4 lb/gal EC	1 lb/A	--	--	Use limited to ID, OR, and WA (ID940013, OR940030, and WA940003).

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Grass (grown for seed)					
Broadcast application Foliar Ground or aerial equipment	4 lb/gal EC	1 lb/A	3	--	Use limited to OR and NV (OR940032 and NV940002). Grazing of livestock in treated areas or feeding treated grass, straw, or seed screenings to livestock or using hay for livestock bedding is prohibited.[b]
Kale					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."
Kohlrabi					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Kohlrabi (continued)					
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."
Lemon					
Spray application Foliar Ground or aerial equipment	4 lb/gal EC	6 lb/A	2	30	See "Citrus."
Spray application Foliar or transplant Ground or aerial equipment	4 lb/gal EC	3.5 lb/A	2	30	See "Citrus."
Spray application Foliar Ground equipment	4 lb/gal EC	0.5 lb/100 gal	2	30	See "Citrus."
Spray application Foliar Ground or aerial equipment	1 lb/gal EC	0.4 lb/100 gal	2	30	See "Citrus."
Macadamia Nuts					
Trunk spray (bark) application Ground equipment	50% WP	1 lb/A	8	30	Use limited to HI (HI930010 and HI930011). Maximum seasonal application rate of 8 lb ai/A is in effect. A 14-day PHI has been established. Grazing of livestock in treated areas is prohibited.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Mint - Peppermint					
Soil incorporated treatment Preplant Ground equipment	4 lb/gal EC	2 lb/A	1	NA	Use limited to OR (OR940027). Application following a broadcast foliar spray is not permitted.
Broadcast foliar application Preharvest and postharvest Ground or sprinkler irrigation equipment	1 lb/gal EC 4 lb/gal EC	2 lb/A	1 preharvst + 1 postharvest	NA	A 90-day PHI has been established.
Mustard greens					
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."
Nectarines					
Spray application Dormant/delayed dormant Branches and Trunk Ground equipment	1 lb/gal EC 4 lb/gal EC	0.5 lb/100 gal [200-600 gal finished spray/A, 1 lb/A-3 lb/A]	1	NA	Application may be made alone or as a tank mix with petroleum spray oil. Grazing of meat or dairy animals in treated orchards is prohibited.
Spray application Dormant/delayed dormant Branches and Trunk Ground equipment	4 lb/gal EC	2/ lb/A	1	NA	Use limited to CA (CA940013)

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Nectarines (continued)					
Trunk spray (bark) application Ground equipment	1 lb/gal EC 4 lb/gal EC	3 lb/100 gal	1	NA	A 14-day PHI has been established. Grazing of meat or dairy animals in treated orchards is prohibited.
Onions, bulb					
Soil application At seeding Ground equipment	0.5% G 1% G 15% G	0.035 lb/1,000 ft. of row	1	NA	Maximum seasonal application rate of 1 lb ai/A is in effect for the 15% G formulation.
Soil drench application At seeding Ground equipment	1 lb/gal EC 4 lb/gal EC	0.04 lb/1,000 ft. of row (1 lb/gal EC) 0.03 lb/1,000 ft. of row (4 lb/gal EC)	1	NA	
Soil drench application Post planting Ground equipment	4 lb/gal EC	1 lb/A	2		Use limited to MI (MI940002. 60 day PHI. Total number of applications should include both at planting and post crop uses.
Oranges					
Spray application Foliar Ground or aerial equipment	4 lb/gal EC	6 lb/A	2	30	See "Citrus."
Spray application Foliar or transplant Ground or aerial equipment	4 lb/gal EC	3.5 lb/A	2	30	See "Citrus."

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Oranges (continued)					
Spray application Foliar Ground equipment	4 lb/gal EC	0.5 lb/100 gal	2	30	See "Citrus."
Spray application Foliar Ground or aerial equipment	1 lb/gal EC	0.4 lb/100 gal	2	30	See "Citrus."
Peaches					
Spray application Dormant/delayed dormant Ground equipment	1 lb/gal EC 4 lb/gal EC	0.5 lb/100 gal [200-600 gal finished spray/A]	1	NA	See "Nectarines."
Trunk spray (bark) application Ground equipment	4 lb/gal EC	3 lb/100 gal	1	NA	See "Nectarines."
Dip application Preplant (nonbearing)	4 lb/gal EC	3 lb/100 gal	1	NA	

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Peanuts					
Soil incorporated treatment Preplant Ground equipment	4 lb/gal EC	2 lb/A	1	NA	A combined maximum seasonal application rate of 4 lb ai/A is in effect for preplant and postplant use. A 21-day PHI has been established. Feeding peanut forage or hay to meat or dairy animals is prohibited.
Soil band application At planting, postplant, or early pegging Ground equipment	0.5% G 1% G 15% G	2.25 oz ai/1,000 ft. of row (2 lb/A)	2	NA	A maximum seasonal application rate of 4.5 oz ai/1,000 ft. of row or 4 lb ai/A for the 15% G formulation is in effect. A maximum seasonal rate of 2.25 oz ai/1,000 ft. of row is in effect. A 21-day PHI has been established. Feeding peanut forage or hay to meat or dairy animals is prohibited.
Broadcast application Prior to or at pegging	15% G	1.95 lb/A	--	10	A maximum seasonal application rate of 4 lb ai/A is in effect. A 21-day PHI has been established. Feeding peanut forage or hay to meat or dairy animals is prohibited.
Directed spray application Foliar Ground equipment	1 lb/gal EC	2 lb/A	1	NA	A 21-day PHI has been established. A maximum seasonal application rate of 2 lb ai/A is in effect.
Pears					
Spray application Dormant/delayed dormant Ground equipment	1 lb/gal EC 4 lb/gal EC	0.5 lb/100 gal [200-600 gal finished spray/A]	1	NA	See "Apples."
Spray application Dormant/delayed dormant Branches and Trunk	4 lb/gal EC	2 lb/A	1	NA	Use limited to CA (CA940013).

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Peas (black-eyed, field, and garden)					
Slurry seed treatment Preplant	50% WP	1 oz/cwt	(1)	--	See "Bean."
Slurry seed treatment Stored seed	50% WP	19.3 oz/23.5 gal [3 fl.oz/cwt]	(1)	--	See "Bean."
Pecans					
Spray application Foliar Ground or aerial equipment	50% WP 50% DF 1 lb/gal EC 4 lb/gal EC	1 lb/100 gal or 1 lb/A (50% WP, 50% DF, and 1 lb/gal EC) 2 lb/A (4 lb/gal EC)	5	--	A maximum seasonal application rate of 10 lb ai/A is in effect for the 4 lb/gal EC formulation. Application may be made alone or as a tank mix with other pesticides. A 28-day PHI has been established. The grazing of livestock in treated orchards is prohibited.
Soil broadcast application Orchard floor Ground equipment	50% WP 1 lb/gal EC 4 lb/gal EC	1 lb/100 gal or 1 lb/A (50% WP and 1 lb/gal EC) 2 lb/A (4 lb/gal EC)	5	--	
Peppers					
Broadcast application Foliar Ground equipment	50% WP	1 lb/A	8	-	Use limited to FL and GA (FL920007, FL920009, GA930003, and GA930004). A 7-day PHI has been established.
Broadcast application Foliar Ground equipment	50% WP	1 lb/A	8		Use limited to NM and TX (NM95001). A 14 day PHI has been established.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Plum/Prune					
Spray application Dormant/delayed dormant Ground equipment	1 lb/gal EC 4 lb/gal EC	0.5 lb/100 gal [200-600 gal finished spray/A, 1 lb/A -3 lb/A]	1	NA	See "Apples."
Spray application Dormant/delayed dormant Ground or aerial equipment	4 lb/gal EC	2 lb/A	1	NA	Use limited to CA (CA940013)
Pumpkin					
Slurry seed treatment Preplant	50% WP	1 oz/cwt	(1)	--	See "Bean."
Radish					
Soil in-furrow treatment At planting Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	0.5 oz/1,000 ft. of row (2.75 lb/A)	1	NA	A maximum seasonal application rate of 2.75 lb ai/A is in effect for the 0.5-15% G, 1 lb/gal EC and 4 lb/gal EC formulations.
Radish (grown for seed)					
Soil incorporated treatment Preplant Ground equipment	4 lb/gal EC	2 lb/A	(1)	NA	Use limited to OR (OR94033). Grazing of livestock in treated areas or the feeding of radish cuttings or seed screenings to livestock is prohibited.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Rape					
Broadcast application Foliar Ground or aerial equipment	50% WP	1 lb/A	3	10	See "Broccoli."
Rutabagas					
Soil band treatment At planting/transplanting Ground equipment	0.5% G 1% G 15% G	1.4 oz/1,000 ft. of row (2.25 lb/A)	1	NA	Maximum seasonal application rate of 2.25 lb ai/A is in effect. The use of rutabaga tops for food/feed purposes is prohibited.
	4 lb/gal EC	1.6 oz/1,000 ft. of row (2.25 lb/A)	1	NA	
Soil band treatment At planting Ground equipment	1 lb/gal EC	1.3 oz/1,000 ft. of row	1	NA	Maximum seasonal application rate of 1.9 lb ai/A is in effect. The use of rutabaga tops for food/feed purposes is prohibited.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Sorghum					
Soil T-band incorporated treatment At planting Ground equipment	15% G	8 oz/1,000 ft. of row (1.5 lb/A)	1	NA	
Broadcast application Foliar Ground, sprinkler irrigation, or aerial equipment or Directed spray application Foliar Ground equipment	4 lb/gal EC	1 lb/A	3	10	Maximum seasonal application rate of 1.5 lb ai/A is in effect. A 30-day PHI/PGI/PFI for rates 0.5 lb ai/A and a 60-day PHI/PGI/PFI for rates >0.5 lb ai/A have been established. Use on sweet sorghum is prohibited.
Slurry seed treatment Stored seed	50% WP	19.3 oz/23.5 gal [3 fl.oz/cwt]	(1)	NA	See "Bean."

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Soybean					
Soil T-band incorporated treatment At planting or postemergence Ground equipment	15% G	1.2 oz/1,000 ft. of row	1	NA	
Soil band application At planting Ground equipment or Directed soil band application, Postemergence Ground equipment or Broadcast spray application Foliar Ground, sprinkler irrigation, or aerial equipment	4 lb/gal EC	1 lb/A	3	14 (between final two applications)	Maximum seasonal application rate of 3 lb ai/A is in effect. A 28-day PHI has been established. Grazing of livestock in treated areas or the feeding of treated soybean forage, hay, and straw to meat or dairy animals is prohibited.
Strawberry					
Soil incorporated treatment Preplant Ground equipment	4 lb/gal EC	2 lb/A	1	NA	Use limited to ID, OR, and WA (ID940012, OR940035, and WA94004) Application made one year before harvest season.
Broadcast foliar application Prebloom Ground equipment	1 lb/gal EC 4 lb/gal EC	1 lb/A	2	10	A 21-day PHI has been established.

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Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Strawberry (continued)					
Broadcast foliar application Prebloom Ground equipment	1 lb/gal EC 4 lb/gal EC	1 lb/A	1 pre-plant 2 foliar	10 (foliar)	A 21-day PHI has been established.
Directed spray application Postharvest Ground equipment	4 lb/gal EC	1 lb/A	2	14	Use limited to OR (OR940034).
Sugar beet					
Soil T-band application At planting or postemergence (two- to four-leaf stage) Ground equipment	15% G	1.35 oz/1,000 ft. of row or 2 lb/A (based on a 22-inch row spacing)	1	NA	
Soil incorporated treatment Preplant Ground equipment or Soil band application At planting Ground equipment	4 lb/gal EC	4.6 oz/100 ft row (30 in row) or 1 lb/A	(1)	NA	Maximum seasonal application rate of 4 lb ai/A is in effect. A 30-day PHI/PGI have been established. Application may be made alone or as a tank mix with other pesticides.

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).					
Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Sugar beet (continued)					
Broadcast application Foliar Ground or aerial equipment or Soil band application Foliar Ground equipment	4 lb/gal EC	1 lb/A	3	10	Maximum seasonal application rate of 4 lb ai/A is in effect. A 30-day PHI/PGI have been established.
Sugar beet (grown for seed)					
Soil broadcast application Preplant Ground equipment	4 lb/gal EC	2 lb/A	1 - fall before harvest season	NA	Use limited to ID and OR (ID950018 and OR940028).
Sunflower					
Soil band application At planting Ground equipment	0.5% G 1% G 15% G	1.25 oz/1,000 ft. of row	1	NA	
Soil incorporated treatment Preplant Ground equipment	4 lb/gal EC	2 lb/A	1	NA	Maximum seasonal application rate of 3 lb ai/A is in effect. A 42-day PHI has been established. Grazing of livestock in treated areas is prohibited.
Broadcast foliar application Postemergence Ground or aerial equipment	4 lb/gal EC	1.5 lb/A	3	7	

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Sweet Potato					
Soil incorporated treatment Preplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	2 lb/A	1	NA	A 125-day PHI has been established.
Tobacco					
Soil incorporated treatment Pre-transplant Ground equipment	15% G 4 lb/gal EC	3 lb/A	1	NA	
Soil incorporated treatment Pre-transplant Ground equipment	4 lb/gal EC	2 lb/A	1	NA	Tank mix use in all tobacco growing regions.
Turnip					
Soil band treatment At planting/transplanting Ground equipment or Directed spray application Post-transplant Ground equipment	0.5% G 1% G 15% G 1 lb/gal EC 4 lb/gal EC	1.4 oz/1,000 ft. of row	1	NA	See "Broccoli."

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Walnuts					
Spray application Dormant/delayed dormant Ground or aerial equipment	50% WP	2 lb/A or 2 lb/100 gal	1	NA	
Spray application Foliar Ground or aerial equipment	50% WP 50% DF 1 lb/gal EC 4 lb/gal EC	2 lb/A or 2 lb/100 gal	2	--	A 14-day PHI has been established. Grazing of livestock in treated orchards is prohibited.
Soil spray application Ground equipment	4 lb/gal EC	4 lb/A	2		A 14-day PHI has been established. Grazing of livestock in treated orchards is prohibited. Ant control for orchard floors.
Wheat					
Broadcast application Foliar Ground, sprinkler irrigation, or aerial equipment	4 lb/gal EC	0.5 lb/A	2	--	A 14-day PHI for forage and hay, and a 28-day PHI for grain and straw have been established.

Appendix A. Food/Feed Use Patterns Subject to Reregistration for Chlorpyrifos (Case 0100).

Site Application Type Application Timing Application Equipment	Form	Max. Single Application Rate (ai)	Max. # Apps.	Min. Retreatment Interval (Days)	Use Limitations
Animal uses					
Cattle (beef, calves, and lactating and non-lactating dairy)					
Ear tag treatment	5% Impr	Two ear tags/animal	--	--	One tag is attached to each ear when pests first appear in the spring. Tags may be replaced as needed.
Outdoor turkey pens					
Soil treatment Before turkeys are transferred to pens Ground equipment	50% WP 50% DF	4 lb/A	2	28	Direct application to turkeys is prohibited. A 7-day PSI has been established. Open feed should be covered during spraying and water troughs should be flushed out immediately after spraying operations.
Food-handling establishment uses					
Food-Handling Establishments					
Spot and/or crack and crevice treatment Coarse low pressure sprayer or paint brush	1 lb/gal Mcap 1.7 lb/gal Mcap	0.5% spray	--	14	
Spot and/or crack and crevice treatment Coarse low pressure sprayer or paint brush	2 lb/gal EC 4 lb/gal EC 0.5% RTU	0.5% spray	--	7	Applications may be repeated at 7-day intervals in food service establishments and every 14 days in other types of food handling establishments. Emergency application may be made 2 days after the last treatment; limited to one emergency treatment per month.

[a] Unless protective clothing is worn.
[b] According to Table 1 (OPPTS, 860.1000) label restrictions on these commodities are not practical and will no longer be accepted.

Appendix B. Table of Generic Data Requirements and Studies Used to Make the Reregistration Decision

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Data Supporting Guideline Requirements for the Reregistration of Chlorpyrifos

REQUIREMENT			USE PATT	CITATION(S)
PRODUCT CHEMISTRY				
New Guideline Number	Old Guid. Number	Guideline Name		MRID
830.1550	61-1	Product Identity and Composition	All	00146506, 00146508, 45434001, data gap for MPs
830.1600	61-2A	Start. Mat. & Mnfg. Process	All	00146506, 00146508, 40105301, 40411301, 45434001, data gap for MPs
830.1670	61-2B	Formation of Impurities	All	00146506, 00146508, 40105301, 42495401, 45434001, data gap for MPs
830.1700	62-1	Preliminary Analysis	All	00146506, 00146508, 40144101, 42544901, 45434001, data gap for MPs
830.1750	62-2	Certification of limits	All	00146506, 00146508, 40105301, 45434001, data gap for MPs
830.1800	62-3	Analytical Method	All	00146506, 00146508, 40144101, 45434001, 42527203, data gap for MPs
830.6302	63-2	Color	All	00146506, 00146508, data gap for MPs
830.6303	63-3	Physical State	All	00146506, 00146508, data gap for MPs
830.6304	63-4	Odor	All	00146506, 00146508, data gap for MPs
830.7200	63-5	Melting Point	All	00146506, 00146508, data gap for MPs
830.7300	63-7	Density	All	00146506, 00146508, 42495402, 41747202, data gap for MPs
830.7840 830.7860	63-8	Solubility	All	00146506, 00146508, data gap for MPs
830.7950	63-9	Vapor Pressure	All	00146506, 00146508, data gap for MPs
830.7370	63-10	Dissociation Constant	All	N/A

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REQUIREMENT			USE PATT	CITATION(S)
830.7550	63-11	Octanol/Water Partition Coefficient	All	00146506, 00146508, 42652601, data gap for MPs
830.7000	63-12	pH	All	N/A
830.6313	63-13	Stability	All	00146506, 00146508, data gap for MPs
830.6314	63-14	Oxidizing/Reducing Action	All	41742705, 43428701
830.6315	63-15	Flammability	All	N/A
830.6316	63-16	Explodability	All	00146506, 43046602, 43428702, data gap for MPs
830.6317	63-17	Storage Stability	All	00146506, 00146508, 41747204, 43633901, data gap for MPs
830.7100	63-18	Viscosity	All	N/A
830.6319	63-19	Miscibility	All	N/A
830.6320	63-20	Corrosion characteristics	All	00146506, 00146508, 41653503, 42527201, data gap for MPs
830.7050	None	UV/Visible Absorption	A,B	data gap for MPs
	ECOLOGICAL EFFECTS			
850.2100	71-1	Avian Acute Oral Toxicity	A,B	00046954, 40854701, 41043901, 41885201, 44057101, 44057102, 44585403
850.2200	71-2A	Avian Dietary Toxicity - Quail	A,B	00046955, 00095123, 00095304, 00095305, 40854703, 41965502, 44055101, 44062601, 44585401
850.2200	71-2B	Avian Dietary Toxicity - Duck	A,B	00046958, 00095007, 00095446, 00095449, 40854702, 41965501
850.2400	71-3	Earthworm Toxicity	A,B	00078524, 00095371,
850.2300	71-4A	Avian Reproduction - Quail	A,B	00046951, 42144902
850.2300	71-4B	Avian Reproduction - Duck	A,B	00046952, 00046953, 42144901

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REQUIREMENT			USE PATT	CITATION(S)
850.1075	72-1A	Fish Toxicity - Bluegill	A,B	00095013, 00095125, 00095298, 00095296, 00095321, 00154732, 40840904, 41043903, 41885203
850.1075	72-1C	Fish Toxicity, Rainbow Trout	A,B	00095013, 00095297, 00155781, 40840903, 41885204
850.1010	72-2A	Invertebrate Toxicity	A,B	00024400, 00095338, 00095365, 00095366, 00095368, 00095370, 00102520, 00154727, 05000774, 05000821, 05000841, 40840902, 41073401
850.1010	72-2B	Invertebrate Toxicity TEP	A,B	41885202
None	72-3A	Estuarine/Marine Toxicity - Fish	A,B	00102758, 00154718, 42144904
None	72-3B	Estuarine/Marine Toxicity - Mollusk	A,B	42144905, 42495405, 42495406
None	72-3C	Estuarine/Marine Toxicity - Shrimp	A,B	00095363, 42144906, 42245902
None	72-4A	Fish- Early Life Stage	A,B	00154732, 41043903
None		Estuarine Field Studies	A,B	00095130, 00095301, 00095367, 00104696, 00158261, 05000928, 41205409, 41228801, 44585408
850.1500	72-5	Life Cycle Fish	A,B	42834401, 00154721
		Terrestrial Field Toxicity Study	A,B	00095114, 42144903, 43483101, 43483102, 43730301, 43706701, 43785201, 43785202, 44692001, 44709401
850.4400	123-2	Aquatic Plant Growth	A,B	00024400, 41063402
850.3020	141-1	Honey Bee Acute Contact	A,B	00040602, 00060632, 41654701
		Water Monitoring	A,B	43065601, 43760601, 43760602, 43760603, 43760604, 43760605, 43760608, 43760609, 43760610, 43760611, 43786901, 43823901, 43853201, 43853202, 43918301, 44033401, 44033402, 44223601, 44235001, 44711601, 45013101, 43319201. Data gap for collection of water monitoring data to confirm reduction of residues in surface water.

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REQUIREMENT			USE PATT	CITATION(S)
		Amphibian Toxicity	**A,B**	**44692201, 45506303**
		Simulated Freshwater Field Studies	**A,B**	**00024400, 00095366, 00154717, 44823801**
		Freshwater Microcosm/Fish Toxicity	**A,B**	**00092775, 00095128, 00095370, 41205403, 43216401, 43216402, 43216403, 44692101, 44585405**
TOXICOLOGY				
870.1100	**81-1**	**Acute Oral Toxicity-Rat**	**A,B**	**44209101, 42495404, 44884301**
870.1200	**81-2**	**Acute Dermal Toxicity-Rabbit/Rat**	**A,B**	**44209102**
870.1300	**81-3**	**Acute Inhalation Toxicity-Rat**	**A,B**	**00146507, 40055001**
870.2400	**81-4**	**Primary Eye Irritation-Rabbit**	**A,B**	**44209103**
870.2500	**81-5**	**Primary Skin Irritation**	**A,B**	**44209104**
870.2600	**81-6**	**Dermal Sensitization**	**A,B**	**44209105**
870.6100	**81-7**	**Acute Delayed Neurotoxicity - Hen**	**A,B**	**00097144, 00405106**
		Special Acute Rat Neurotoxic Esterase	**A,B**	**44273901**
		Acute Pharmacokinetic Study - rat	**A,B**	**44648102**
		Cognitive Rat Study	**A,B**	**44020901**
870.3100	**82-1A**	**90-Day Feeding - Rodent**	**A,B**	**40436406, 40952801**
870.3150	**82-1B**	**90-Day Feeding - Non-rodent**	**A,B**	**42172801**
870.3200	**82-2**	**21-Day Dermal - Rabbit/Rat**	**A,B**	**40972801, 41340201**
870.3465	**82-4**	**90-Day Inhalation-Rat**	**A,B**	**40013901, 40166501, 40908401**
	82-8	**13-Week Rat Neurotoxicity study**	**A,B**	**42929801, 43426601**
870.4100	**83-1A**	**Chronic Feeding Toxicity - Rodent**	**A,B**	**40952802, 42172802, 42534201**

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REQUIREMENT			USE PATT	CITATION(S)
870.4100	83-1B	Chronic Feeding Toxicity - Non-Rodent	A,B	00064933, 00146519, 45360101
870.4200	83-2A	Oncogenicity - Rat	A,B	40952802, 42172802
870.4200	83-2B	Oncogenicity - Mouse	A,B	00054352, 00142902, 42534201
870.3700	83-3A	Developmental Tox. - Rat	A,B	00095268, 00130400, 40436407
870.3700	83-3B	Developmental Toxicity - Rabbit	A,B	40436408
870.3800	83-4	2-Generation Repro. - Rat	A,B	00029064, 00064934, 41930301
870.6200	83-6	Developmental Neurotoxicity - rat	A,B	44556901, 44648101, 45360102
870.5140 870.5375	84-2A 84-2B	Mutagenicity Studies	A,B	00152683, 00152684, 00157058, 00157057, 40057201 40436401, 40436409, 40436411, 41340203, 44533401
870.7485	85-1	General Metabolism	A,B	40458901, 44648102, 44810701
		6-Week Dietary Study Acetylcholinesterase Inhibition in the Dog	A,B	45467301, 45467302
		Human data	A,B	42008401, 42031701, 44035001, 44811002, 44889501, 45098001, 45144101, 45195701, 45195702, 45195703, 45195704, 45195705
OCCUPATIONAL/RESIDENTIAL EXPOSURE				
875.2100	132-1A	Foliar Residue Dissipation	A,B	42974501, 42994401, 43062701, 43062702, 44748101, 44748102, data gap for ornamentals grown in greenhouses, biological monitoring data to develop transfer coefficientf for various greenhouse/nursery activities
875.2200	132-1B	Soil Residue Dissipation	A,B	41540202, 42974501, data gap for reentry into treated areas with soil incorporated/directed applications

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REQUIREMENT			USE PATT	CITATION(S)
875.2400	133-3	Dermal Passive Dosimetry Exposure	A,B	42974501, 42994401, 42994401, 43027901, 43042002, 43138101, 43138102, 44483501, 44739302,
875.2500	133-4	Inhalation Passive Dosimetry Exposure	A,B	42974501, 42994401, 42994401, 43027901, 43042002, 43138101, 43138102, 44483501, 44739302,
875.1100	231	Estimation of Dermal Exposure at Outdoor Sites	A,B	40026001, 43013501, 43013502, 43013503, 43042001 44167101, 44444801, 44729401, 44729402, 44739301, 44589001, data gap for seed treatment uses, dip applications (preplant peach), mixing wettable powders for aerial/chemigation application, loading and applying granulars for aerial applications, groundboom application, and backpack spray applications
875.1300	232	Estimation of Inhalation Exposure at Outdoor Sites	A,B	40026001, 43013501, 44167101, 44444801, 44729401, 44729402, 44739301, 44589001, data gap for seed treatment uses, dip applications (preplant peach), mixing wettable powders for aerial/chemigation application, loading and applying granulars for aerial applications , groundboom application, and backpack spray applications
	233	Estimation of Dermal Exposure at Indoor Sites	A,B	40094001, 44458201, 42887201, Data gap for treated wood in residential structures.
	234	Estimation of Inhalation Exposure at Indoor Sites	A,B	40094001, 44458201, 43963701, Data gap for treated wood in residential structures.
ENVIRONMENTAL FATE				
None	160-5	Chemical Identity	A,B	00146506, 00146508
835.2120	161-1	Hydrolysis	A,B	00155577
835.2240	161-2	Photodegradation - Water	A,B	41747206, 40026101
835.2410	161-3	Photodegradation - Soil	A,B	42495403
835.2370	161-4	Photodegradation - Air	A,B	40234801, waived

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REQUIREMENT			USE PATT	CITATION(S)
835.4100	162-1	Aerobic Soil Metabolism	A,B	00025619, 42144911, 42144912
835.4200	162-2	Anaerobic Soil Metabolism	A,B	00025619
835.4400	162-3	Anaerobic Aquatic Metabolism	A,B	waived
835.4300	162-4	Aerobic Aquatic Metabolism	A,B	44083401, waived
835.1240	163-1	Leaching/Adsorption/ Desorption	A,B	00155636, 00155637, 40050401, 41892801, 41892802, 42493901
835.6100	164-1	Terrestrial Field Dissipation	A,B	40059001, 40395201, 42874702, 42874703, 42874704, 42924801, 42924802,
835.1850	165-1	Confined Rotational Crop	A,B	43210801
None	165-4	Bioaccumulation in Fish	A,B	40056401, 42495405, 42495406
RESIDUE CHEMISTRY				
None	171-2	Chemical Identity	A,B	00146506, 00146508
860.1300	171-4A	Nature of Residue - Plants	A,B	00066724, 00066725, 00072657, 00072660, 00157541, 00157542, 00157543, 40638801, 40638802, 41829007
860.1300	171-4B	Nature of Residue- Livestock	A,B	00077055, 00154734,00161743, 40638802
860.1340	171-4C+D	Residue Analytical Method - Plants and Animals	A,B	00034031, 00037455, 00037457, 00037458, 00039642, 00039643, 00051801, 00058089, 00084330, 00084331, 00095179, 00095201, 00095216, 00095251, 00095383, 00095387, 00095436, 00134720, 00141725, 00148881, 00155578, 00155579, 00155580, 00157713, 00158566, 00158567, 00158568, 00158569, 00162109, 00164187, 40131301, 40131302, 40288501
860.1380	171-4E	Storage Stability	A,B	00033586, 00034031, 00044555, 00051798, 00077120, 00095227, 00095260, 00095374, 00101566, 00116675, 00134720, 00162109

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REQUIREMENT			USE PATT	CITATION(S)
860.1480	171-4J	Magnitude of Residues - Meat/Milk/Poultry/Egg	A,B	00058087, 00095179, 00095438, 42542701
860.1500	171-4K	Magnitude of Residue in Plants (Root and Tuber Vegetables Group)	A,B	Radish, fresh - 0095259 Rutabagas, root - 0095259 Sugar beets, root - 00039641, 00101566 Sweet potatoes, root - 00095227 Turnip, root - 0095259
860.1500	171-4K	Mag. of Res.- Plants (Leaves of Root and Tuber Veg. Group)	A,B	Sugar beets, tops - 00039641, 00101566 Turnip, tops - 00095259
860.1500	171-4K	Mag. of Res.- Plants (Bulb Veg. Group)	A,B	Leeks - 00157909 Onions, dry bulb(only) - 00154019, 42649001
860.1500	171-4K	Mag. of Res.- Plants (Brassica Leafy Vegetables group)	A,B	Broccoli - 00095273, 00155580, 00158566 Brussels sprouts - 00095273, 00158566 Cabbage - 00095273, 00155580, 00158566 Cabbage, Chinese - 00095273 Cauliflower - 00095273, 00158566
860.1500	171-4K	Mag. of Res.- Plants Legume Vegetables (succulent or dried) Group	A,B	Beans, lima - 42245907 Beans, snap - 42245907 Soybeans - 00095270, data gap for aspirated grain fractions
860.1500	171-4K	Mag. of Res.- Plants (Foliage of Legume Vegetables Group)	A,B	Beans, vines - 00095264, 42245907 Beans, lima, vines - 00095264, 42245907 Beans, snap, vines - 42245907 Peas, vines - 00095264 Soybeans, forage - 00095270
860.1500	171-4K	Mag. of Res.- Plants [Fruiting Vegetables (except cucurbits) Group]	A,B	Tomatoes -00095251, 00131864, (tomato tolerance being revoked)

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REQUIREMENT			USE PATT	CITATION(S)
860.1500	171-4K	Mag. of Res.- Plants (Cucurbit Veg Group)	A,B	Cucumbers - 00095264 Pumpkins - 00095264
860.1500	171-4K	Mag. of Res.- Plants Citrus Fruits Group	A,B	00084326, 00095260
860.1500	171-4K	Mag. of Res.- Plants (Pome Fruits Group)	A,B	Apples - 00044555, 00088978, 00095264 Pears - 00044555, 43445601
860.1500	171-4K	Mag. of Res.- Plants (Stone Fruits Group)	A,B	Cherries - 00044555, 00077120, data gap Nectarines - 00044555, 00095179 Peaches - 00044555, 00095179 Plums (fresh prunes) - 00044555
860.1500	171-4K	Mag. of Res.- Plants (Small Fruits and Berries Group)	A,B	Bluberry - 00164187 Caneberries - PP#7E3557 Cranberries - 00108813 Grapes - 00085785, 00126713, 00134499, PP#3F02872/3H05393 Strawberries - 00095271, 40131302
860.1500	171-4K	Mag. of Res.- Plants (Tree Nuts Group)	A,B	00132786, 00044555, 00116675, 41424401
860.1500	171-4K	Mag. of Res.- Plants Cereal Grains Group	A,B	Corn, field, grain - 00070509 Corn, sweet (K+CWHR) - 00095216, 42245904 Sorghum, grain (milo) - 00046785, 00095249, 42245905, data gap for aspirated grain fractions Wheat, grain - data gap for aspirated grain fractions

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REQUIREMENT			USE PATT	CITATION(S)
860.1500	171-4K	Mag. of Res.- Plants (Forage, Fodder, and Straw of Cereal Grains Group)	A,B	Corn, Fodder - 00070509, 00078962, data gap Corn, Forage - 00070509, 00078962, data gap Sorghum, Fodder (milo) - 00046785, 00158569, Sorghum, Forage (milo) - 00046785, 00158569, Wheat, forage - PP#3F2947/FAP#3H5411, data gap for aspirated grain fractions Wheat, straw - PP#3F2947/FAP#3H5411, data gap for aspirated grain fractions
860.1500	171-4K	Mag. of Res.- Plants (Non-grass Animal Feeds (forage, fodder, straw, and hay) Group)	A,B	Alfalfa, forage - 00125686, 00158567, 00158568, 41739001 Alfalfa, hay - 00125686, 00158567, 00158568, 41739001
860.1500	171-4K	Mag. of Res.- Plants (Miscellaneous Commodities)	A,B	Asparagus - 00094088 Bananas - 00125686 Cherimoya - PP#7E3536 Cottonseed - 00095373, 40131303, data gap for cotton gin by-products Dates - 00162109 Feijoa (pineapple guava) - PP#7E3536 Figs - 00098580 Kiwifruits - 00115260 Mint - 00034031 Mushrooms - 00129295 Peanuts - 00025942, 00083840, 00095263 Sapote - PP#7E3536 Sugarcane - 42645401 Sunflower - 00084845, 42245906, 43181401 Tobacco - 40265201
860.1500	171-4K	Mag. of Res.- Plants (Crops Grown Solely for Seed)	A,B	Clover forage, seed and hay - data gap Grass forage and hay - data gap

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REQUIREMENT			USE PATT	CITATION(S)
860.1520	171-4L	Magnitude of the Residues in Processed Food/Feed	A,B	Alfalfa - 00125686, 00158567, 00158568 Apples - 00044555, 00088978, 00095264 Citrus - 00084326 Corn, field - 00084266, 42649002 Corn, sweet - 42649002 Cottonseed - 00037455 Grapes - 00085785, 00126713, 00134499 Mint - 00034031 Peanuts - 00025942, 00083840, 00095263 Plums - 00044555 Sorghum - 00046785, 00095249 Soybeans - 00095270 Sugar beet - 00039641, 00101566 Sugarcane - 42645401 Sunflower - 00084846, 42245906, 43181401 Tomatoes - 00095251 Wheat - PP#3F2947/FAP#3H5411
860.1460	171-4I	Magnitude of Residue in Food Handling Establishments	A,B	00090562, 00090563
OTHER				
810.1000	90-1	Usage Data for hydraulic handheld equipment	A,B	Data gap for usage data of amount of ai handled per day, per season and types of equipment.
810.1000	90-1	Usage Data for high pressure hand-wand equipment	A,B	Data gap for usage data of amount of ai handled per day, per season and types of equipment.
810.1000	90-1	Usage Data for groundboom applications to sodfarms	A,B	Data gap for usage data of acres treated per day at the 3 lb/A rate on sodfarms.
810.1000	90-1	Usage Data for greenhouse activities	A,B	use pattern information, timing of application relative to post-application activities

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REQUIREMENT		USE PATT	CITATION(S)
201-1	Droplet Size Spectrum	A,B	43760606, 43760607, 43786902
202-1	Drift Field Evaluation	A,B	41887501, 43786903
	Incident data	A,B	43798001, 44039901, 44186301, 44245801

Appendix C.Technical Support Documents

Appendix C. TECHNICAL SUPPORT DOCUMENTS

Additional documentation in support of this RED is maintained in the OPP docket, located in Room 119, Crystal Mall #2, 1921 Jefferson Davis Highway, Arlington, VA. It is open Monday through Friday, excluding legal holidays, from 8:30 am to 4 pm.

The docket initially contained preliminary risk assessments and related documents as of August 10, 1998. Sixty days later the first public comment period closed. The EPA then considered comments, revised the risk assessment, and added the formal "Response to Comments" document and the revised risk assessment to the docket on June 16, 1999.

All documents, in hard copy form, may be viewed in the OPP docket room or downloaded or viewed via the Internet at the following site:

www.epa.gov/pesticides/op

These documents include:

HED Documents:

1. David Soderberg (USEPA/OPPTS/OPP/HED). Acute Dietary Risk Assessment for Chlorpyrifos, Revised after Public Comments. June 22, 2000.

2. David Soderberg (USEPA/OPPTS/OPP/HED). Chronic Dietary Exposure Assessment for Chlorpyrifos RED with Updated Values for Anticipated Residues, Revised after Public Comments. June 22, 2000.

3. Steven A. Knizner (USEPA/OPPTS/OPP/HED). Chlorpyrifos - Revised Product and Residue Chemistry Chapters of the HED Chapter of the RED. June 20, 2000.

4. Tim Leighton (USEPA/OPPTS/OPP/HED). Agricultural and Occupational Exposure Assessment and Recommendations for the RED Document for Chlorpyrifos. June 19, 2000.

EFED Document:

1. William Rabert (USEPA/OPPTS/OPP/EFED). EFED Review of Lorsban-4E, Lock-On, and Lorsban 15G Label Changes. July 31, 2001.

Appendix D. Citations Considered to be Part of the Data Base Supporting the Interim Reregistration Decision (Bibliography)

Appendix D. CITATIONS CONSIDERED TO BE PART OF THE DATA BASE SUPPORTING THE INTERIM REREGISTRATION DECISION (BIBLIOGRAPHY)

GUIDE TO APPENDIX D

1. CONTENTS OF BIBLIOGRAPHY. This bibliography contains citations of all studies considered relevant by EPA in arriving at the positions and conclusions stated elsewhere in the Reregistration Eligibility Document. Primary sources for studies in this bibliography have been the body of data submitted to EPA and its predecessor agencies in support of past regulatory decisions. Selections from other sources including the published literature, in those instances where they have been considered, are included.

2. UNITS OF ENTRY. The unit of entry in this bibliography is called a "study". In the case of published materials, this corresponds closely to an article. In the case of unpublished materials submitted to the Agency, the Agency has sought to identify documents at a level parallel to the published article from within the typically larger volumes in which they were submitted. The resulting "studies" generally have a distinct title (or at least a single subject), can stand alone for purposes of review and can be described with a conventional bibliographic citation. The Agency has also attempted to unite basic documents and commentaries upon them, treating them as a single study.

3. IDENTIFICATION OF ENTRIES. The entries in this bibliography are sorted by Master Record Identifier, or "MRID" number. This number is unique to the citation, and should be used whenever a specific reference is required. It is not related to the six-digit "Accession Number" which has been used to identify volumes of submitted studies (see paragraph 4(d)(4) below for further explanation). In a few cases, entries added to the bibliography late in the review may be preceded by a nine character temporary identifier. These entries are listed after all MRID entries. This temporary identifying number is also to be used whenever specific reference is needed.

4. FORM OF ENTRY. In addition to the Master Record Identifier (MRID), each entry consists of a citation containing standard elements followed, in the case of material submitted to EPA, by a description of the earliest known submission. Bibliographic conventions used reflect the standard of the American National Standards Institute (ANSI), expanded to provide for certain special needs.

   a Author. Whenever the author could confidently be identified, the Agency has chosen to show a personal author. When no individual was identified, the Agency has shown an identifiable laboratory or testing facility as the author. When no author or laboratory could be identified, the Agency has shown the first submitter as the author.

   b. Document date. The date of the study is taken directly from the document. When the date is followed by a question mark, the bibliographer has deduced the date

from the evidence contained in the document. When the date appears as (1999), the Agency was unable to determine or estimate the date of the document.

c. Title. In some cases, it has been necessary for the Agency bibliographers to create or enhance a document title. Any such editorial insertions are contained between square brackets.

d. Trailing parentheses. For studies submitted to the Agency in the past, the trailing parentheses include (in addition to any self-explanatory text) the following elements describing the earliest known submission:

(1) Submission date. The date of the earliest known submission appears immediately following the word "received."

(2) Administrative number. The next element immediately following the word "under" is the registration number, experimental use permit number, petition number, or other administrative number associated with the earliest known submission.

(3) Submitter. The third element is the submitter. When authorship is defaulted to the submitter, this element is omitted.

(4) Volume Identification (Accession Numbers). The final element in the trailing parentheses identifies the EPA accession number of the volume in which the original submission of the study appears. The six-digit accession number follows the symbol "CDL," which stands for "Company Data Library." This accession number is in turn followed by an alphabetic suffix which shows the relative position of the study within the volume.

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Chlorpyrifos Bibliography

Appendix E. Generic Data Call-In

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See the following table for a list of generic data requirements. Note that a complete Data Call-In (DCI), with all pertinent instructions, is being sent to registrants under separate cover.

The following documents are part of the Generic Data Call-in.

DCI Response

Requirements Status and Registrant's Response

Footnotes and Key Definitions for Guideline Requirements

Appendix F. Product Specific Data Call-In

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See attached table for a list of product-specific data requirements. Note that a complete Data Call-In (DCI), with all pertinent instructions, is being sent to registrants under separate cover.

Appendix G. EPA's Batching of Chlorpyrifos Products for Meeting Acute Toxicity Data Requirements for Reregistration

Appendix G. EPA'S BATCHING OF CHLORPYRIFOS PRODUCTS FOR MEETING ACUTE TOXICITY DATA REQUIREMENTS FOR REREGISTRATION

In an effort to reduce the time, resources and number of animals needed to fulfill the acute toxicity data requirements for reregistration of products containing ***Chlorpyrifos*** as an active ingredient, the Agency has batched products which can be considered similar for purposes of acute toxicity. Factors considered in the sorting process include each product's active and inert ingredients (identity, percent composition and biological activity), type of formulation (e.g., emulsifiable concentrate, aerosol, wettable powder, granular, etc.), and labeling (e.g., signal word, use classification, precautionary labeling, etc.). Note the Agency is not describing batched products as "substantially similar" since some products with in a batch may not be considered chemically similar or have identical use patterns.

Using available information, batching has been accomplished by the process described in the preceding paragraph. Notwithstanding the batching process, the Agency reserves the right to require, at any time, acute toxicity data for an individual product should need arise.

Registrants of products within a batch may choose to cooperatively generate, submit or cite a single battery of six acute toxicological studies to represent all the products within that batch. It is the registrants' option to participate in the process with all other registrants, only some of the other registrants, or only their own products within in a batch, or to generate all the required acute toxicological studies for each of their own products. If the registrant chooses to generate the data for a batch, he/she must use one of the products within the batch as the test material. If the registrant chooses to rely upon previously submitted acute toxicity data, he/she may do so provided that the data base is complete and valid by to-days standards (see acceptance criteria attached), the formulation tested is considered by EPA to be similar for acute toxicity, and the formulation has not been significantly altered since submission and acceptance of the acute toxicity data. Regardless of whether new data is generated or existing data is referenced, the registrants must clearly identify the test material by EPA Registration Number. If more than one confidential statement of formula (CSF) exists for a product, the registrant must indicate the formulation actually tested by identifying the corresponding CSF.

In deciding how to meet the product specific data requirements, registrants must follow the directions given in the Data Call-In Notice and its attachments appended to the RED. The DCI Notice contains two response forms which are to be completed and submitted to the Agency within 90 days of receipt. The first form, "Data Call-in Response, " asks whether the registrant will meet the data requirements for each product. The second form, "Requirements Status and Registrant's Response," lists the product specific data required for each product, including the standard six acute toxicity tests. A registrant who wishes to participate in a batch must decide whether he/she will provide the data or depend on someone else to do so. If the registrant supplies the data to support a batch of products, he/she must select the one of the following options: Developing data (Option 1), Submitting an existing Study (Option 4), Upgrading an existing Study (Option 5), or Citing an Existing Study (Option). If a registrant depends on another's data, he/she must choose

among: Cost sharing (Option 2), Offers to Cost Share (Option 3) or Citing an Existing Study (Option 6). If a registrant does not want to participate in a batch, the choices are Options 1, 4, 5 or 6. However, a registrant should know that choosing not to participate in a batch does not preclude other registrants in the batch from citing his/her studies and offering to cost share (Option 3) those studies.

Two hundred twenty four products were found which contain ***Chlorpyrifos*** as the active ingredient. These products have been placed into 27 batches and a "No Batch" category in accordance with the active and inert ingredients and type of formulation. Please note that this batching scheme may not apply to products with CSFs that have been revised after generation of this document.

Batch 1	EPA Reg. No.	Percent active ingredient	Formulation Type
	4787-38	99.7	Solid
	4787-40	98.5	Solid
	4748-41	97.0	Solid
	11678-58	97.0	Solid
	34704-826	99.0	Solid
	42519-23	97.0	Solid
	62719-353	97.0	Solid
	62719-355	99.0	Solid
	70907-19	99.3	Solid

Batch 2	EPA Reg. No.	Percent active ingredient	Formulation Type
	1812-446	62.5	Liquid
	4787-37	62.2	Liquid
	4787-39	61.9	Liquid
	51036-350	61.5	Liquid
	62719-77	62.5	Liquid
	62719-349	62.5	Liquid
	62719-351	62.5	Liquid
	70907-17	60.6	Liquid

Batch 3	EPA Reg. No.	Percent active ingredient	Formulation Type
	7501-29	50.0	Solid
	34704-693	50.0	Solid
	62719-38	50.0	Solid

Batch 4	EPA Reg. No.	Percent active ingredient	Formulation Type
	62719-39	50.0	Solid
	62719-68	50.0	Solid
	62719-72	50.0	Solid
	62719-221	50.0	Solid
	62719-255	50.0	Solid
	62719-352	50.0	Solid
	70907-8	50.0	Solid

Batch 5	EPA Reg. No.	Percent active ingredient	Formulation Type
	655-499	44.8	Liquid
	829-280	44.9	Liquid
	1022-543	44.9	Liquid
	1386-649	44.9	Liquid
	34704-66	41.2	Liquid
	51036-122	42.8	Liquid
	51036-154	44.7	Liquid
	60061-82	44.9	Liquid
	60061-108	44.9	Liquid

Batch 6	EPA Reg. No.	Percent active ingredient	Formulation Type
	10163-158	40.7	Liquid
	19713-504	45.0	Liquid
	19713-518	44.9	Liquid
	19713-520	40.2	Liquid

	51036-216	44.7	Liquid
	51036-291	44.7	Liquid
	51036-294	44.7	Liquid
	62719-382	42.0	Liquid
	66222-3	44.9	Liquid
	66222-17	44.9	Liquid
	66222-19	40.7	Liquid
	67760-7	44.6	Liquid
	67760-27	44.2	Liquid
	67760-28	44.2	Liquid
	70907-3	45.0	Liquid
	70904-4	45.0	Liquid
	70907-7	45.0	Liquid
	70907-13	45.0	Liquid
	70907-18	45.0	Liquid

Batch 7	EPA Reg. No.	Percent active ingredient	Formulation Type
	19713-300	44.9	Liquid
	42519-19	44.9	Liquid
	42519-21	44.9	Liquid
	62719-11	44.9	Liquid
	62719-35	44.9	Liquid
	62719-69	44.9	Liquid
	62719-220	44.9	Liquid
	62719-245	44.9	Liquid
	62719-254	44.9	Liquid

Batch 8	EPA Reg. No.	Percent active ingredient	Formulation Type
	655-466	24.6	Liquid
	829-279	24.7	Liquid

	28293-200	24.1	Liquid
	51036-152	24.6	Liquid
	66222-5	24.5	Liquid
	66222-6	24.9	Liquid

Batch 9	EPA Reg. No.	Percent active ingredient	Formulation Type
	42519-20	24.8	Liquid
	51036-257	24.6	Liquid
	62719-65	24.8	Liquid
	67760-6	24.7	Liquid
	67760-31	24.7	Liquid

Batch 10	EPA Reg. No.	Percent active ingredient	Formulation Type
	62719-166	23.5	Liquid
	62719-167	23.5	Liquid

Batch 11	EPA Reg. No.	Percent active ingredient	Formulation Type
	499-367	20.0	Liquid
	499-419	20.0	Liquid

Batch 12	EPA Reg. No.	Percent active ingredient	Formulation Type
	10350-22	20.0	Liquid
	62719-88	20.0	Liquid
	62719-364	20.0	Liquid

Batch 13	EPA Reg. No.	Percent active ingredient	Formulation Type
	19713-505	15.0	Solid
	62719-383	15.0	Solid
	70907-5	15.0	Solid

Batch 14	EPA Reg. No.	Percent active ingredient	Formulation Type
	19713-521	15.0	Solid
	66222-18	15.0	Solid

Batch 15	EPA Reg. No.	Percent active ingredient	Formulation Type
	829-290	12.9	Liquid
	1386-615	12.6	Liquid
	28293-210	12.6	Liquid
	62719-380	12.6	Liquid

Batch 16	EPA Reg. No.	Percent active ingredient	Formulation Type
	655-764	2.32	Solid
	769-825	2.5	Solid
	1386-653	2.0	Solid
	8378-34	2.32	Solid
	9198-39	2.5	Solid
	9198-127	2.32	Solid
	10404-15	2.32	Solid
	28293-201	2.5	Solid
	32802-22	2.32	Solid
	34704-423	2.0	Solid
	51036-247	2.5	Solid
	51036-259	2.32	Solid
	51036-264	2.32	Solid
	53883-52	2.5	Solid

Batch 17	EPA Reg. No.	Percent active ingredient	Formulation Type
	829-292	2.5	Solid
	62719-276	2.5	Solid

Batch 18	EPA Reg. No.	Percent active ingredient	Formulation Type
	769-679	1.0	Solid
	769-726	1.0	Solid
	829-291	1.0	Solid
	1386-652	1.0	Solid
	8329-26	1.0	Solid
	8378-33	1.14	Solid
	8378-46	1.0	Solid
	9198-68	1.0	Solid
	9198-132	0.97	Solid
	9198-167	1.34	Solid
	10404-67	1.0	Solid
	10404-81	0.97	Solid
	28293-202	1.0	Solid
	32802-20	1.14	Solid
	32802-49	1.0	Solid
	34704-448	1.0	Solid
	51036-153	1.0	Solid
	51036-220	1.0	Solid
	62719-54	1.0	Solid
	62719-210	1.0	Solid

Batch 19	EPA Reg. No.	Percent active ingredient	Formulation Type
	8378-26	0.92	Solid
	8378-27	1.14	Solid
	9198-32	0.92	Solid
	10404-27	0.97	Solid
	32802-21	1.14	Solid
	62719-271	1.0	Solid

Batch 20	EPA Reg. No.	Percent active ingredient	Formulation Type
	655-766	0.5	Solid
	829-223	0.5	Solid
	829-272	0.5	Solid
	2724-487	0.5	Solid
	4822-153	0.5	Solid
	4822-335	0.03	Solid
	4822-411	0.528	Solid
	8329-23	0.5	Solid
	8378-28	0.5	Solid
	8848-61	0.5	Solid
	9198-137	0.5	Solid
	9688-67	0.50	Solid
	32802-19	0.7	Solid
	32802-39	0.5	Solid
	34704-55	0.5	Solid
	47006-5	0.5	Solid
	51036-117	0.5	Solid
	51036-263	0.5	Solid
	53883-48	0.5	Solid
	62719-14	0.5	Solid

Batch 21	EPA Reg. No.	Percent active ingredient	Formulation Type
	228-161	0.7	Solid
	8378-42	0.7	Solid
	8378-43	0.5	Solid
	8378-44	0.6	Solid
	9198-82	0.52	Solid
	9198-84	0.65	Solid
	9198-85	0.71	Solid

	9198-166	0.55	Solid
	10404-29	0.74	Solid
	10404-40	0.42	Solid
	35512-36	0.67	Solid
	62719-316	0.7	Solid

Batch 22	EPA Reg. No.	Percent active ingredient	Formulation Type
	572-329	0.5	Liquid
	10088-84	0.5	Liquid
	28293-99	0.5	Liquid
	62719-89	0.4	Liquid
	62719-90	0.2	Liquid

Batch 23	EPA Reg. No.	Percent active ingredient	Formulation Type
	10088-94	Chlorpyrifos- 0.5 Resmethrin - 0.11	Liquid
	28293-121	Chlorpyrifos - 0.5 Resmethrin - 0.11	Liquid

Batch 24	EPA Reg. No.	Percent active ingredient	Formulation Type
	655-786	Chlorpyrifos-0.5 PBO-0.26 Pyrethrins- 0.052	Liquid
	11474-66	Chlorpyrifos - 0.5 PBO- 0.26 Pyrethrins - 0.052	Liquid
	28293-87	Chlorpyrifos - 0.5 PBO- 0.26 Pyrethrins - 0.052	Liquid

Batch 25	EPA Reg. No.	Percent active ingredient	Formulation Type
	28293-142	Chlorpyrifos - 0.5 N-octyl bicycloheptene dicarboximide -0.4 Allethrin - 0.05	Liquid
	28293-149	Chlorpyrifos - 0.5 N-octyl bicycloheptene dicarboximide - 0.4 Allethrin - 0.05	Liquid

Batch 26	EPA Reg. No.	Percent active ingredient	Formulation Type
	11474-40	Chlorpyrifos-0.5 N-octyl bicycloheptene dicarboximide - 0.4 Allethrin- 0.054	Liquid

	11474-93	Chlorpyrifos- 0.5 N-octyl bicycloheptene dicarboximide- 0.4 Allethrin - 0.054	Liquid

Batch 27	EPA Reg. No.	Percent active ingredient	Formulation Type
	9198-98	Chlorpyrifos- 0.57 Benefin - 0.77 Trifluralin - 0.38	Liquid
	9198-99	Chlorpyrifos- 0.57 Benefin - 0.38 Trifluralin- 0.19	Liquid

No Batch	EPA Reg. No.	Percent active ingredient	Formulation Type
	499-405	Chlorpyrifos- 8.0 Cyfluthrin - 1.6	Liquid
	499-413	Chlorpyrifos- 0.5	Liquid
	665-441	Chlorpyrifos- 13.0 Dichlorvos- 4.82	Liquid
	1386-613	Chlorpyrifos- 6.97	Liquid
	7501-31	Chlorpyrifos-30.0	Liquid
	8329-18	Chlorpyrifos- 24.6	Liquid
	8329-20	Chlorpyrifos- 19.36	Liquid
	8329-24	Chlorpyrifos- 13.6	
	8329-36	Chlorpyrifos- 12.0 Permethrin- 4.0	Liquid
	9198-168	Chlorpyrifos-0.92	Solid
	9198-200	Chlorpyrifos- 0.45 Pendimethalin- 0.68	Solid
	9444-184	Chlorpyrifos- 0.5	Liquid
	9444-202	Chlorpyrifos- 0.50	Liquid
	9688-131	Chlorpyrifos- 0.50 Sulfluramid- 1.0	Liquid
	10088-85	Chlorpyrifos- 0.5 PBO-0.1 Pyrethrins-0.05 N-octyl bicycloheptene dicarboximide- 0.166	Liquid
	11474-55	Chlorpyrifos- 0.5 PBO- 0.260 Pyrethrins - 0.052	Liquid
	11474-90	Chlorpyrifos -0.5 PBO- 0.260 Pyrethrins - 0.052	Liquid
	13283-14	Chlorpyrifos- 5.0	Liquid
	13283-17	Chlorpyrofos-7.0	Solid
	26693-2	Chlorpyrifos- 2.0	Liquid
	28293-203	Chlorpyrifos- 1.0	Solid
	28293-204	Chlorpyrifos- 44.4	Liquid
	28293-205	Chlorpyrifos- 12.6	Liquid

	28293-265	Chlorpyrifos- 6.7	Liquid
	34704-65	Chlorpyrifos- 22.4	Liquid
	39039-2	Chlorpyrifos- 5.0 Cypermethrin-7.0 PBO- 3.5	Solid
	39039-6	Chlorpyrifos-9.5 Diazinon- 30.0	Solid
	45600-1	Chlorpyrifos- 0.86	Liquid
	48273-14	Chlorpyrifos- 44.9	Liquid
	51036-300	Chlorpyrifos- 15.0	Solid
	55431-1	Chlorpyrifos- 42.4	Liquid
	60061-100	Chlorpyrifos- 0.1 3-Iodo-2-Propynyl butyl Carbamate- 0.5	Liquid
	62719-34	Chlorpyrifos- 15.0	Solid
	62719-47	Chlorpyrifos- 44.9	Liquid
	62719-79	Chlorpyrifos- 22.9	Liquid
	62719-293	Chlorpyrifos- 75.0	Solid
	62719-295	Chlorpyrifos-30.0	Solid
	62719-350	Chlorpyrifos- 22.8	Liquid
	62719-354	Chlorpyrifos-30.0	Liquid
	66222-4	Chlorpyrifos-2.3	Solid
	67517-36	Chlorpyrifos-9.4 Permethrin- 7.2 PBO- 2.0	Solid
	67760-10	Chlorpyrifos- 43.2	Liquid
	67760-14	Chlorpyrifos- 15.0	Solid

Appendix H. List of Registrants Sent this Data Call-In

ATTACHMENT A

Appendix I. List of Available Related Documents and Electronically Available Forms

Appendix I. LIST OF AVAILABLE RELATED DOCUMENTS AND ELECTRONICALLY AVAILABLE FORMS

Pesticide Registration Forms are available at the following EPA internet site:

http://www.epa.gov/opprd001/forms/

Pesticide Registration Forms (These forms are in PDF format and require the Acrobat reader)

Instructions

1. Print out and complete the forms. (Note: Form numbers that are bolded can be filled out on your computer then printed.)

2. The completed form(s) should be submitted in hardcopy in accord with the existing policy.

3. Mail the forms, along with any additional documents necessary to comply with EPA regulations covering your request, to the address below for the Document Processing Desk.

DO NOT fax or e-mail any form containing 'Confidential Business Information' or 'Sensitive Information.'

If you have any problems accessing these forms, please contact Nicole Williams at (703) 308-5551 or by e-mail at williams.nicole@epa.gov.

The following Agency Pesticide Registration Forms are currently available via the internet: at the following locations:

8570-1	Application for Pesticide Registration/Amendment	http://www.epa.gov/opprd001/forms/8570-1.pdf
8570-4	Confidential Statement of Formula	http://www.epa.gov/opprd001/forms/8570-4.pdf
8570-5	Notice of Supplemental Registration of Distribution of a Registered Pesticide Product.	http://www.epa.gov/opprd001/forms/8570-5.pdf
8570-17	Application an Experimental Use Permit	http://www.epa.gov/opprd001/forms/8570-17.pdf
8570-25	Application for/Notification of State Registration of a Pesticide To Meet a Special Local Need	http://www.epa.gov/opprd001/forms/8570-25.pdf
8570-27	Formulator's Exemption Statement	http://www.epa.gov/opprd001/forms/8570-27.pdf
8570-28	Certification of Compliance with Data Gap Procedures	http://www.epa.gov/opprd001/forms/8570-28.pdf

8570-30	Pesticide Registration Maintenance Fee Filing_	http://www.epa.gov/opprd001/forms/8570-30.pdf
8570-32	Certification of Attempt to Enter into an Agreement with other Registrants for Development of Data	http://www.epa.gov/opprd001/forms/8570-32.pdf
8570-34	Certification with Respect to Citations of Data (PR Notice 98-5)	http://www.epa.gov/opppmsd1/PR_Notices/pr98-5.pdf
8570-35	Data Matrix (PR Notice 98-5)	http://www.epa.gov/opppmsd1/PR_Notices/pr98-5.pdf
8570-36	Summary of the Physical/Chemical Properties (PR Notice 98-1)	http://www.epa.gov/opppmsd1/PR_Notices/pr98-1.pdf
8570-37	Self-Certification Statement for the Physical/Chemical Properties (PR No 98-1)	http://www.epa.gov/opppmsd1/PR_Notices/pr98-1.pdf

Pesticide Registration Kit www.epa.gov/pesticides/registrationkit/

Dear Registrant:

For your convenience, we have assembled an online registration kit which contains the following pertinent forms and information needed to register a pesticide product with the U.S. Environmental Protection Agency's Office of Pesticide Programs (OPP):

1. The Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and the Federal Food, Drug and Cosmetic Act (FFDCA) as Amended by the Food Quality Protection Act (FQPA) of 1996.

2. Pesticide Registration (PR) Notices

   a. 83-3 Label Improvement Program--Storage and Disposal Statements
   b. 84-1 Clarification of Label Improvement Program
   c. 86-5 Standard Format for Data Submitted under FIFRA
   d. 87-1 Label Improvement Program for Pesticides Applied through Irrigation Systems (Chemigation)
   e. 87-6 Inert Ingredients in Pesticide Products Policy Statement
   f. 90-1 Inert Ingredients in Pesticide Products; Revised Policy Statement
   g. 95-2 Notifications, Non-notifications, and Minor Formulation Amendments
   h. 98-1 Self Certification of Product Chemistry Data with Attachments (This document is in PDF format and requires the Acrobat reader.)

   Other PR Notices can be found at http://www.epa.gov/opppmsd1/PR_Notices

3. Pesticide Product Registration Application Forms (These forms are in PDF format and will require the Acrobat reader).

a. EPA Form No. 8570-1, Application for Pesticide Registration/Amendment
b. EPA Form No. 8570-4, Confidential Statement of Formula
c. EPA Form No. 8570-27, Formulator's Exemption Statement
d. EPA Form No. 8570-34, Certification with Respect to Citations of Data
e. EPA Form No. 8570-35, Data Matrix

4. General Pesticide Information (Some of these forms are in PDF format and will require the Acrobat reader).

a. Registration Division Personnel Contact List
B. Biopesticides and Pollution Prevention Division (BPPD) Contacts
A. Antimicrobials Division Organizational Structure/Contact List
d. 53 F.R. 15952, Pesticide Registration Procedures; Pesticide Data Requirements (PDF format)
e. 40 CFR Part 156, Labeling Requirements for Pesticides and Devices (PDF format)
f. 40 CFR Part 158, Data Requirements for Registration (PDF format)
g. 50 F.R. 48833, Disclosure of Reviews of Pesticide Data (November 27, 1985)

Before submitting your application for registration, you may wish to consult some additional sources of information. These include:

1. The Office of Pesticide Programs' website.

2. The booklet "General Information on Applying for Registration of Pesticides in the United States", PB92-221811, available through the National Technical Information Service (NTIS) at the following address:

National Technical Information Service (NTIS)
5285 Port Royal Road
Springfield, VA 22161

The telephone number for NTIS is (703) 605-6000.

3. The National Pesticide Information Retrieval System (NPIRS) of Purdue University's Center for Environmental and Regulatory Information Systems. This service does charge a fee for subscriptions and custom searches. You can contact NPIRS by telephone at (765) 494-6614 or through their website.

4. The National Pesticide Information Center (NPIC) can provide information on active ingredients, uses, toxicology, and chemistry of pesticides. You can contact NPIC by telephone at 1-800- 858-7378 or through their website: http://npic.orst.edu.

The Agency will return a notice of receipt of an application for registration or amended registration, experimental use permit, or amendment to a petition if the applicant or

petitioner encloses with his submission a stamped, self-addressed postcard. The postcard must contain the following entries to be completed by OPP:

1. Date of receipt;
2. EPA identifying number; and
3. Product Manager assignment.

Other identifying information may be included by the applicant to link the acknowledgment of receipt to the specific application submitted. EPA will stamp the date of receipt and provide the EPA identifying file symbol or petition number for the new submission. The identifying number should be used whenever you contact the Agency concerning an application for registration, experimental use permit, or tolerance petition.

To assist us in ensuring that all data you have submitted for the chemical are properly coded and assigned to your company, please include a list of all synonyms, common and trade names, company experimental codes, and other names which identify the chemical (including "blind" codes used when a sample was submitted for testing by commercial or academic facilities). Please provide a chemical abstract system (CAS) number if one has been assigned.

Documents Associated with this RED

The following documents are part of the Administrative Record for this RED document and may be included in the EPA's Office of Pesticide Programs Public Docket. Copies of these documents are not available electronically, but may be obtained by contacting the person listed on the respective Chemical Status Sheet.

1. Health Effects Division and Environmental Fate and Effects Division Science Chapters, which include the complete risk assessments and supporting documents.
2. Detailed Label Usage Information System (LUIS) Report.

ATTACHMENT B

U.S. ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

Office of Chemical Safety
and Pollution Prevention

MEMORANDUM

DATE: July 13, 2012

SUBJECT: **Chlorpyrifos, PC Code 059101, DP Barcode 399483 and 399485**; Evaluation of the Potential Risks from Spray Drift and the Impact of Potential Risk Reduction Measures

PC Code: 059101	**DP Barcodes:** D399483 & 399485
Decision No.: 461963	**Registration No.:** varied
Petition No.: N/A	**Regulatory Action:** Registration Review
Risk Assessment Type: Spray Drift	**Case No.:** N/A
TXR No.: N/A	**CAS No.:** 2921-88-2
MRID No.: NA	**40 CFR:** N/A

FROM:

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THROUGH:

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TO:

Joel Wolf, Chemical Review Manager
Pesticide Re-Evaluation Division (7508P)

ATTACHMENT B

Table of Contents

Executive Summary

This analysis supplements the June 2011 *Chlorpyrifos Preliminary Human Health Risk Assessment for Registration Review* (HHRA) where limited monitoring data indicate risks to bystanders.[1] Bystanders are those who live on, work in or frequent areas adjacent to treated fields. Spray drift can be characterized as the movement of aerosols and volatile components away from the treated area as a result of the application process. Bystanders can be exposed to spray drift directly or by contact with resulting deposited residues. Spray drift of chlorpyrifos was examined using Tier I and II spray drift models to evaluate the impact of varied application conditions.[2] The degree of such impacts is governed by many processes (*e.g.*, application method, nozzles used, release height) and the conditions at the time of application (*e.g.*, windspeed and direction). Timing and frequency of exposures are also important. Chlorpyrifos is generally not applied multiple times in a year on a per acre basis, so spray drift is sporadic for adjacent properties (most applications are once per year). Residues also quickly degrade on plant surfaces. As such, single day exposures for bystanders are the focus of this analysis.

Chlorpyrifos labels indicate sprays should not directly or through drift contact workers or other persons. Some also indicate sprays should not drift to structures people occupy, associated property, parks or recreation areas. Buffer zones up to 150 feet from the field edge are also required to protect aquatic areas. Ensuring these provisions are adhered to is primarily the responsibility of the user, which is reinforced through training, compliance assistance, and enforcement actions, if needed. The degree of drift in field situations is difficult for users to ascertain at times because observing drift to prevent all deposits and potential exposure situations can be complex and at times physically impossible. These issues have been discussed extensively in efforts to develop good management practices and spray drift labeling language.[3]

This analysis focuses on chlorpyrifos applications that comply with application rates and methods identified on product labeling. As explained below, such applications may still result in drift. This analysis did not focus on circumstances leading to the majority of reported drift incidents since it is EPA's assessment that the majority of reported incidents resulted from applications that were plainly not in compliance with label requirements.

Results indicate spray drift from application of chlorpyrifos using current label requirements generally results in risk estimates of concern for locations immediately adjacent to treated fields. While risk estimates are of concern adjacent to a field edge for all application methods, implementing buffer zones as appropriate for residential areas (*e.g.*, similar to those in place to address aquatic risk concerns) could alleviate many of the potential human health risks.[4] An additional analysis shows shorter buffers coupled with drift reduction technologies and practices could further alleviate risks in some cases (*e.g.*, lower application rates, larger droplets, and larger swath displacement).

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[1] www.regulations.gov (docket ID EPA-HQ-OPP-2008-0850-0025)
[2] Peer reviewed modeling approaches were used http://www.epa.gov/scipoly/sap/tools/atozindex/spray.htm and, http://www.epa.gov/pesticides/science/residential-exposure-sop.html.
[3] http://www.epa.gov/pesticides/factsheets/spraydrift.htm provides links to pertinent sources of information
[4] http://www.cdms.net/ldat/ld02A011.pdf (pg 3) as an example

1 Introduction

Chlorpyrifos is an organophosphate (OP) insecticide, acaricide, and miticide that controls many foliar and soil borne insect pests.[5] Chlorpyrifos was first registered in 1965 for use on a variety of food and feed crops. As chlorpyrifos use expanded in agriculture, it also grew in the residential marketplace. Residential use has since diminished because such uses, for the most part, except those with a low exposure potential, were voluntarily cancelled and removed from labels in 2001. Even given this trend, exposures can occur in residential environments or other areas frequented by the general public through spray drift. Exposures can also occur as a result of volatilization of chlorpyrifos from agricultural crops or other areas after application. These exposure pathways were addressed in the 2011 HHRA using empirical monitoring data. This analysis supplements the previous risk assessment by using spray drift models to estimate the potential exposure from a number of different chlorpyrifos application scenarios. This approach allows for a broader understanding of the potential for risk associated with chlorpyrifos spray drift.

Off-target movement of chlorpyrifos can occur via many types of pathways and its magnitude is governed by many processes. Sprays that are off-target can directly lead to exposure. They can also deposit on surfaces where contact with residues can lead to exposure (*e.g.*, children playing on lawns next to treated fields). The potential exposure and risk estimates from these residues can be calculated using drift modeling and methods employed for typical residential risk assessments. There is precedent for using this approach as it mirrors the methods used in a response to a petition to cancel 14 pesticides as well as methods used to develop buffer zone estimates for use of two organophosphate insecticides on orchard crops in the Pacific Northwest.[6]

Volatilization can occur during or after application. It can result from aerosols evaporating during application, while deposited sprays are still drying, or after application as dried deposited residues volatilize (*e.g.*, possibly via co-distillation). This document addresses only those exposures associated with spray drift and whatever volatilization is anticipated to occur during application.

AgDRIFT (V2.1.1) and AgDISP (V8.26) were used to provide deposition values for residential lawns, as a fraction of the application rate, at different distances downwind of a treated field. AgDRIFT (V2.01) was used to estimate air concentrations at different distances downwind of a treated field. Distances up to 125 feet away were considered in both of these analyses. Both the drift and exposure methods used to quantify the potential risks from pesticide spray drift were previously peer reviewed through the FIFRA Scientific Advisory Panel).[6]

[5] http://www.cdms.net/manuf/mprod.asp?mp=11&lc=0&ms=3691&manuf=11 (see Dursban or Lorsban labels)
[6] http://www.epa.gov/scipoly/sap/tools/atozindex/residentexp.htm, http://www.epa.gov/scipoly/sap/meetings/2009/100609meeting.html, and http://www.epa.gov/scipoly/sap/tools/atozindex/spray.htm

In addition to spray drift deposition and air concentration determinations, other aspects of this analysis are based on methods commonly used for evaluating risks from pesticides used on turf because the risk concern is based on drift depositing on lawns adjacent to treatment sites. This scenario is utilized because it represents the highest potential for exposure associated with spray drift and it also considers different human lifestages, including those associated with children at different developmental stages.[7] Data from a chlorpyrifos study that determined turf residue levels and dissipation rates after application were also available.[8] These data were used in conjunction with the standard residential methods to estimate exposure from treated turf. Finally, the endpoints and points of departure (PODs) used for this analysis are similar to those used in the 2011 HHRA. The PODs used were defined based on route-specific studies that can be considered of high quality and suitable for risk assessment purposes. In some cases, PODs differ slightly from the 2011 HHRA because these values are more appropriate for evaluating infrequent, single day exposures like those anticipated with spray drift.[9]

Section 2, **Use Patterns** provides an overview of how chlorpyrifos is used with a specific focus on spray drift. **Section 3**, **Context** includes information on related topics such as the nature and frequency of incidents and efforts to develop best management practices for spray drift that must be considered when interpreting the results of this analysis. **Section 4**, **Methods and Inputs** provides more details on the specific calculations and values used to define risks from chlorpyrifos spray drift. The calculated risk estimates are provided in **Section 5**, **Results**. Issues that should be considered, specific to how the risk estimates from spray drift were calculated, are presented in **Section 6**, **Issues for Consideration**. **Section 7**, **Conclusions and Recommendations**, presents the conclusions of this assessment and describes potential mitigation measures that could be adopted and their impact on the calculated risk estimates.

2 Use Patterns

Chlorpyrifos is a broadly used organophosphate insecticide currently registered for the control of various insects. Registered use sites include: food crops including fruits, nuts, vegetables, and grains and non-food crops such as golf course turf, industrial sites, greenhouses, nurseries, sod farms, and wood products. Public health uses include aerial and ground-based fogger treatments to control adult mosquitoes. There is a wide range of currently registered chlorpyrifos application rates. In general, current maximum chlorpyrifos application rates do not exceed 4 lb ai/a nationwide; however, single application rates greater than 4 lb ai/a are currently permitted for some specific use patterns. For example, a single chlorpyrifos application of 6 lb ai/a is permitted on citrus in a limited number of counties in California. In addition there are also spot or trunk drench applications that may result in single application rates higher than 4 lb ai/a. A summary of the chlorpyrifos use patterns including application rates is presented in **Appendix A**.

Other information related to how chlorpyrifos is used germane to the exposure potential from spray drift and characterizing potential mitigation options is presented below. Specifically,

[7] http://www.epa.gov/pesticides/science/residential-exposure-sop.html
[8] (Stafford et al,1999) Determination of Dislodgeable Foliar Residues on Turf Treated With Formulations Containing Chlorpyrifos (EPA MRID 44829601).
[9] Should changes in the hazard evaluation occur they will be reflected in this analysis as appropriate (*e.g.*, impact of findings of 2012 FIFRA SAP, http://www.epa.gov/scipoly/sap/meetings/2012/041012meeting.html).

Section 2.1 summarizes the requirements to ensure applications are made using a *best management practices* approach. Typical chlorpyrifos use practices are presented in **Section 2.2**. **Section 2.3** discusses the significance of aerial application as a method for using chlorpyrifos (*e.g.*, prevalence on certain crops).

2.1 Label Requirements Related to Spray Drift

Current chlorpyrifos labels provide guidance for users on how to reduce the potential for spray drift under actual use conditions. Many of these criteria have long been recognized in the scientific literature as key factors related to how changes in application technology will impact spray patterns and the subsequent drift. The Dow AgroSciences product Lorsban Advanced label has been used to illustrate this guidance (EPA Reg. No. 62719-591). All current chlorpyrifos labels contain similar labeling language. This guidance serves as the basis for the drift modeling described below. The label language under the section, ***Spray Drift Management***, is included as **Appendix B**. A few key elements are summarized below.

The label prohibits drift from contacting people, the structures they occupy, and associated property at any time. The label also indicates avoiding drift is the responsibility of the applicator and that they should consider equipment and weather related factors when making applications. Buffer zones are also specified for applications around aquatic areas as described in **Table 1**.

Table 1. Current Buffer Zones for Chlorpyrifos Around Aquatic Areas

Application Method	Required Buffer Zone (feet)
Groundboom	25
Chemigation	25
Airblast	50
Aerial (fixed wing or helicopter)	150

The Lorsban label also provides best management practices to reduce spray drift. Guidance is provided that describes how applicators should consider the conditions of the site when deciding how and when to make an application (*e.g.*, presence of possible atmospheric inversion conditions, location of sensitive sites such as residences relative to site location). Additionally, the label provides information for specific application methods. Some examples include:

- ***For Aerial Applications*** **-** Nozzles must produce a medium or coarser droplet size (255 to 340 microns volume median diameter) per ASABE Standard 572 under application conditions and applications must not be made at a height greater than 10 feet above the top of the target plants unless required for aircraft safety.

- ***For Groundboom Applications*** **-** Choose only nozzles and pressures that produce a medium or coarse droplet size (255 to 400 microns volume median diameter), per ASABE Standard 572 and do not apply product when wind speed exceeds 10 mph.

•***For Orchard Airblast Applications*** **-** Apply only when wind speed is 3 to 10 mph and spray the outside rows of orchards from outside in, directing the spray into the orchard and shutting off nozzles on the side of the sprayer away from the orchard.

2.2 Typical Uses

This section provides use information such as the application rate and number of applications for national-level total chlorpyrifos used (in all formulations and with all application methods combined) on use sites where information was available. Details are available in **Appendix C**.

Approximately 8 million pounds of chlorpyrifos are used annually in agriculture. Total chlorpyrifos usage varies widely with average percent crop treated (PCT) - a measure of the area treated with chlorpyrifos in the survey years (2006-2010, unless otherwise noted) - as low as 1% for several crops and as high as 62 % (for apples) (**Appendix C**). The five crops with the highest PCT are apples (62% PCT), broccoli (55%), walnuts (46%), onions (45%), and cauliflower (41%).

Corn and soybean are the two crops with highest total amount of chlorpyrifos used in terms of average pounds applied – both show over 1,000,000 lb used across the years (**Appendix C**). Crops that have low total chlorpyrifos usage (arbitrarily defined as less than 20,000 pounds used, on average) but show relatively large area treated (arbitrarily defined as greater than 25% average percent crop treated, or PCT) include strawberries, asparagus, and cauliflower.

Nineteen crops (out of a total of 39 crops surveyed) show average application rates that are approximately 1 lb ai/a. These crops are apples, almonds, broccoli, cabbage, cauliflower, cherries, grapefruit, hazelnuts, lemons, oranges, peaches, peanuts, pears, plums/prunes, pumpkins, squash, sweet corn, tobacco, and walnuts. The frequency of application, on average, for most crops is one time per year. In a few crops, the annual average frequency of application is approximately two times per year. This information supports the supposition that focusing on single day exposures on a per acre basis from spray drift is an appropriate premise.

2.3 Use of Aerial Application

The usage information on aerial applications was obtained from a private marketing research database that stores results of annual market surveys that cover the continental United States and the majority of crops produced (**Table 2**).[10] These should be considered in the context of the percent crop treated information described above in **Section 2.2**. The surveys utilize sampling procedures that result in statistically valid results for most crops. However, due to limited sample size, precision is reduced for some crops and for crops with limited use of chlorpyrifos. Data from 2006 to 2010 were used in this analysis, and the results presented are an average of the five years of data. The rate range information for each crop was obtained by querying the proprietary source database.

[10] A similar analysis was conducted in order to evaluate the import of ground applications. A table analogous to Table 2, excerpted from that memo, is included as part of Appendix C for informational purposes. It is not included here since ground applications are predominant and they do not represent the most significant risk concern, as will be detailed below.

The application method rate range data was summed over the five years for each rate range in which applications were reported and the percentage of applications in each rate range was calculated for aerial applications. The maximum observed rate for aerial applications is the last column in **Table 2**. The maximum label rates may be higher than these maximum observed so it is possible that higher rates might have been used. However the maximums reported were the highest rates observed over a five year period so a significant number of applications at higher rates are unlikely.

The approximate percentage of the crop treated by air was calculated. This calculation assumes that the average number of applications were the same for aerial applications as for all applications. This assumption is justified because the average number of applications for most crops is once per year.

For most crops the acreage treated with aerially applied chlorpyrifos represents only a small percentage (<10%) of the total crop acreage. Nevertheless, for some crop aerial applications are important (*e.g.*, 80% of sunflower chlorpyrifos applications are made by air). Nine percent of asparagus acres were estimated to have been treated by air. An estimated five percent of sunflowers and sweet corn were treated with aerial applications of chlorpyrifos and for all other crops less than five percent of planted acres received aerial applications of chlorpyrifos. Maximum observed application rates varied significantly among crops, with some receiving maximum application rates of one pound of active ingredient per acre while maximum application rates for lemons and oranges were 5.7 and 6 pounds per acre respectively. Citrus crops (alone) have an additional label, limited for use only in the San Joaquin Valley of California (Fresno, Tulare, Kern, Kings, and Madera counties), that permits use of rates up to 6 lb ai/a; all other citrus has a maximum application rate of 4 lb ai/a.

Table 2. Aerial Applications of Chlorpyrifos to Crops (2006-2010)

Crop	% of Applications Applied by Air	% of Total Crop Acreage Treated by Air	Average Lbs AI Year Applied by Air	Average Application Rate (lb ai/a) for Aerial Applications	Maximum Rate (lb ai/a) Observed for Aerial Applications (rounded up)
Alfalfa	31	1	130,000	0.5	1.0
Almonds	14	3	60,000	1.9	2.0
Apples	5	3	15,000	1.0	2.0
Asparagus	24	9	4,000	1.0	1.0
Beans (Snap)	<1	<1	<100	0.8	0.8
Broccoli	<1	<1	<100	1.0	1.0
Cabbage	<1	<1	<100	1.0	1.0
Cauliflower	1	1	200	0.9	1.0
Cherries	2	1	700	0.9	1.0
Corn	2	<1	20,000	0.8	2.0
Cotton	8	<1	40,000	0.8	1.0
Cucumbers	None				
Dry Beans	17	<1	2,000	0.5	1.0
Grapefruit	2	<1	700	1.5	1.5

Crop	% of Applications Applied by Air	% of Total Crop Acreage Treated by Air	Average Lbs AI Year Applied by Air	Average Application Rate (lb ai/a) for Aerial Applications	Maximum Rate (lb ai/a) Observed for Aerial Applications (rounded up)
Grapes, Raisin	None				
Grapes, Table	<1	<1	150	1.0	1.0
Grapes, Wine	None				
Hazelnuts	32	4	1,000	1.0	2.0
Lemons	4	2	3,000	2.7	5.7
Onions	<1	<1	<100	1.0	1.0
Oranges	7	2	30,000	1.8	(2.5) 6.0
Peaches	5	1	2,000	1.1	3.0
Peanuts	2	<1	2,500	1.4	1.8
Pears	3	1	800	2.0	2.0
Peas (Fresh)	None				
Pecans	15	4	40,000	0.9	(1.5) 2.0
Peppers	7	<1	<100	0.3	0.3
Plums/Prunes	None				
Potatoes	8	<1	200	1.0	1.0
Pumpkins	None				
Sorghum	18	<1	2,000	0.5	0.8
Soybeans	16	1	600,000	0.4	(0.8) 1.0
Squash	1	<1	<100	3.0	3.0
Strawberries	None				
Sugar Beets	13	1	10,000	0.6	1.2
Sunflowers	80	5	50,000	0.5	(0.7) 1.0
Sweet Corn	43	5	30,000	0.6	2.0
Tobacco	None				
Walnuts	6	3	25,000	1.9	(2.0) 3.0
Wheat, Spring	38	1	50,000	0.4	(0.5) 1.0
Wheat, Winter	30	1	110,000	0.4	(0.5) 1.0
Note: For some crops there are single/very few observations so this should be considered in the interpretation of the information presented in this table.					

3 Context

In late 2009, a policy paper focusing on the development of updated methods for conducting risk assessments for workers and for those who live in proximity to agricultural pesticide use sites was released.[11] Status updates of this policy have also been presented at several meetings of the Pesticide Program Dialogue Committee (PPDC). A public comment period was utilized to solicit input from stakeholders.[12] A key aspect of this policy is that the Agency committed to

[11] http://www.epa.gov/pesticides/health/worker-rsk-assmnt.html, also see www.regulations.gov docket ID EPA-HQ-OPP-2009-0889-0002

[12] For example, refer to http://www.epa.gov/oppfead1/cb/ppdc/2011/april/april2011.html

developing an approach for characterizing potential exposure and risk estimates associated with off-target chemical movement via spray drift. The plan for implementing this policy is through the Registration Review process with an initial focus on uses with a perceived high risk potential.

There are many ways the potential risk associated with pesticide spray drift can be considered in the regulatory process. It is important to consider all mitigation alternatives in order to be expedient, thorough, and cost effective (*e.g.*, changes to application rates, changes in application parameters, buffer zones). For example, if a pesticide can be used to treat lawns directly without a risk estimate of concern, then calculating risks via spray drift, given similar application rates, may not be needed because the risks from drift could be the same or lower than that associated with the direct application to lawns. In other cases, like with chlorpyrifos, uses in the residential marketplace are restricted. In these situations, many factors are important for describing the potential for risks via drift including: application methods for particular crops, prevalence of use on a particular crop or application method, need for a pesticide relative to the pest of concern, environmental fate characteristics of a pesticide, and the toxicological characteristics of the pesticide. Many of these factors were considered in this analysis and are described.

In addition to label stipulations that dictate how a pesticide can be used and the factors that can impact exposures via drift (described above in **Section 2.1**), other relevant information should be considered including a discussion of incident rates and causes. Incidents are important for characterizing the results of this analysis and are summarized based on the 2011 human health risk assessment in **Section 3.1**. The Agency and stakeholders have also been working to develop guidance related to managing spray drift using a best practices approach as described below in **Section 3.2**.[13] These efforts have prompted methods development for evaluating the performance of drift reduction technologies, an important basis for developing viable mitigation options.

3.1 Incident Rates

The conclusions of the most recent evaluation of the incidents associated with chlorpyrifos use indicate incidents have declined substantially among residential users. This would be expected given that most residential uses, except products that are known to produce lower levels of exposure, were voluntarily cancelled and removed from labels at the end of 2001.[14] At the same time, occupational incident rates have remained fairly constant over time. The conclusions that were made associated with the 2011 HHRA indicate that the overall numbers and severity of incidents associated with chlorpyrifos use do not represent an immediate public health concern (*e.g.*, serious non-reversible effects or deaths have not been seen as a result of chlorpyrifos drift). For the most part, they also have been caused by some manner of accident, operator error, or misuse (*e.g.*, direct drift onto individuals in adjacent properties). These incidents differ from the drift calculations contained in this analysis because this analysis is based on the premise of label compliance.

An evaluation of the National Institute for Occupational Safety and Health (NIOSH) *Sentinel Event Notification System for Occupational Risks* (SENSOR) information indicates issues

[13] http://www.epa.gov/pesticides/factsheets/spraydrift.htm
[14] Recore and Oo (6/27/11) Chlorpyrifos: Tier II Incident Report (D388406)

associated with application appear to be the leading cause of the reported incidents involving chlorpyrifos, and responsible for 46% of those incidents reported from 1998 to 2007. The California Pesticide Illness Surveillance Program (PISP) also reported incidents associated with chlorpyrifos and it appears that drift from adjacent fields may be the cause of most of the reported incidents, unlike the SENSOR results. Spray drift is responsible for 56% of the cases reported to PISP from 1999 to 2008 (**Figure 1**). These incidents appear to be predominantly caused by drift contacting workers in fields adjacent to application events, which is a misuse based on current labels. An example includes a large 2007 incident (*i.e.*, impact on large number of people) in Tulare County California associated with an airblast application of chlorpyrifos to almonds that impacted 28 workers who were working in an adjacent vineyard.

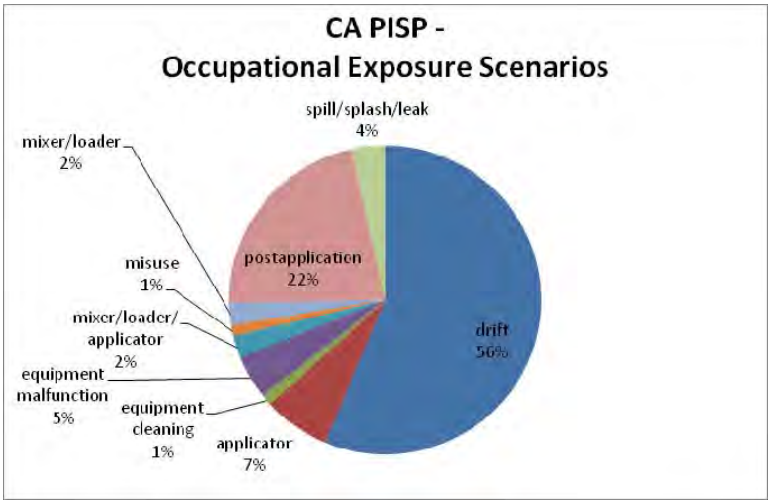

Figure 1. Chlorpyrifos CA PISP Occupational Incident Summary

A separate published analysis of drift incidents developed by NIOSH SENSOR investigators was also considered. This analysis indicated chlorpyrifos has a relatively high rate of incidents due to drift compared to the other pesticides addressed in the analysis.[15] The authors indicated 49 out of 1809 total cases (*i.e.*, incidents) across all pesticides considered, where the cause was related to a single pesticide, could definitively be associated with exposure to chlorpyrifos. Ten of these were defined as being of moderate to high severity. Additionally, the authors noted the Tulare County incident described above was one of the 10 largest (*i.e.*, most impacted people) incidents that occurred between 1998 and 2006 associated with spray drift.

3.2 Stewardship

As mentioned previously, the issue of spray drift, including developing a workable strategy for ensuring proper stewardship by users, has been a longstanding issue. These efforts have been formalized with the development of a draft PR notice (2009-X: Draft Pesticide Drift Labeling) which provides guidance to pesticide registrants on how to minimize drift and to protect people, non-target organisms, and the environment from adverse effects that may be caused by off-target

[15] Lee *et al* (2011) Acute Pesticide Illnesses Associated with Off-Target Pesticide Drift from Agricultural Applications — 11 States, 1998–2006, available at the following: http://ehp03.niehs.nih.gov/article/info%3Adoi%2F10.1289%2Fehp.1002843

pesticide drift.[16] Current chlorpyrifos labels in many ways reflect the guidance outlined in the PR Notice 2009-X: Draft Pesticide Drift Labeling (see **Appendix B**). This includes overall guidance and recommendations for users to evaluate field and atmospheric conditions prior to and during application to ensure drift potential is minimized, particularly if there are areas nearby where people live or that could be considered sensitive.

The draft PR Notice also outlines a plan for developing a Drift Reduction Technology (DRT) program. DRTs represent many possible application technologies that offer the potential to reduce drift from pesticide applications. Even though some of these techniques are widely recognized for reducing drift potential, limited information exists to quantify their actual effectiveness. The DRT provides application equipment and spray adjuvant manufacturers with a test method and a process for voluntarily testing and validating technologies for drift reduction potential. Tested technologies that are proven to significantly reduce drift will be assigned a rating and listed on the Agency's website. Pesticide registrants will then be able to choose to recommend or require through pesticide product labels the use of validated DRTs for product application as a drift mitigation measure. EPA would consider label DRTs in its risk assessment and risk management decisions for the registration of these products. Labeling that cites DRTs could provide applicators who employ them more flexibility for modifying equipment and techniques in order to reduce drift potential for the specific use scenario. In effect, use of DRTs is a performance based approach that lets users employ the means at their disposal to reduce drift. *Note: DRTs are described in detail in the draft PR Notice if further information is desired.*

Managing the drift potential from applications of chlorpyrifos is a key concern. In the PR Notice example, different application parameters are associated with different possible buffer distances. Such application parameters include varying the droplet size, allowing for different wind speeds at application, and fixing boom size relative to the wingspan of a plane or the helicopter rotor. The evaluation of drift potential for chlorpyrifos and the associated risk estimates were developed in a manner that is consistent with the draft PR Notice and the DRT performance-based construct. A series of possible drift reduction measures and the associated impacts were evaluated as part of this analysis as potential refinements to existing chlorpyrifos labels and may also be used as a guide for the development of data consistent with the DRT approach.

Finally, many of the approaches that are currently being implemented as part of the overall strategy for reducing risk estimates associated with fumigants could also be effective for spray drift concerns.[17] This is especially true of some of the qualitative measures like enhanced training, site specific management plans, and record keeping requirements.

4 Methods and Inputs

The approach used to evaluate the drift potential and associated risks for chlorpyrifos is based on standard methods and inputs. **Section 4.1** describes the overall approach used for this spray drift assessment. **Section 4.2** describes the models used, inputs, and limitations used to define the degree of drift associated with each application method evaluated. **Section 4.3** summarizes the

[16] Available at www.regulations.gov (docket ID EPA-HQ-OPP-2009-0628-0002), also refer to (docket ID EPA-HQ-OPP-2009-0628-0003) which provides examples to aid with interpretation of the PR Notice

[17] http://www.epa.gov/opp00001/reregistration/soil_fumigants/implementing-new-safety-measures.html

toxicological characteristics of chlorpyrifos. **Section 4.4** describes the values and methods used to calculate the exposures, once the drift potential was defined, for risk assessment purposes. Similarly, **Section 4.5** describes the values and methods used for risk calculation purposes. Finally, **Section 4.6** describes the possible DRTs that were considered as viable measures for reducing risk estimates and how they were integrated into the analysis in order to illustrate their potential impacts.

4.1 Approach

Pesticide spray drift can potentially impact those who are in proximity to application events through a variety of possible exposure pathways. In order to evaluate the drift potential and associated risks, an approach based on drift modeling coupled with techniques used to evaluate residential uses of pesticides was utilized. Essentially, a residential turf assessment based on exposure to deposited residues has been completed for each scenario, but the source term used to define these residues was determined in a different manner and is based on the amount of spray drift that may occur as illustrated in **Figure 2**.

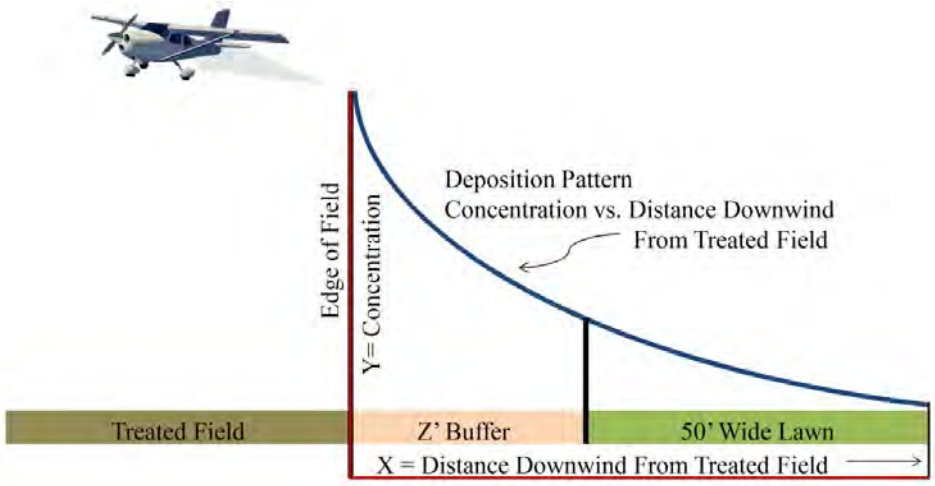

Figure 2. Conceptual Model of Exposure Pathway Associated With Spray Drift

In this case, drift modeling was completed for a variety of application scenarios including different application equipment, spray types, and windspeeds. This approach is also protective of other possible types of pathways because the exposures used as the basis for these calculations are protective of other possible exposure situations (*i.e.,* children's exposure on treated turf yields significantly higher estimates than other possible scenarios). It also is a logical exposure pattern that would reasonably be expected to occur with a spray drift event, given that children could play on lawns adjacent to treated fields. Along with these types of exposures, airborne concentrations of chlorpyrifos were also defined and used to estimate risks from inhalation exposure. For this calculation, concentrations were defined in the breathing zone (area around the nose, ~ 3 to 5 ft above ground for this analysis) at the edge of the 50' wide lawn closest to the treated field. Chlorpyrifos is generally not applied multiple times in a year on a per acre basis, so

spray drift is only anticipated sporadically on adjacent properties. As such, there is a focus in this assessment on single day exposures for individuals who may live on or frequent such properties.

4.2 Drift Modeling

Tier I spray drift analysis for aerial, ground, and orchard air blast applications of chlorpyrifos were completed using AgDRIFT[18] (v2.1.1) to investigate the deposition pattern of chlorpyrifos downwind of a treated field.[19] The AgDRIFT spray drift model is a Microsoft Windows-based computer program provided to the U.S. Environmental Protection Agency's Office of Pesticide Programs as a product of the Cooperative Research and Development Agreement (CRADA) between EPA's Office of Research and Development and the Spray Drift Task Force (SDTF). AgDRIFT predicts the motion of spray material released from aircraft, including the mean position of the material and the position variance about the mean as a result of turbulent fluctuations. AgDRIFT has undergone extensive validation including evaluation by a FIFRA Scientific Advisory Panel.[20, 21]

Tier I AgDRIFT is a regression model developed from SDTF data including more than 300 applications made in 10 field studies covering a range of application practices for each type of application (*i.e.*, aerial, ground, and orchard air blast applications). AgDRIFT has been formally validated by comparing its predicted outputs to 180 SDTF field trials. These data were generated for aerial applications to a distance of about 800 meters downwind of the application field edge. These field trials were conducted using pesticide (*i.e.*, dissolved or emulsifiable) in a liquid spray and therefore they may not fully represent spray drift resulting from encapsulated or granular based pesticide formulations. If these formulations (*e.g.*, dispersible capsules or dispersible granular) are applied using a liquid carrier and the droplet size distribution is the same as those droplet size distribution included in AgDRIFT the resulting spray drift values are assumed to be the same. Granular formulations have different drift patterns than for liquid sprays, and as such need to be considered in a separate analysis. However, from a human health perspective, off-site movement of granular products would result in lower risk estimates than those for liquid products (*e.g.*, less drift occurs and there is less exposure uptake for granules).

Deposition rates were determined as a fraction of the application rate (*i.e.*, how much of the target application rate is predicted to be deposited at various distances downwind of the treated field). This fraction of the application rate was adjusted based on the range of currently approved application rates (1 to 6 lb ai/a) for various crops to determine the average effective deposition

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[18] A User's Guide for AgDRIFT 2.1.1: A Tiered Approach for the Assessment of Spray Drift Pesticides (February 2003). AgDRIFT V2.01 was used to model inhalation exposure concentrations.

[19] AgDRIFT and AGDISP models: Models are available through Harold Thistle, US Forestry, hthistle@fs.fed.us.

[20] U.S. Environmental Protection Agency. Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel: Estimating Drinking Water Exposure as a Component of the Dietary Risk Assessment; Spray Drift Program, **December 10-11, 1997**.

[21] U.S. Environmental Protection Agency. Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel: Burholderia cepacia: Risk Assessment of a Biopesticide with Affinities to a Human Opportunistic Pathogen; A Consultation on Protocol Design to Assess Acute Neurotoxicity Studies following Oral Administration of Pesticides; Higher Tier Ecological Risk Assessment for Chlorfunapyr; Spray Drift - Review of Proposed Pesticide Deposition Curves to Adjacent Areas, **July 20-23, 1999**.

rate (lb ai/a) at various distances downwind of the treated field for each respective crop use. Input parameters used in the Tier I spray drift analysis are provided in **Table 3**.

Table 3. Tier I AgDRIFT Input Parameters

Parameter	Value	Comments
Aerial		
Equipment	Air Tractor AT-401	AgDRIFT default option
Release Height	10 ft	AgDRIFT default values
Wind Speed	10 mph	AgDRIFT default values / Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) restricts applications to periods when the wind speed is between 3-10 mph.
Surface Roughness	0.0075 m (0.3 in)	Default
Temperature	Not specified	AgDRIFT Tier I ground models are based on regression equations developed on empirical data.
Swath Displacement	0.3702 (F2M) 0.2781 (M2C) 0.2149 (C2VC)	AgDRIFT default values/ Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) restricts applications to droplet sizes greater than 255 µM.
Droplet Size	F2M, M2C, C2VC	Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) restrict applications to droplet sizes greater than 255 µM; therefore, only droplet distributions with a mean diameter ($D_{v0.5}$) ≥ 255 µM were selected for modeling.
Fine to Medium (F2M): Droplet size ($D_{v0.5}$) = 255 µM Medium to Coarse (M2C): $D_{v0.5}$ = 341 µM Coarse to Very Coarse (C2VC): $D_{v0.5}$ = 439 µM		
Ground[1]		
Equipment	Low Boom (LB) High Boom (HB)	AgDRIFT default options
Release Height	LB: 0.508 m (20 in) HB: 1.27 m (50 in)	AgDRIFT default values
Application Efficiency	LB: 99.27 HB: 99.22	AgDRIFT default values
Wind Speed	Not specified	AgDRIFT Tier I ground models are based on regression equations developed on empirical data.
Surface Roughness	Not specified	AgDRIFT Tier I ground models are based on regression equations developed on empirical data.
Temperature	Not specified	AgDRIFT Tier I ground models are based on regression equations developed on empirical data.
Swath Width	13.72 m (45 ft)	AgDRIFT default value
Droplet Size	F2M/C	Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) restrict applications to droplet sizes greater than 255 µM; therefore, only droplet distributions with a mean diameter ($D_{v0.5}$) ≥ 255 µM were selected for modeling.
Fine to Medium/Coarse (F2M/C): Droplet size ($D_{v0.5}$) = 341 µM (see AgDrift GUI screenshot in **Appendix D**) 1. Regression equations were developed from SDTF field study results with ground boom equipment.		
Orchard Air Blast[1]		
Equipment	Not specified	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Release Height	Not specified	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Application Efficiency	Normal: 99.85 Dense: 98.01 Sparse: 97.69 Vineyard: 99.73	AgDRIFT default/ Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) permits all these types of applications.

Parameter	Value	Comments
	Orchard: 98.63	
Wind Speed	Variable	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Surface Roughness	Not specified	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Temperature	Variable	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Swath Displacement	Not specified	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Droplet Size	Not specified	AgDRIFT Tier I orchard air blast model are based on regression equations developed on empirical data.
Orchard Type[2]	Normal, Dense, Sparse, Vineyard, and Orchard	AgDRIFT default/ Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) permits all these types of applications

1. Regression equations were developed from SDTF field study results with orchard blast equipment.
2. Normal (Stone and Pome Fruit, Vineyard): This composite orchard combines grape and apple orchards
   Dense (Citrus, Tall Trees): This composite orchard combines almond, orange, grapefruit, small grapefruit (mist blower) and pecan orchards.
   Sparse (Young, Dormant): This composite orchard combines small grapefruit and dormant apple orchards.
   Orchard: This composite orchard combines apple, almond, orange, grapefruit, small grapefruit, pecan, and dormant apple orchards.
   Vineyard: This composite curve combines grape air blast sprayer applications and may not apply to other application equipment.

4.3 Hazard Inputs

Chlorpyrifos inhibits the enzyme acetylcholinesterase which leads to accumulation of acetylcholine and ultimately to neurotoxicity. In previous risk assessments, including the 2011 risk assessment, the Agency concluded that data on the inhibition of cholinesterase (ChE) provided the most sensitive dose-response data for use in deriving points of departure (POD) for all durations, routes of exposure, and lifestages. The 2011 risk assessment included the typical scenarios used for risk assessment [acute dietary, chronic dietary, short-term (1-30 days), intermediate-term (1 month-6 months), and long-term (>6 months)]. This spray drift assessment relies on the endpoint selection from the 2011, with refinements to account for the focus on single day exposures from spray drift. Specifically, revisions were made to provide a more appropriate match between the duration of the toxicology endpoints to the duration of exposure. It is noted below where values varied from the 2011 risk assessment and the rationale for modifying them.

The toxicity database of laboratory animal studies spans multiple routes of exposure (oral, dermal, and inhalation), animal species, lifestages, and durations. The database consists of studies that address different durations of exposure from a single exposure day (acute) to subchronic and chronic toxicity. The metabolism and pharmacokinetics of chlorpyrifos are well-characterized due to a variety of studies in different species and lifestages. The studies used to complete this assessment are of high quality and provide information that are appropriate for evaluating risks associated with single day exposures which are the focus of this assessment. A comparative cholinesterase assay (CCA) provides information on comparative sensitivity in adult and juvenile rats from acute and repeated exposures to chlorpyrifos. The POD for acute dietary risk was defined using the CCA study. The POD was derived using a benchmark dose (BMD)

approach; details of the BMD analysis can be found in the 2011 risk assessment. This POD was also considered appropriate for children's mouthing behavior, as described later in this document, for a single day duration of exposure and is thus used here.

Dermal exposures were evaluated in this assessment using a POD derived from a dermal toxicity study where sacrifices occurred at 4 and 21 days of dosing. It is notable that the results from the 4 day sacrifice are similar to those from 21 days of dosing. In addition, a single dose dermal administration study conducted in human volunteers (Nolan, et al, 1982)[22] indicates that effects can occur with only a single dermal dose. This means that the resulting risks using the 4-day study can be reasonably characterized as being representative of single day dermal risk estimates.

Finally, inhalation risks were evaluated in the 2011 risk assessment using the POD derived from an acute inhalation toxicity study. In the previous assessment the POD was a HEC (Human Equivalent Concentration) calculated based on a 24 hour basis, but realistic exposure durations are much shorter, so the HEC was revised using a 2 hour duration. This 2 hour period was selected because it both represented a dosing time in the study and the potential duration of exposure from spray drift. *Note: The impact of using a 2 hour duration is discussed in more detail below, including how the exposure source is considered after the first hour of application given how the drift estimates are modeled.*

The endpoints and PODs used for this analysis are summarized in **Table 4**. A 1x FQPA safety factor (SF) was used in all cases, except for the 10X FQPA database uncertainty factor (UF) that was applied, to account for LOAEL to NOAEL extrapolation in the acute inhalation study because a NOAEL was not determined (LOAEL = Lowest observed adverse effect level and NOAEL = No observed adverse effect level). As discussed in the 2011 risk assessment, benchmark dose modeling of the acute inhalation study results was attempted, but the dose-response data were not amenable for this type of analysis.

Table 4. Toxicological Doses, Endpoints and Points of Departure for Chlorpyrifos Drift Risk Assessment

Exposure Scenario	Point of Departure (mg/kg/day)	Study and Toxicological Effects
Acute Dietary (all populations) [Also used in this case to represent single day Incidental Oral exposures]	BMDL10 = 0.36 UFA = 10x UFH = 10x FQPA SF = 1x Residential LOC for MOE=100	Inhibition of RBC ChE in male and female rat pups. Weight of evidence from several acute oral studies: • CCA Study (MRID 48139301) in the rat – PND 11 males and females

[22] This study was reviewed by Human Studies Review Board in 2009. Materials are available at: http://www.epa.gov/osa/hsrb/jun-24-25-2009-public-meeting.htm. Additionally it should be noted that a 2008 FIFRA Scientific Advisory Panel review indicated that this study provides useful information but is not appropriate for defining PODs http://www.epa.gov/scipoly/sap/meetings/2008/september/sap0908report.pdf .

Exposure Scenario	Point of Departure (mg/kg/day)	Study and Toxicological Effects
		• Data on PND17 males , Moser et al.(2006) • Qualitative support from Timchalk et al. (2006) and Zheng et al. (2000) studies
Dermal Short- (1 – 30 days) and Intermediate-Term (1 to 6 months) [Also used in this case to represent single day dermal exposures]	NOAEL = 5 mg/kg/day UF_A = 10x UF_H = 10x FQPA SF = 1x (residential) Residential LOC for MOE=100	Plasma and RBC ChE inhibition. 21-day dermal study (NOAEL) and 4 day probe study (LOAEL) at 10 mg/kg/day in adult rats (MRID 40972801).
Acute Inhalation [A 2 hour HEC has been calculated which better reflects actual exposure conditions and the available dosing regimen from the study.]	Inhalation LOAEL = 3.7 mg/m^3 HEC = 2.48 mg/m^3 UF_A = 3x UF_H = 10x FQPA UF_{DB} = 10x (LOAEL to NOAEL extrapolation (residential) Residential LOC for MOE=300	Lung ChE inhibition. Special acute inhalation study (MRID 48139303). (Aerosol) [2, 4, and 6 hour sacrifice data were used to define the 2 hour HEC]

4.4 Exposure Inputs

The exposure aspects of this analysis were completed based on the algorithms and input values specified in the recently revised (2012) *Standard Operating Procedures For Residential Risk Assessment (SOPs)*. These are publicly available and address many broad issues pertinent to the development of risk assessments for pesticides used in a residential environment.[23] These SOPs have been extensively peer reviewed and information on this process is also publicly available.[24] The calculations in this analysis were developed by considering the variety of possible pathways and all pertinent routes of exposure that could occur associated with spray drift. As a result of this process, two pathways were identified and used to evaluate the potential risks associated with spray drift including (1) inhalation exposures at discrete distances downwind of a treated field during application and (2) exposures from lawns adjacent to treated fields where drift occurs.

Inhalation exposure calculations were based on modeled air concentrations of chlorpyrifos in a vertical plane at specified distances downwind from the edge of a treated field. These represent 1

[23] http://www.epa.gov/pesticides/science/residential-exposure-sop.html and http://www.epa.gov/pesticides/science/EPA-OPP-HED_Residential%20SOPS_Feb2012.pdf

[24] http://www.epa.gov/scipoly/sap/meetings/2009/100609meeting.htm, http://www.epa.gov/scipoly/sap/meetings/1999/092199_mtg.htm, and http://www.epa.gov/scipoly/sap/meetings/1997/090997_mtg.htm

hour time weighted average concentrations during application and can be defined only for aerial applications. In this case these outputs were used to estimate breathing zone exposure concentrations. Specifically, these concentrations were estimated using Tier II in the AgDRIFT model (V2.01) with varied spray/application rates. The only modifications to the Tier II inputs were altering the volatile and nonvolatile components of the spray solution variables to account for the specific application rates and chlorpyrifos products considered in this assessment. Vertical flux planes were considered at different distances ranging from the edge of a treated field to 125 feet downwind of the field. This analysis is only possible for aerial applications because both the ground boom and airblast aspects of AgDRIFT are empirical in nature, thus making upper tier analyses, which provide this type of output, not possible. In order to account for dilution, a dissipation rate of 25 percent per hour was used. This is protective as defined by an analysis conducted using the well-mixed box model, which is described in the residential SOPs. Exposure concentrations were used to compare to the acute inhalation HEC presented in **Table 4** to calculate risks.

The calculations for the second pathway included in this analysis are based on the premise that chlorpyrifos residues drift away from a treated area, are deposited downwind (*e.g.*, on a lawn) and, then serve as a potential source of exposure. Once the amount of drift has been quantified, the resulting levels are considered like any pesticide intended for use on lawns. In this assessment, agricultural application rates are adjusted by the amount of drift averaged over a 50 foot wide lawn. These lawns are considered at different distances up to 300 feet (125 reported) downwind from a treated field (closest edge to the treated field defines the distance). In this assessment, dermal exposures were added to non-dietary incidental ingestion exposure sources to calculate total risks because the toxicological endpoint was similar for all routes of exposure (*i.e.*, cholinesterase inhibition).

When risks associated with lawn pesticides are calculated, if residue data referred to as turf transferable residues (TTRs) are available, they are used as the basis of the calculation. If TTRs are not available, they are estimated using an established default methodology as described in the SOPs. TTR data, generated when chlorpyrifos use on turf was allowable, are available in this case and have been used directly as prescribed in the SOPs. The chlorpyrifos TTR data were used in the 2011 risk assessment to evaluate other scenarios involving possible contact with treated turf, including deposits resulting from the mosquito adulticide use of chlorpyrifos and its use on golf courses. The data have been summarized and used for the purposes of this assessment in a manner similar to what was done in the 2011 risk assessment.

The available TTR study (MRID 44829601) was conducted in three states including California, Indiana, and Mississippi. This study examined the residues of chlorpyrifos on turf after application of Dursban® Pro (emulsifiable concentrate), Dursban® 50W (wettable powder), Dursban® 2.5G (granular) and Dursban® 1F (flowable granular). Single applications of the test products were made to the treatment plots using tractor mounted boom sprayers for the liquid applications. The liquid applications were made at a nominal application rate of 4 lb ai/a.[25] Duplicate samples were collected using the California roller technique at the following intervals:

[25] In the calculations, residue levels from this study are adjusted based on the amount of drift and the varied application rates considered in the assessment.

one day prior to the application, just after the application (2.5 hours), and days 1, 2, 4, and 7 after application. Samples were analyzed for residues of chlorpyrifos.[26] The overall average concurrent recovery was 98% ± 3%. The LOD and LOQ were reported as 0.001 µg/cm^2 and 0.003 µg/cm^2, respectively. The field study samples did not require correction for field fortification recovery. Average field fortification recoveries (corrected for concurrent recovery), at each of three fortification levels were greater than 90% at all sites. Dursban® Pro, a liquid formulation, results are presented in **Table 5**. The results for this formulation are presented because they represent the highest relative levels of residues resulting from the products used in this study. The chlorpyrifos dissipation half-life, at all sites, was approximately 14 hours or less.

Table 5. Chlorpyrifos (Dursban Pro EC) TTR Data Collected Using California Roller Method

Site	Target App Rate (lb ai/A)	Deposition At Appl. Rate (µg/cm^2)	Day of Application Mean TTR (µg/cm^2)	TTR as % App. Rate
CA	3.80	42.60	0.124	0.29
IN	3.98	44.62	0.090	0.20
MS	3.83	42.93	0.146	0.34
average	3.87	43.38	0.120	0.28

4.5 Risk Assessment

As there is a wide variety of use scenarios described on chlorpyrifos labels, all calculations in this assessment were completed using an approach that groups currently registered chlorpyrifos application rates and crops into appropriate categories. The use information included in this document is presented in order to provide context for the groupings. The application rates listed for the individual groupings represent the maximum single application rate for the crops in that grouping (see **Table 6**).

Table 6. Chlorpyrifos Application Groupings

Single Application Rate of Chlorpyrifos lb ai/a	Example Use Site	Comments
6	Citrus	Use only permitted in specific counties in Arizona and California
3.5 or 4	other citrus, grass grown for seed	Use permitted nationwide
2.3	Citrus	Aerial application used to control psyllid, the vector for citrus greening[1] (pest present in Florida and possibly California and Texas)
2	nursery and orchard trees, tree fruit, nuts	Use permitted nationwide
1	alfalfa, asparagus, sorghum,	Use permitted nationwide

[26] Chlorpyrifos oxon was not measured but this would not impact the results of this analysis because only the values immediately following application were considered in this assessment.

	strawberry, and sugarbeet	
1. Based on recent communications between EPA/OPP/PRD and DAS, et al. (May 17, 2012)		

In addition to these groupings, calculations were further delineated by application method and the types of spray patterns and spray conditions used. These are based on the pre-defined options incorporated into the AgDRIFT model, and they include:[27]

- Orchard Airblast (normal, dense, sparse, and vineyard foliage/canopy levels)[28]
- Aerial [spray droplet patterns/sizes include fine to medium ($Dv_{0.5}$ = 255 µM), medium ($Dv_{0.5}$ = 294 µM), medium to coarse ($Dv_{0.5}$ = 341 µM), coarse to very coarse ($Dv_{0.5}$ = 439 µM)]
- Groundboom [low (20 in) and high (50 in) booms, fine to medium/coarse spray pattern ($Dv_{0.5}$ = 341 µM)]

As described in detail above, two types of calculations were completed in this analysis. All of the calculations follow the guidance contained in the residential SOPs and the drift/deposition outputs described above. The appendices of this document contain the detailed algorithms and inputs used for this analysis, as well as the results.

4.6 Risk Reduction Options

The concept behind the development of the DRTs and the overall guidance of the PR notice for labeling related to spray drift described above is that pesticide applicators can control drift through the use of specific equipment and application parameters. These parameters include spray nozzle size, application pressure, vehicle type and configuration, release height, formulation choice, use of additives such as surfactants, and considering terrain as well as atmospheric conditions at the time of application. Additional spray drift modeling was completed for several droplet size distributions to investigate the impact that the aircraft, wind speed, swath displacement (the horizontal offset distance between the flight line and the target area), and crop canopy have on the deposition pattern of chlorpyrifos downwind of treated fields. These represent implementable means that applicators could potentially use to reduce drift

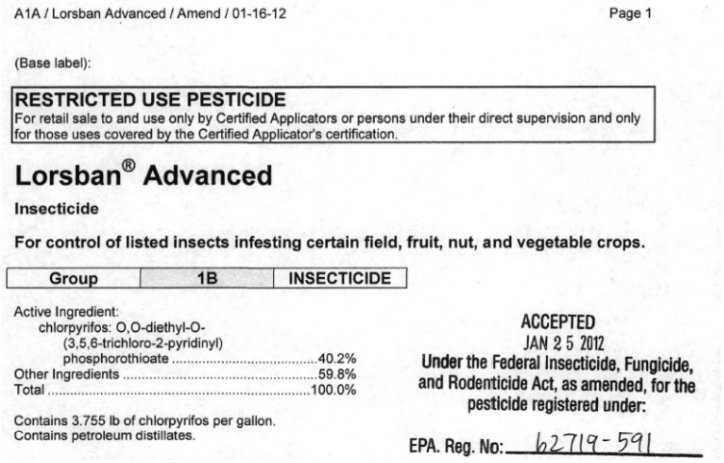
Tier II modeling for ground and orchard applications was not completed as AgDRIFT does not have the capability to refine the spray drift estimates for these application types; however, Tier II spray drift analysis can and was completed using AgDRIFT for aerial applications. As a Tier II assessment tool, AgDRIFT is a parameterized model that can be used to investigate specific aerial application parameters. For chlorpyrifos, aerial application parameters investigated

[27] Note: Orchard airblast and groundboom are only available for Tier I analysis. A "medium" droplet size ($Dv_{0.5}$ = 294 µM) is only attainable for analysis in Tier II.

[28] Normal (Stone and Pome Fruit, Vineyard): This composite orchard combines grape and apple orchards
Dense (Citrus, Tall Trees): This composite orchard combines almond, orange, grapefruit, small grapefruit (mist blower) and pecan orchards.
Sparse (Young, Dormant): This composite orchard combines small grapefruit and dormant apple orchards.
Orchard: This composite orchard combines apple, almond, orange, grapefruit, small grapefruit, pecan, and dormant apple orchards.
Vineyard: This composite curve combines grape air blast sprayer applications and may not apply to other application equipment.

included: aircraft (fixed and rotary winged), wind speed (10 and 5 mph), and swath displacement (default value based on droplet size see **Table 3** and 100%). AgDRIFT input values for the Tier II analyses are provided in **Table 7**.

Table 7. Tier II AgDRIFT Input Parameters

Parameter	Value	Comments
Aerial		
Equipment	Air Tractor AT-401 WASP Helicopter	There are no aircraft restrictions on chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591); therefore, one fixed and one rotary winged aircraft was chosen. The default airplane (fixed wing) used in the Tier I analysis was used for consistency between analyses. One of the two default Tier II options for helicopter (rotary wing) was selected by the assessor.
Release Height	10 ft	Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) restrict applications to 10 ft above the crop canopy.
Wind Speed	10 mph 5 mph	Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) limit applications to time periods when the wind speed is between 3 and 10 mph.
Surface Roughness	0.0075 m (0.3 in)	AgDRIFT default value
Temperature	86 °F	AgDRIFT default value
Swath Displacement	0.3702 and 1 (F2M) 0.3351 and 1(M) 0.2781 and 1 (M2C) 0.2489 (C) 0.2149 and 1 (C2VC) 0.2005 (VC)	While an upwind swath displacement is recommended on chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) a specific displacement value is not prescribed. The default Tier I value was selected by the assessor for consistency with the Tier I analysis. An alternative swath displacement of 100% was selected by the assessor to evaluate the impact of swath displacement.
Droplet Size	F2M, M, M2C, C, C2VC	Chlorpyrifos labels (*e.g.*, EPA Reg. No. 62719-591) restrict applications to droplet sizes greater than 255 µM; therefore, only droplet distributions with a mean diameter ($D_{v0.5}$) ≥ 255 µM were selected for modeling. These droplet sizes are consistent with the Tier I analysis.
Nonvolatile Rate	1.49 lb/a 1.35 lb/a	Chlorpyrifos label EPA Reg. No. 62719-591 (59.8% other ingredients /40.2% a.i. x 1 lb ai/a = 1.49 lb/a other ingredients based on data provided on the label as shown below).  A1A / Lorsban Advanced / Amend / 01-16-12 Page 1 (Base label): RESTRICTED USE PESTICIDE For retail sale to and use only by Certified Applicators or persons under their direct supervision and only for those uses covered by the Certified Applicator's certification. Lorsban® Advanced Insecticide For control of listed insects infesting certain field, fruit, nut, and vegetable crops. Group 1B INSECTICIDE Active Ingredient: chlorpyrifos: O,O-diethyl-O-(3,5,6-trichloro-2-pyridinyl) phosphorothioate ..... 40.2% Other Ingredients ..... 59.8% Total ..... 100.0% Contains 3.755 lb of chlorpyrifos per gallon. Contains petroleum distillates. ACCEPTED JAN 25 2012 Under the Federal Insecticide, Fungicide, and Rodenticide Act, as amended, for the pesticide registered under: EPA. Reg. No: 62719-591 Chlorpyrifos label EPA Reg. No. 66222-19 (57.5% other ingredients /42.5% a.i. x 1 lb ai/a = 1.35 lb/a other ingredients based on data provided on the label as shown below)

Parameter	Value	Comments
Aerial		
		CHLORPYRIFOS 4E AG For control of listed insects infesting certain field, fruit, nut, and vegetable crops. **ACTIVE INGREDIENT** **% BY WT.** Chlorpyrifos: 0,0-diethyl-0-(3,5,6-trichloro-2-pyridinyl) phosphorothioate ..... 42.5% **INERT INGREDIENTS*:** ..... 57.5% TOTAL 100.0% Contains 4 pounds of chlorpyrifos per gallon * Contains petroleum distillates Contains chlorpyrifos, the active ingredient used in Lorsban® 4E. Chlorpyrifos 4E AG is not manufactured or distributed by Dow AgroSciences LLC. EPA Reg. No. 66222-19 EPA Est. No. 11678-ISR-002 **Manufactured for:** Makhteshim Agan of North America, Inc. 4515 Falls of Neuse Rd., Suite 300 Raleigh, NC 27609 **NET CONTENTS: 2.5 GALLONS**
Spray Volume	2 gal/a	Minimum required spray volume
Carrier Type	Water	Chlorpyrifos labels (*e.g.*, EPA Reg. No. 66222-19)
Fine to Medium (F2M): Average Droplet size ($D_{v0.5}$) = 255 µM Medium (M): $D_{v0.5}$ = 294 µM Medium to Coarse (M2C): $D_{v0.5}$ = 341 µM Coarse (C) $D_{v0.5}$ = 385 µM Coarse to Very Coarse (M2C): $D_{v0.5}$ = 439 µM Very Coarse (VC): $D_{v0.5}$ = 478 µM Other input parameters were not changed or adjusted based on standard AgDRIFT recommendation via program prompts.		

The Tier I and Tier II analysis for aerial applications are based on bare ground applications. A Tier III analysis for aerial applications to investigate the effect that a crop canopy may have on the deposition pattern downwind of a treated field following an aerial application of chlorpyrifos was contemplated. However, the use of Tier III results are limited primarily for the following reasons: 1) canopy simulations in AgDISP are based on theoretical calculations and the results have not be validated (compared to field data); and 2) AgDISP simulations represent canopy structures that are expected to be different from chlorpyrifos use sites (*e.g.*, AgDISP simulations are based on a uniformly closed canopy which is generally not expected for tree crops in an orchard setting). These limitations were discussed with and acknowledged by the model developer, Dr. Harold Thistle with the U.S. Forest Service.[29] Tier III results are not presented in this document.

5 Results

The results of the analyses completed to evaluate the potential risks from chlorpyrifos spray drift are presented below. **Section 5.1** describes the different spray drift modeling outputs that were calculated for the current label prescribed application conditions as well as those that were calculated representing potential risk mitigation options. **Section 5.2** presents the inhalation risk estimates that were calculated. **Section 5.3** presents the risk estimates associated with contact of residues which potentially deposit on lawns adjoining treated fields.

[29] Personal communication on May 30, 2012

5.1 Drift Estimates

The spray drift deposition curves for the Tier I analysis for aerial applications are shown in **Figure 3**. This analysis includes fine to medium [average droplet size ($D_{v0.5}$) = 255 µM, which represents the current label], medium to coarse ($D_{v0.5}$ = 341 µM) and coarse to very coarse ($D_{v0.5}$ = 439 µM) droplet sizes. The estimated drift concentrations over a 50' wide lawn for various distances downwind from the treated field are provided in **Table 8**. These estimated drift concentrations are shown for a range of chlorpyrifos application rates (1 to 6 lb ai/a).

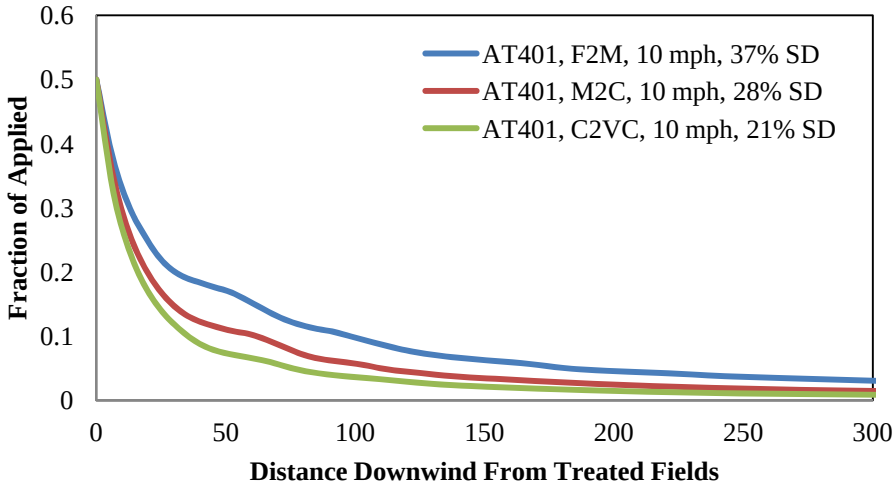


Figure 3. Tier I Spray Drift Analysis: Aerial Deposition Curves for Three Different Droplet Sizes

Table 8. Tier I Spray Drift Analysis: Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn Starting at Various Distances Downwind From a Field Treated with Chlorpyrifos Using Aerial Equipment

Application Rate (lb ai/a)	Droplet Size	Distance Downwind From Treated Field (feet)										
		0	10	25	50	75	100	125	150	200	250	300
		Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn										
6	F2M	1.64	1.23	1.02	0.77	0.58	0.46	0.38	0.32	0.25	0.20	0.17
	M2C	1.38	0.91	0.70	0.49	0.34	0.26	0.21	0.18	0.13	0.10	0.08
	C2VC	1.23	0.71	0.50	0.31	0.22	0.17	0.13	0.11	0.08	0.06	0.05
4	F2M	1.10	0.82	0.68	0.51	0.38	0.30	0.25	0.22	0.16	0.13	0.11
	M2C	0.92	0.60	0.46	0.32	0.22	0.17	0.14	0.12	0.08	0.07	0.05
	C2VC	0.82	0.48	0.33	0.21	0.15	0.11	0.22	0.07	0.05	0.04	0.03
2.3	F2M	0.63	0.47	0.39	0.29	0.22	0.18	0.14	0.12	0.09	0.08	0.06
	M2C	0.53	0.35	0.27	0.19	0.13	0.10	0.08	0.07	0.05	0.04	0.03
	C2VC	0.47	0.27	0.19	0.12	0.08	0.06	0.05	0.04	0.03	0.02	0.02
2	F2M	0.55	0.41	0.34	0.26	0.19	0.15	0.13	0.11	0.08	0.07	0.06
	M2C	0.46	0.30	0.23	0.16	0.11	0.09	0.07	0.06	0.04	0.03	0.03
	C2VC	0.41	0.24	0.17	0.10	0.07	0.06	0.04	0.04	0.03	0.02	0.02

1[a]	F2M	0.27	0.20	0.17	0.13	0.10	0.08	0.06	0.05	0.04	0.03	0.03
	M2C	0.23	0.15	0.12	0.08	0.06	0.04	0.03	0.03	0.02	0.02	0.01
	C2VC	0.20	0.12	0.08	0.05	0.04	0.03	0.02	0.02	0.01	0.01	0.01

Fine to Medium (F2M): Avg. Droplet size ($D_{v0.5}$) = 255 µM, Medium to Coarse (M2C): $D_{v0.5}$ = 341 µM, Coarse to Very Coarse (C2VC): $D_{v0.5}$ = 439 µM
The double box indicates the current aquatic buffer distance of 150 feet.
a. The average deposition concentration for a 50' wide lawn at each of the distances downwind. This is equivalent in terms of the average fraction of the applied that deposits over the same area [*e.g.*, 0.27 lb ai/a is equivalent to a fraction applied of 0.27 (or 27% of the agricultural application rate)].

Tier I results are presented in **Figure 4** and

Table 9 for fine to medium droplet sizes for applications using ground boom application equipment with high and low spray booms. The results suggest that potential spray drift exposure is higher near the field when a high boom is used as compared to a low boom; however, once several feet (≥10 ft) downwind the exposure resulting from the use of either boom is about the same.

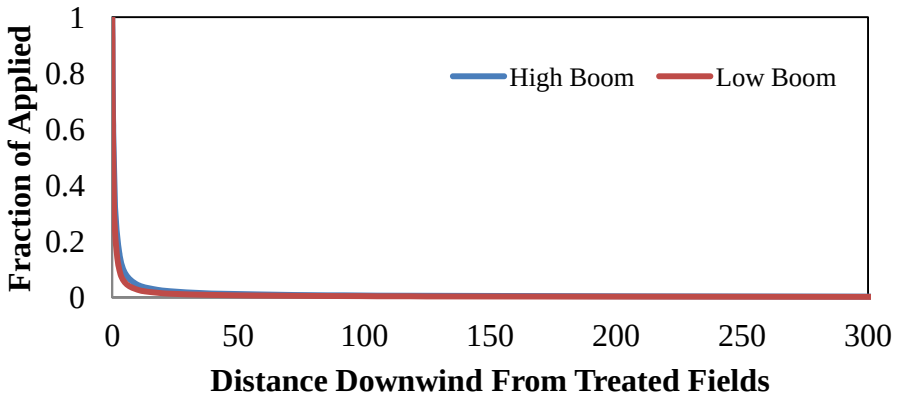


Figure 4. Tier I Spray Drift Analysis: Deposition Curves Resulting From the Application of Chlorpyrifos Using Ground Equipment

Table 9. Tier I Spray Drift Analysis: Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn Starting at Various Distances Downwind From a Field Treated with Chlorpyrifos Using Ground Equipment (high and low boom options)

Application Rate (lb ai/a)	Boom Height	Distance Downwind From Treated Field (feet)										
		0	10	25	55	75	100	125	150	200	250	300
		Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn										
6	High	1.63	0.11	0.08	0.05	0.04	0.04	0.03	0.03	0.02	0.02	0.02
	Low	1.37	0.07	0.05	0.03	0.03	0.02	0.02	0.02	0.01	0.01	0.01
4	High	1.08	0.07	0.05	0.04	0.03	0.02	0.02	0.02	0.01	0.01	0.01
	Low	0.91	0.04	0.03	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01
2	High	0.54	0.04	0.03	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.01
	Low	0.46	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.00
1[a]	High	0.27	0.02	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.00	0.00
	Low	0.23	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Low Boom 0.508 m (20 in), High Boom 1.27 m (50 in)
Fine to Medium/Coarse (F2M/C): Avg. Droplet size ($D_{v0.5}$) = 341 µM
The double box indicates the current aquatic buffer distance of 25 feet.

a. The average deposition concentration for a 50' wide lawn at each of the distances downwind. This is equivalent in terms of the average fraction of the applied that deposits over the same area [*e.g.*, 0.27 lb ai/a is equivalent to a fraction applied of 0.27 (or 27% of the agricultural application rate)].
b. Data are reported for the 90th percentile.

Finally, Tier I results are presented in **Figure 5** and **Table 10** for fine to medium droplet sizes for applications using orchard airblast application equipment. These results show that the highest exposure from spray drift is when chlorpyrifos applications are made to sparsely vegetated or dormant orchards. This is consistent with the fact that canopy density and spray interception likely correlates to potential reductions of spray drift.

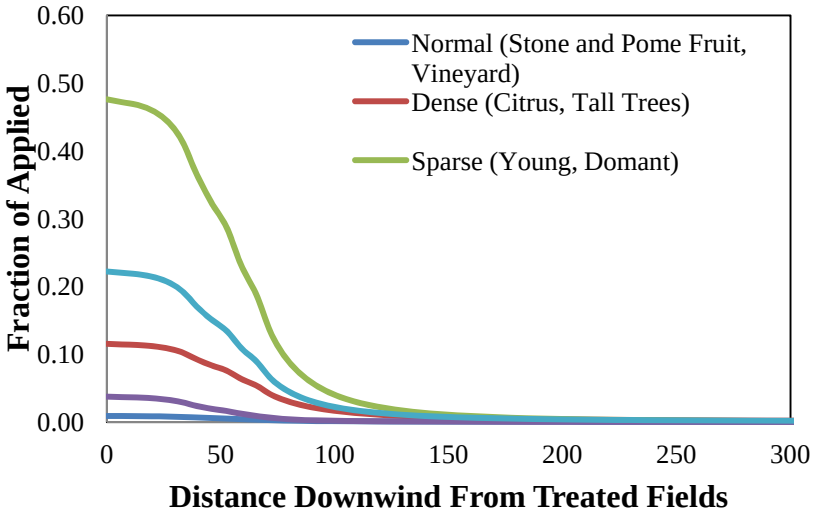


Figure 5. Tier I Spray Drift Analysis: Deposition Curves Resulting from the Application of Chlorpyrifos Using Orchard Blast Equipment in Various Orchard Types

Table 10. Tier I Spray Drift Analysis: Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn Starting at Various Distances From a Field Treated with Chlorpyrifos Using Orchard Blast Equipment in Various Orchard Types

Application Rate (lb ai/a)	Crop Canopy	Distance Downwind From Treated Field (feet)										
		0	10	25	50	75	100	125	150	200	250	300
		Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn										
6	Normal	0.03	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Dense	0.44	0.16	0.06	0.03	0.04	0.03	0.02	0.02	0.01	0.01	0.01
	Sparse	1.70	0.48	0.27	0.12	0.06	0.04	0.03	0.02	0.01	0.01	0.00
	Vineyard	0.12	0.02	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Orchard	0.80	0.24	0.15	0.07	0.05	0.03	0.02	0.02	0.01	0.01	0.01
4	Normal	0.02	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Dense	0.29	0.11	0.04	0.02	0.03	0.02	0.02	0.01	0.01	0.01	0.01
	Sparse	1.13	0.32	0.18	0.08	0.04	0.03	0.02	0.01	0.01	0.00	0.00
	Vineyard	0.08	0.02	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Orchard	0.53	0.16	0.10	0.05	0.03	0.02	0.02	0.01	0.01	0.01	0.01
2	Normal	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Dense	0.15	0.05	0.02	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.00
	Sparse	0.57	0.16	0.09	0.04	0.02	0.01	0.01	0.01	0.00	0.00	0.00
	Vineyard	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Orchard	0.27	0.08	0.05	0.02	0.02	0.01	0.01	0.01	0.00	0.00	0.00
1[a]	Normal	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Dense	0.07	0.03	0.01	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.00
	Sparse	0.28	0.08	0.05	0.02	0.01	0.01	0.00	0.00	0.00	0.00	0.00
	Vineyard	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Orchard	0.13	0.04	0.02	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00

Normal (Stone and Pome Fruit, Vineyard): This composite orchard combines grape and apple orchards
Dense (Citrus, Tall Trees): This composite orchard combines almond, orange, grapefruit, small grapefruit (mist blower) and pecan orchards.
Sparse (Young, Dormant): This composite orchard combines small grapefruit and dormant apple orchards.
Orchard: This composite orchard combines apple, almond, orange, grapefruit, small grapefruit, pecan, and dormant apple orchards.
Vineyard: This composite curve combines grape air blast sprayer applications and many not apply to other application equipment.
The double box indicates the current aquatic buffer distance of 50 feet.
a. The average deposition concentration for a 50' wide lawn at each of the distances downwind. This is equivalent in terms of the average fraction of the applied that deposits over the same area [*e.g.*, 0.27 lb ai/a is equivalent to a fraction applied of 0.27 (or 27% of the agricultural application rate)].

The spray drift deposition curves for the Tier II analysis for aerial chlorpyrifos applications are shown in **Figure 6** for a medium droplet size ($D_{v0.5}$ = 295 µM). Included in **Appendix E** are the results for fine to medium droplet size ($D_{v0.5}$ = 255 µM), medium to coarse ($D_{v0.5}$ = 341 µM) and coarse to very coarse ($D_{v0.5}$ = 439 µM) droplet sizes (**Figures E.1**, **E.2** and **E.3**, respectively). For all the droplet size spectra analyzed, the fraction of chlorpyrifos applied that is expected to

be deposited over a 50' wide lawn for various distances downwind from the treated field are provided in **Table 11**.[30]

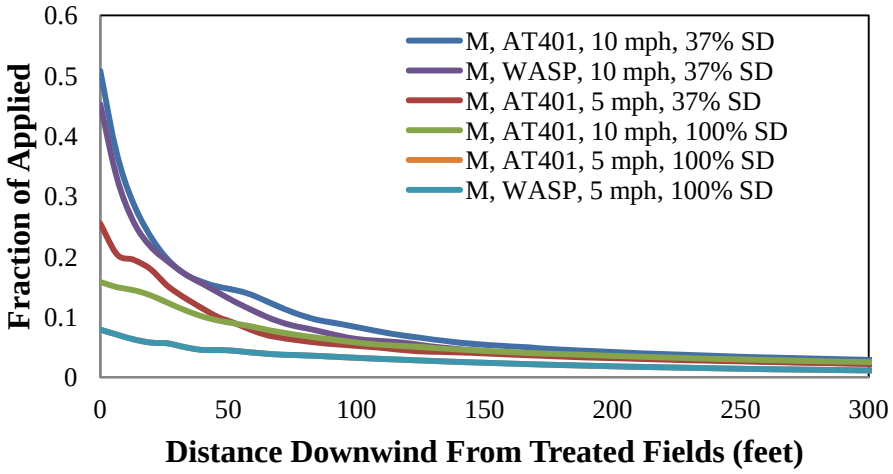


Figure 6. Tier II Spray Drift Analysis: Deposition Curves for a Medium Droplet Size Following Chlorpyrifos Application Using Several Different Aerial Application Configurations

Table 11. Tier II Spray Drift Analysis: Average Deposition Concentrations (lb ai/a) for a 50' Wide Lawn Starting at Various Distances From a Field Treated with Chlorpyrifos Using Several Different Aerial Application Configurations and a Medium Droplet Size

Configuration	Distance Downwind From Treated Field (feet)										
	0	10	25	50	75	100	125	150	200	250	300
	Average Fraction of Applied Deposited on a 50' Wide Lawn Downwind of a Treated Field[1]										
AT401, F2M, 10 mph, 37% SD	0.28	0.21	0.18	0.14	0.10	0.09	0.07	0.06	0.05	0.04	0.04
WASP, F2M, 10 mph, 37% SD	0.27	0.21	0.17	0.11	0.09	0.07	0.06	0.05	0.04	0.03	0.03
AT401, F2M, 5 mph, 37% SD	0.19	0.16	0.12	0.09	0.07	0.06	0.05	0.05	0.04	0.03	0.03
AT401, F2M, 10 mph, 100% SD	0.16	0.14	0.12	0.09	0.08	0.07	0.06	0.05	0.04	0.04	0.03
AT401, F2M, 5 mph, 100% SD	0.10	0.09	0.08	0.06	0.06	0.05	0.05	0.04	0.03	0.03	0.02
WASP, F2M, 5 mph, 100% SD	0.08	0.07	0.06	0.05	0.04	0.04	0.03	0.03	0.02	0.02	0.01
AT401, M, 10 mph, 37% SD	0.24	0.18	0.15	0.11	0.08	0.07	0.05	0.05	0.04	0.03	0.03
WASP, M, 10 mph, 37% SD	0.22	0.16	0.13	0.09	0.06	0.05	0.04	0.04	0.03	0.02	0.02
AT401, M, 5 mph, 37% SD	0.16	0.13	0.10	0.06	0.05	0.04	0.04	0.04	0.03	0.02	0.02
AT401, M, 10 mph, 100% SD	0.13	0.11	0.09	0.07	0.06	0.05	0.04	0.04	0.03	0.03	0.02
AT401, M, 5 mph, 100% SD	0.08	0.07	0.06	0.05	0.04	0.04	0.03	0.03	0.02	0.02	0.02
WASP, M, 5 mph, 100% SD	0.06	0.05	0.04	0.04	0.03	0.03	0.02	0.02	0.02	0.01	0.01
AT401, M2C, 10 mph, 28% SD	0.24	0.16	0.12	0.09	0.06	0.05	0.04	0.03	0.03	0.02	0.02

[30] To obtain the estimated chlorpyrifos concentrations (lb ai/a), as presented for Tier I analysis, the fraction of applied should be multiplied by the range of chlorpyrifos application rates (1 to 6 lb ai/a).

Configuration	Distance Downwind From Treated Field (feet)										
	0	10	25	50	75	100	125	150	200	250	300
	Average Fraction of Applied Deposited on a 50' Wide Lawn Downwind of a Treated Field[1]										
WASP, M2C,10 mph, 28% SD	0.21	0.14	0.11	0.07	0.05	0.04	0.03	0.03	0.02	0.02	0.01
AT401, M2C,5 mph, 28% SD	0.15	0.11	0.08	0.05	0.04	0.03	0.03	0.02	0.02	0.02	0.01
AT401, M2C,10 mph, 100% SD	0.10	0.08	0.07	0.05	0.04	0.03	0.03	0.03	0.02	0.02	0.01
AT401, M2C,5 mph, 100% SD	0.05	0.04	0.04	0.03	0.03	0.02	0.02	0.02	0.02	0.01	0.01
WASP, M2C, 5 mph, 100% SD	0.04	0.03	0.03	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01
AT401, C, 10 mph, 25% SD	0.22	0.14	0.10	0.07	0.05	0.04	0.03	0.03	0.02	0.02	0.01
WASP, C, 10 mph, 25% SD	0.19	0.12	0.09	0.05	0.04	0.03	0.02	0.02	0.01	0.01	0.01
AT401, C2VC, 10 mph, 21% SD	0.21	0.12	0.09	0.06	0.04	0.03	0.02	0.02	0.01	0.01	0.01
WASP, C2VC, 10 mph, 21% SD	0.17	0.10	0.07	0.04	0.03	0.02	0.02	0.02	0.01	0.01	0.01
AT401, C2VC, 5 mph, 21% SD	0.12	0.08	0.05	0.03	0.02	0.02	0.02	0.01	0.01	0.01	0.01
AT401, C2VC, 10 mph, 100% SD	0.06	0.05	0.04	0.03	0.02	0.02	0.02	0.02	0.01	0.01	0.01
AT401, C2VC, 5 mph, 100% SD	0.03	0.03	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.01
WASP, C2VC, 5 mph, 100% SD	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.00
AT401, VC, 10 mph, 20% SD	0.20	0.11	0.07	0.05	0.03	0.02	0.02	0.02	0.01	0.01	0.01
WASP, VC, 10 mph, 20% SD	0.16	0.09	0.06	0.04	0.03	0.02	0.02	0.01	0.01	0.01	0.01

1. To obtain the estimated chlorpyrifos concentrations (lb ai/a), as presented for Tier I analysis, the fraction of applied should be multiplied by the range of chlorpyrifos application rates (1 to 6 lb ai/a).
Fine to Medium (F2M): Average Droplet size ($D_{v0.5}$) = 255 µM
Medium (M): $D_{v0.5}$ = 294 µM
Medium to Coarse (M2C): $D_{v0.5}$ = 341 µM
Coarse (C) $D_{v0.5}$ = 385 µM
Coarse to Very Coarse (M2C): $D_{v0.5}$ = 439 µM
Very Coarse (VC): $D_{v0.5}$ = 478 µM
The double box indicates the current aquatic buffer distance of 150 feet.

The results suggest that swath displacement and wind speed have the most impact, of all the parameters tested, on reducing the amount of chlorpyrifos spray drift downwind of a treated field. A comparison of the different droplet sizes also shows that increasing the size of the spray droplet reduces the spray drift as well. This is also supported by the Tier I analysis.

5.2 Risk Estimates From Inhalation Exposure

The risk estimates associated with potential inhalation exposures are presented in this section (**Table 12**). *Note: Detailed calculations are included in* ***Appendix F***. The toxicological input for this calculation is a *Human Equivalent Concentration (HEC)* based on an acute rat inhalation study with multiple sampling times on day of dosing that was used to calculate a 2 hour HEC (2.48 mg/m^3). A no observed adverse effect level (NOAEL) was not determined in the rat inhalation study, as a result, the total uncertainty factor considered appropriate for identifying risks of concern is 300 (*i.e.*, MOEs < 300 present a risk estimate of concern).

In the 2011 risk assessment a 24 hour exposure period was used, which was a reasonable approach given that the source of exposure was ambient air and emissions occurred over a longer period of time than would be expected for typical spray drift exposure situations. The HEC used

was also based on a 24 hour period. Inhalation exposures from spray drift have instead been evaluated based using a 2 hour duration in order to better reflect how they would likely occur. This also required that an updated HEC based on a 2 hour exposure period be calculated as described above. The exposure considerations related to this approach are discussed below. This study was completed using small aerosols (~2 microns).

Air concentrations in the breathing zone were calculated using the Tier II option in AgDRIFT (V2.01). AgDRIFT provides air concentrations at specified distances downwind of a treated field in a vertical flux plane but this capability is limited only to aerial applications. These values are 1 hour time-weighted averages and have a certain degree of conservatism associated with them because in AgDRIFT the wind vector (*i.e.*, speed and direction) remains constant at all times and this analysis was based on a 10 mph windspeed, the highest allowable under current chlorpyrifos labels which results in the highest possible air concentrations. It also calculates air concentrations for the range of droplet sizes included in the particular spectra used as the basis for the model run. In all cases, droplets less than approximately 10 microns account for a very small percentage of the overall amount of droplets produced for any spectra that would be used for modeling of chlorpyrifos drift. The dichotomy between how the data used as the basis for the HEC and the airborne exposure concentrations should be considered when interpreting the associated risk estimates (*i.e.*, they are conservative because only a small percentage of the airborne exposure concentration is made up of droplets which could reach the lung).

For further consideration relative to the application methods evaluated, especially those that take more time to complete (*i.e.*, groundboom and airblast applications), wind vectors would be expected to change to some extent. In order to reflect actual anticipated exposures over 2 hour periods and account for expected changes in atmospheric conditions an approach to factor dilution over time was used based on the 1 hour concentrations as a starting point. The 1 hour values were amortized using a dilution factor of 25 percent per hour over the 2 hour exposure period. This value, especially when coupled with the premise inherent in AgDRIFT of a constant wind vector, is intended to represent possible conditions but also be conservative (*e.g.*, 20 flight passes for aerial applications which could take a substantial time period and a constant windspeed and vector). Values that represent a higher degree of dilution were considered but not utilized because some atmospheric conditions could potentially impede dilution such as atmospheric inversions or diametric changes in wind direction. However, to be thorough, the impact of conditions where more dilution would be expected was also investigated and these were found not to impact the results in a manner that would alter the conclusions of the assessment.

Finally, it is not appropriate to combine this exposure with those described below in **Section 5.3** because the exposure conditions differ (*e.g.*, duration, time/location issues), there are different uncertainties associated with each pathway, and the HEC in this case is based on lung cholinesterase inhibition using very small droplets (~ 2 microns) which differs from the other types of exposures.

Single day risk estimates based on the current label directives for managing spray drift are of concern for most scenarios at the 4 and 6 lb ai/a rates even at 125 feet from the edge of a treated field. Some scenarios are of concern at application rates of 2 and 2.3 lb ai/a at 75 feet from the

edge of a treated field when using medium to coarse nozzles. If larger droplets are used risks are not of concern at shorter distances. At the lower application rate of 1 lb ai/a risks estimates are not of concern at the field edge for larger droplets and not of concern at 10 feet for medium to coarse droplets. There are possible techniques that applicators can use to reduce drift from treated fields. These are discussed below in **Section 6** but due to a lack of information further refinements to these calculations have not been attempted.

Table 12. Inhalation Risk Estimates Associated With Spray Drift at Differing Distances From Edge of Treated Field

Flux Plane Distance (ft)	Results Based On 2 Hr Amortized Concentration			
	Medium/Coarse Droplets		Coarse/Very Coarse Droplets	
	ng/m3	MOE	ng/m3	MOE
6.0 lb ai/ A - Citrus [CA/AZ]				
0	46875	53	34375	72
10	39063	63	26563	93
25	31875	78	20313	122
50	26250	94	15625	159
75	20000	124	11250	220
100	16250	153	9375	265
125	13750	180	7813	317
4 lb ai/ A - Nursery and Orchard Trees (except apple), Tree Fruit, Tree Nuts				
0	32813	76	23438	106
10	27500	90	18438	135
25	23125	107	14375	173
50	18750	132	11250	220
75	14375	173	8750	283
100	12500	198	7500	331
125	10625	233	5938	418
2.3 lb ai/ A - psyllid, the vector for citrus greening[1]				
0	20625	120	14375	173
10	16875	147	11250	220
25	14375	173	8750	283
50	11875	209	7500	331
75	10000	248	5625	441
100	8125	305	5000	496
125	7500	331	4375	567
2 lb ai/ A - Nursery and Orchard Trees (except apple), Tree Fruit, Tree Nuts				
0	18125	137	12500	198
10	15000	165	10000	248

Flux Plane Distance (ft)	Results Based On 2 Hr Amortized Concentration			
	Medium/Coarse Droplets		Coarse/Very Coarse Droplets	
	ng/m3	MOE	ng/m3	MOE
25	13125	189	8125	305
50	11250	220	6875	361
75	8750	283	5000	496
100	8125	305	4375	567
125	6875	361	3750	661
1 lb ai/ A - Alfalfa, Asparagus, Sorghum, Strawberry, Sugarbeet				
0	10000	248	6875	361
10	8125	305	5000	496
25	7500	331	5000	496
50	6250	397	3750	661
75	5625	441	3125	794
100	5000	496	2500	992
125	4375	567	1875	1323
MOEs<300 would indicate a risk concern				
1. Pest present in Florida and possibly California and Texas				

5.3 Combined Risk Estimates From Lawn Deposition Adjacent to Applications

The risk estimates associated with deposition of residues that could potentially result in exposures akin to those experienced from intentionally treated turf are presented in this section (**Table 13**). These have been calculated using the Tier I option in the AgDRIFT model and the droplet spectra that are allowable per current chlorpyrifos labels. All of the results for orchard airblast and groundboom results are allowable under existing labels. *Note: Cells indicating risk estimates of concern have been highlighted for ease of review. Also, detailed calculations are included in **Appendix G**.*

The results are based on the premise that if a residence with a 50 foot wide lot exists next to a treated field spray drift could result from an application such that residues could be deposited across that lot on the lawn. Children could play on that contaminated turf resulting in exposure. This type of exposure has been assessed using the residential SOPs as described above. The only difference between these calculations that quantify exposure due to chlorpyrifos spray drift landing on residential lawns and an evaluation of a pesticide intended for use on lawns is how the exposure concentration is determined. The amount deposited on lawns in this approach is based on the agricultural application rate which was then adjusted for the average amount of spray drift deposition anticipated for the scenario across the modeled lawn (50' wide). In addition to results included in **Table 13** that are based on AgDrift Tier I, additional scenarios have been evaluated using AgDrift Tier II including considering medium, coarse, and very coarse sized droplets and the impact of using potential spray drift reduction measures (**Table 14**). In all cases, exposures were considered for lawns where the nearest side of the property was

directly adjoining the treated field (at field edge) up to 125 feet downwind of a treated field (see **Figure 2**). The toxicological points of departure for this analysis are route specific and highly refined. Dermal and non-dietary risk estimates were combined because the toxicity endpoint for each route of exposure is cholinesterase inhibition. The total applicable uncertainty factor is 100 so MOEs < 100 would be of concern.

The risk estimates presented in **Table 13** based on AgDrift Tier I indicate that the major risk concern is from aerial applications.[31] Risk estimates from ground boom and orchard airblast applications are not of concern past 25 feet downwind from a field edge for all situations considered, even at higher application rates, except for a couple of airblast application scenarios representing sparse orchard canopy situations. For aerial applications, risks are of concern up to 125 feet downwind from a treated field. Risks are reduced if lower application rates are considered. Also, the impact of changing nozzle types resulting in coarser sprays, which drift less, reduces risks from aerial applications.

Table 13. Risk Estimates Associated With Spray Drift Deposition Based On AgDrift Tier 1 From All Pertinent Application Methods

Application Scenario (Rep. Crop)	Appl. Rate (lb ai/A)	Nozzle/ Spray Type	Day (0) Highest Measured TTR (ug/cm2)			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	Day 0 Levels	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
Aerial												
Tier I [Citrus (CA/AZ)]	6.0	Fine to Medium	Pro - MS	3.83	0.146	10	13	15	20	27	34	42
			Mean-All sites	3.87	0.120	12	16	19	25	33	42	51
		Medium to Coarse	Pro - MS	3.83	0.146	11	17	22	32	46	60	75
			Mean-All sites	3.87	0.120	14	21	28	40	57	74	93
		Coarse to Very Coarse	Pro - MS	3.83	0.146	13	22	31	50	72	93	121
			Mean-All sites	3.87	0.120	16	27	39	61	88	114	148
Tier I [Tree Fruits, Nuts, Orchards, Nursery]	4.0	Fine to Medium	Pro - MS	3.83	0.146	14	19	23	31	41	51	63
			Mean-All sites	3.87	0.120	18	23	28	38	50	63	77
		Medium to Coarse	Pro - MS	3.83	0.146	17	26	34	48	70	90	113
			Mean-All sites	3.87	0.120	21	32	41	59	86	111	139
		Coarse to Very Coarse	Pro - MS	3.83	0.146	19	33	47	75	108	140	181
			Mean-All sites	3.87	0.120	23	40	58	92	133	172	222
Tier I [psyllid, the vector for citrus greening (CA/FL/TX)]	2.3	Fine to Medium	Pro - MS	3.83	0.146	25	33	40	53	71	89	109
			Mean-All sites	3.87	0.120	31	41	49	65	87	110	134
		Medium to Coarse	Pro - MS	3.83	0.146	30	45	59	84	121	156	196
			Mean-All sites	3.87	0.120	36	55	72	103	149	192	241

[31] Note that aggregate exposures that also consider other sources of possible chlorpyrifos exposure (*e.g.*, food and water) have not been incorporated into these risk estimates presented herein. These will be addressed in the registration review risk assessment for chlorpyrifos where all other pertinent aggregate exposure estimates will be presented and discussed.

Application Scenario (Rep. Crop)	Appl. Rate (lb ai/A)	Nozzle/ Spray Type	Day (0) Highest Measured TTR (ug/cm2)			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	Day 0 Levels	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
		Coarse to Very Coarse	Pro - MS	3.83	0.146	33	57	82	130	188	243	315
			Mean-All sites	3.87	0.120	41	70	101	159	230	298	387
Tier I [Tree Fruits, Nuts, Orchards, Nursery]	2	Fine to Medium	Pro - MS	3.83	0.146	29	38	46	61	82	103	125
			Mean-All sites	3.87	0.120	35	47	57	75	100	126	154
		Medium to Coarse	Pro - MS	3.83	0.146	34	52	67	97	139	180	226
			Mean-All sites	3.87	0.120	42	64	83	119	171	221	278
		Coarse to Very Coarse	Pro - MS	3.83	0.146	38	66	94	149	216	279	362
			Mean-All sites	3.87	0.120	47	81	116	183	265	343	445
Tier I Alfalfa, Asparagus. Sorghum, Strawberry, Sugarbeet	1	Fine to Medium	Pro - MS	3.83	0.146	57	76	92	122	163	205	250
			Mean-All sites	3.87	0.120	70	94	113	150	201	252	307
		Medium to Coarse	Pro - MS	3.83	0.146	68	104	135	193	279	360	452
			Mean-All sites	3.87	0.120	83	127	166	237	342	442	555
		Coarse to Very Coarse	Pro - MS	3.83	0.146	76	132	188	298	431	558	724
			Mean-All sites	3.87	0.120	94	162	232	367	530	686	890
Airblast												
Tier I [Citrus (CA/AZ)]	6	Normal	Pro - MS	3.83	0.146	485	1350	1907	3023	4168	5223	6445
			Mean-All sites	3.87	0.120	596	1659	2344	3715	5123	6419	7921
		Dense	Pro - MS	3.83	0.146	36	97	264	531	399	531	685
			Mean-All sites	3.87	0.120	44	119	324	653	491	653	841
		Sparse	Pro - MS	3.83	0.146	9	33	57	132	247	387	589
			Mean-All sites	3.87	0.120	11	40	70	163	303	476	724
		Vineyard	Pro - MS	3.83	0.146	135	672	1149	2218	3387	4500	5810
			Mean-All sites	3.87	0.120	166	825	1413	2725	4163	5531	7140
Tier I [Citrus]	3.5	Normal	Pro - MS	3.83	0.146	832	2314	3270	5182	7146	8954	11049
			Mean-All sites	3.87	0.120	1022	2843	4019	6369	8782	11004	13579
		Dense	Pro - MS	3.83	0.146	61	166	452	911	685	911	1174
			Mean-All sites	3.87	0.120	75	203	555	1119	842	1119	1442
		Sparse	Pro - MS	3.83	0.146	16	56	98	227	423	664	1009
			Mean-All sites	3.87	0.120	19	69	120	279	520	816	1240
		Vineyard	Pro - MS	3.83	0.146	232	1151	1970	3802	5807	7715	9960
			Mean-All sites	3.87	0.120	285	1415	2422	4672	7136	9482	12241
Tier I [Tree Fruits/Nuts/Orchards, Nursery]	2	Normal	Pro - MS	3.83	0.146	1456	4049	5722	9069	12505	15669	19336
			Mean-All sites	3.87	0.120	1789	4976	7033	11145	15368	19257	23763
		Dense	Pro - MS	3.83	0.146	107	290	791	1594	1198	1594	2054
			Mean-All sites	3.87	0.120	131	356	972	1958	1473	1958	2524
		Sparse	Pro - MS	3.83	0.146	28	98	172	397	740	1162	1766
			Mean-All sites	3.87	0.120	34	120	211	488	910	1428	2171

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Application Scenario (Rep. Crop)	Appl. Rate (lb ai/A)	Nozzle/ Spray Type	Day (0) Highest Measured TTR (ug/cm2)			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	Day 0 Levels	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
		Vineyard	Pro - MS	3.83	0.146	406	2015	3448	6653	10162	13501	17430
			Mean-All sites	3.87	0.120	499	2476	4238	8176	12488	16593	21421
Tier I [Christmas Trees, Conifer, Deciduous Nursery, Grape]	1	Normal	Pro - MS	3.83	0.146	2912	8098	11445	18137	25010	31339	38671
			Mean-All sites	3.87	0.120	3578	9952	14065	22290	30737	38515	47526
		Dense	Pro - MS	3.83	0.146	213	580	1581	3187	2397	3187	4108
			Mean-All sites	3.87	0.120	262	712	1943	3917	2946	3917	5048
		Sparse	Pro - MS	3.83	0.146	55	196	343	794	1481	2325	3533
			Mean-All sites	3.87	0.120	68	240	422	975	1820	2857	4342
		Vineyard	Pro - MS	3.83	0.146	811	4029	6896	13306	20323	27002	34860
			Mean-All sites	3.87	0.120	997	4952	8475	16352	24976	33185	42843
Groundboom												
Tier I [Grape (E of Cont. Divide)]	6	Low Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	11	235	327	467	587	687	794
			Mean-All sites	3.87	0.120	14	289	402	574	722	844	976
		High Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	10	143	201	292	373	441	515
			Mean-All sites	3.87	0.120	12	176	247	359	458	542	633
Tier I [Orchard Floor]	4	Low Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	17	353	490	701	881	1030	1191
			Mean-All sites	3.87	0.120	21	434	602	861	1083	1266	1464
		High Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	14	215	301	439	560	662	773
			Mean-All sites	3.87	0.120	18	264	370	539	688	813	950
Tier I [Fig, Peanut, Mint, Strawberry, etc.]	2	Low Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	34	706	980	1401	1762	2061	2382
			Mean-All sites	3.87	0.120	42	867	1205	1722	2166	2533	2927
		High Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	29	429	603	877	1119	1323	1546
			Mean-All sites	3.87	0.120	35	527	741	1078	1375	1626	1900
Tier I [Brussel Sprouts, Cauliflower, Onion, Soybean, Wheat]	1	Low Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	69	1412	1961	2802	3524	4121	4764
			Mean-All sites	3.87	0.120	84	1735	2410	3444	4331	5065	5854
		High Boom/Fine to Med/Coarse	Pro - MS	3.83	0.146	58	858	1206	1755	2238	2646	3093
			Mean-All sites	3.87	0.120	71	1055	1482	2156	2751	3252	3801
MOEs<100 indicate a risk estimate of concern												

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Additional results were calculated using AgDrift Tier II in order to evaluate risks associated with a variety of droplet spectra and to evaluate the impact of potential spray drift reduction measures (**Table 14**). This type of analysis is only feasible for aerial applications due to limitations with the AgDRIFT model. **Section 4.6** above describes the types of application parameters that were considered in detail. Some of the factors are believed to be readily implementable under actual field conditions as described in **Section 3.2** (*e.g.*, use of different droplet spectra). Others have been evaluated as well to identify how sensitive the results would be relative to changes in such parameters even though they may be less viable as mitigation measures because they are harder to routinely implement (*e.g.*, swath width and windspeed at time of application).

To summarize, the possible mitigation measures that were considered include:

- use of a helicopter (WASP) rather than fixed wing aircraft (AT 401),
- varied swath displacement (100% of the width of the aircraft shift inward from the field edge rather than the standard values for each droplet spectra – *e.g.*, 34% for medium) ,
- use of coarser droplets than the label indicates, specific size ranges are noted, and
- a reduction in maximum allowable wind speed during application from 10 to 5 mph.

These were developed in the context of the draft PR Notice guidance for spray drift labeling and the DRTs discussed in **Section 3.2** above.

It is clear that applicators can utilize measures that will be effective for reducing spray drift levels and that the methods will reduce risk estimates depending upon the situation. Altered nozzle size appears to be an effective approach for reducing risks compared to the current label that could be readily implementable.

Table 14. Risk Estimates Associated With Spray Drift Deposition Based On AgDrift Tier II From Aerial Application

Application Scenario	App Rate (lb ai/A)	Nozzle Configuration	Day (0) Highest Measured TTR			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	(ug/cm2)	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
Tier II (AT 401/10 mph/34%SD)	6.0	Medium	Pro - MS	3.87	0.146	10	14	18	24	32	40	48
Tier II (AT 401/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	16	20	26	40	51	59	67
Tier II (AT 401/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	21	24	28	36	45	53	60
Tier II (AT 401/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	33	40	46	56	64	71	80
Tier II (Wasp/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	11	15	19	29	40	49	60
Tier II (Wasp/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	24	30	35	46	56	65	75
Tier II (Wasp/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	18	26	35	52	65	75	100
Tier II (Wasp/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	45	52	59	70	82	95	110
Tier II (AT 401/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	12	19	25	38	53	68	84
Tier II (Wasp/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	14	22	30	49	69	88	110
Tier II (AT 401/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	13	24	35	57	81	106	136

Application Scenario	App Rate (lb ai/A)	Nozzle Configuration	Day (0) Highest Measured TTR			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	(ug/cm2)	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
Tier II (Wasp/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	17	31	45	73	104	136	175
Tier II (AT 401/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	11	17	22	31	44	55	68
Tier II (AT 401/5 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	18	25	34	56	74	86	101
Tier II (AT 401/10 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	27	32	39	52	65	76	89
Tier II (AT 401/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	48	59	68	82	97	110	126
Tier II (Wasp/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	13	19	25	40	57	71	87
Tier II (Wasp/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	65	77	87	109	135	160	189
Tier II (AT 401/10 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	13	22	31	48	68	86	109
Tier II (AT 401/5 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	22	34	49	84	114	136	167
Tier II (AT 401/10 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	45	53	63	84	107	127	151
Tier II (AT 401/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	78	93	109	134	164	192	222
Tier II (Wasp/10 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	15	27	38	63	88	113	145
Tier II (Wasp/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	114	135	156	199	248	295	350
Tier II (AT 401/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	15	21	26	36	48	60	73
Tier II (AT 401/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	24	30	39	60	77	89	100
Tier II (AT 401/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	31	36	42	55	68	79	90
Tier II (AT 401/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	50	60	69	84	97	107	120
Tier II (Wasp/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	17	23	29	44	60	74	91
Tier II (Wasp/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	36	44	53	68	84	98	112
Tier II (Wasp/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	27	40	53	78	97	112	150
Tier II (Wasp/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	68	79	88	105	123	142	165
Tier II (AT 401/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	18	28	38	56	80	101	126
Tier II (Wasp/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	21	33	45	73	103	132	165
Tier II (AT 401/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	20	36	53	86	122	159	204
Tier II (Wasp/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	25	46	67	110	156	205	263
Tier II (AT 401/10 mph/28%SD)	4.0	Medium to Coarse	Pro - MS	3.87	0.146	17	25	33	46	65	83	102
Tier II (AT 401/5 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	27	37	50	85	111	129	151
Tier II (AT 401/10 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	41	48	58	78	98	114	134
Tier II (AT 401/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	72	89	102	123	145	165	189
Tier II (Wasp/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	19	28	37	60	86	106	131
Tier II (Wasp/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	98	115	130	164	203	240	284
Tier II (AT 401/10 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	19	32	46	72	102	129	163
Tier II (AT 401/5 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	33	51	73	126	170	204	250
Tier II (AT 401/10 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	67	80	95	126	160	190	226
Tier II (AT 401/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	117	139	163	200	246	288	332
Tier II (Wasp/10 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	23	40	57	94	132	170	218
Tier II (Wasp/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	171	203	233	298	371	442	524
Tier II (AT 401/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	29	39	47	63	85	106	128
Tier II (AT 401/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	43	55	72	107	135	155	176
Tier II (AT 401/10 mph/100%SD)	2.3	Medium	Pro - MS	3.87	0.146	54	62	73	95	118	137	157
Tier II (AT 401/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	87	105	121	146	168	185	208
Tier II (Wasp/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	31	42	53	79	107	130	159

Application Scenario	App Rate (lb ai/A)	Nozzle Configuration	Day (0) Highest Measured TTR			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	(ug/cm2)	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
Tier II (Wasp/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	63	77	92	119	146	170	195
Tier II (Wasp/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	50	73	97	138	170	196	230
Tier II (Wasp/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	118	137	153	183	215	247	287
Tier II (AT 401/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	31	49	66	98	139	176	219
Tier II (Wasp/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	36	58	78	127	180	230	288
Tier II (AT 401/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	35	62	92	150	212	277	355
Tier II (Wasp/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	43	80	116	191	271	356	457
Tier II (AT 401/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	29	44	57	80	114	144	178
Tier II (AT 401/5 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	47	64	87	147	192	224	263
Tier II (AT 401/10 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	72	84	101	136	170	199	233
Tier II (AT 401/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	126	154	178	214	253	288	328
Tier II (Wasp/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	33	49	65	104	149	185	228
Tier II (Wasp/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	171	200	227	284	353	418	493
Tier II (AT 401/10 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	33	56	80	125	177	224	283
Tier II (AT 401/5 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	57	89	127	219	296	354	435
Tier II (AT 401/10 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	116	138	166	219	278	330	393
Tier II (AT 401/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	203	242	284	348	428	501	578
Tier II (Wasp/10 mph/21%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	39	70	98	163	230	295	378
Tier II (Wasp/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	298	353	406	519	646	769	912
Tier II (AT 401/10 mph/34%SD)	2	Medium	Pro - MS	3.87	0.146	33	45	55	73	98	122	147
Tier II (AT 401/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	50	63	82	123	155	179	202
Tier II (AT 401/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	62	71	84	109	136	158	181
Tier II (AT 401/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	100	121	139	168	193	213	239
Tier II (Wasp/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	36	48	60	91	123	150	183
Tier II (Wasp/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	72	89	106	137	168	196	225
Tier II (Wasp/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	58	84	111	159	195	226	264
Tier II (Wasp/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	135	157	176	211	247	284	330
Tier II (AT 401/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	36	57	76	113	160	203	251
Tier II (Wasp/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	41	67	90	146	207	264	331
Tier II (AT 401/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	40	71	106	172	243	318	409
Tier II (Wasp/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	50	93	134	220	312	409	525
Tier II (AT 401/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	33	50	65	92	131	166	204
Tier II (AT 401/5 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	54	74	101	169	221	258	302
Tier II (AT 401/10 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	82	97	116	156	196	229	267
Tier II (AT 401/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	144	177	205	246	291	331	378
Tier II (Wasp/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	38	57	74	119	172	213	262
Tier II (Wasp/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	196	230	261	327	406	481	567
Tier II (AT 401/10 mph/28%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	38	65	92	143	203	258	326
Tier II (AT 401/5 mph/28%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	65	102	146	252	341	407	500
Tier II (AT 401/10 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	134	159	190	252	320	380	452
Tier II (AT 401/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	233	279	326	401	492	576	665
Tier II (Wasp/10 mph/28%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	45	80	113	188	264	340	435

ATTACHMENT B

Application Scenario	App Rate (lb ai/A)	Nozzle Configuration	Day (0) Highest Measured TTR			MOEs At Various Distances Downwind From Treated Fields						
			Source	Appl. Rate	(ug/cm2)	At Edge	10 Feet	25 Feet	50 Feet	75 Feet	100 Feet	125 Feet
Tier II (Wasp/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	342	406	467	597	743	884	1049
Tier II (AT 401/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	66	89	109	145	196	243	295
Tier II (AT 401/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	100	126	165	246	311	358	404
Tier II (AT 401/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	124	143	167	218	272	316	362
Tier II (AT 401/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	201	242	278	335	387	427	478
Tier II (Wasp/10 mph/34%SD)		Medium	Pro - MS	3.87	0.146	71	97	121	182	246	300	367
Tier II (Wasp/10 mph/100%SD)		Medium	Pro - MS	3.87	0.146	145	177	211	274	335	392	450
Tier II (Wasp/5 mph/34%SD)		Medium	Pro - MS	3.87	0.146	115	168	223	318	390	452	529
Tier II (Wasp/5 mph/100%SD)		Medium	Pro - MS	3.87	0.146	271	314	353	422	494	567	659
Tier II (AT 401/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	71	114	153	225	320	406	503
Tier II (Wasp/10 mph/25%SD)		Coarse	Pro - MS	3.87	0.146	83	133	180	293	413	528	662
Tier II (AT 401/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	80	142	212	344	487	637	818
Tier II (Wasp/10 mph/20%SD)		Very Coarse	Pro - MS	3.87	0.146	99	185	267	440	623	818	1051
Tier II (AT 401/10 mph/28%SD)	1	Medium to Coarse	Pro - MS	3.87	0.146	66	101	131	185	262	332	408
Tier II (AT 401/5 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	107	148	201	338	442	515	605
Tier II (AT 401/10 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	165	194	232	312	392	457	535
Tier II (AT 401/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	289	354	409	493	581	661	755
Tier II (Wasp/10 mph/28%SD)		Medium to Coarse	Pro - MS	3.87	0.146	75	114	149	239	343	425	525
Tier II (Wasp/5 mph/100%SD)		Medium to Coarse	Pro - MS	3.87	0.146	393	459	522	654	812	961	1134
Tier II (AT 401/10 mph/28%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	75	129	184	287	406	516	652
Tier II (AT 401/5 mph/28%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	131	204	292	503	682	814	1000
Tier II (AT 401/10 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	267	318	381	505	640	759	905
Tier II (AT 401/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	466	558	653	801	984	1152	1329
Tier II (Wasp/10 mph/28%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	91	160	226	376	529	679	870
Tier II (Wasp/5 mph/100%SD)		Coarse to Very Coarse	Pro - MS	3.87	0.146	685	812	934	1194	1485	1769	2098
MOEs<100 indicate a risk estimate of concern												

6 Issues For Consideration

The available incident data and limited monitoring data included in the risk assessment indicated spray drift can be of concern. The analysis in this document supplements the findings in the 2011 HHRA by using modeling approaches which allow for a broader consideration of the potential impacts on risk estimates due to varied application scenarios. This analysis was developed by coupling established methods for estimating spray drift and residential exposures. As such, the caveats and concerns that can be associated with those techniques also apply herein.

Even given this premise, there are a significant number of issues that should be considered in the interpretation of the results including:

- The risk estimates calculated are based on the premise that spray drift occurs in a manner similar to that predicted and that an exposed person is at the locations and exhibits the behaviors mimicked in the risk assessment. These activities are believed to be plausible.

Examples could include homebound individuals at a location downwind and lawns adjacent to fields that could be contaminated followed by children playing on them.

- The analysis and resulting recommendations have been developed in a manner consistent with guidance in the draft PR Notice for spray drift labeling and the DRTs. The evaluation of possible risk mitigation approaches considered information developed in a manner consistent with this context. One example may be to develop droplet spectra data that evaluates the impact of adjuvants on the spray pattern (*e.g.,* sticking and drift reduction agents).

- Spray drift values are based on the general application of the Tier I output values from AgDRIFT and the subsequent higher tier analyses that were completed. There are uncertainties associated with all of these approaches which have been discussed extensively during the activities of the SDTF and the development of these models. All of the associated uncertainties apply to the results of this analysis (please refer to the FIFRA SAP reviews cited above). There are additional analyses which could be completed in order to elicit how sensitive spray drift levels may be to such changes (*e.g.*, evaluate impact of alternative aircraft, evaluate nozzle placement, impacts of surface roughness). It should also be noted that in the drift analyses completed in the assessment, wind vectors were moving from treated areas to the downwind area of the field 100 percent of the time and in most analyses (except some mitigation option analyses that considered lower values) windspeed was set to 10 mph which would maximize spray drift levels under the model conditions.

- The residential aspects of the calculation are based on the recently revised SOPs that have undergone an extensive review process as cited above. Associated with each calculation there is a degree of uncertainty which is described in detail in the SOPs themselves. A couple of issues should be noted in the interpretation of the calculated risks. In this case a TTR study for chlorpyrifos using multiple formulations in three different locations was available. The formulation that provided the highest relative TTR was used for estimating risks (Dursban Pro – which is a commonly used liquid formulation). TTR levels in the cited study varied by about a factor of two depending upon location and the type of formulation. Also, the TTR half-life in all cases was less than 1 day and in most cases less than 12 hours. Using this formulation, as compared with granules as an example, more closely mimics the exposures that would be expected from liquid spray drift.

- Aerial application is used on a small percentage of the crops for which chlorpyrifos is labeled, with the percent crop treated for most crops on the order of 1 to 2 percent. However, when acreage is treated using aircraft, it can account for a substantial number of the total applications for that crop on an annual basis (*e.g.*, alfalfa, almonds, apples, asparagus).

- Single day risk estimates were calculated by comparing calculated exposures to PODs defined based on animal studies that are of high quality and appropriate for that purpose. For many pesticides, defining PODs for single day non-dietary exposure scenarios is difficult, but for chlorpyrifos the data exists. For the inhalation exposure pathway, a state

of the art study that defined lung cholinesterase as the endpoint was used to establish the POD (*i.e.*, the HEC). For non-dietary ingestion a single day POD, the same used for defining the aPAD in the 2011 HHRA, was used. It was established based on the results of a single dose CCA (comparative cholinesterase assay) study that measured cholinesterase inhibition. Finally, 4 day sacrifice data in the same 21 day dermal toxicity study used to define the dermal POD in the 2011 HHRA also supports using the existing POD also as the basis of a the single day assessment approach. This characterization of the dermal study is also supported by a human volunteer dermal study (Nolan et al, 1992). Finally, the dichotomy between the nominal droplet size used to generate the HEC and the airborne exposure concentration should be considered in any evaluation of the related risk estimates (*i.e.*, they are conservative because only a small percentage of the airborne exposure concentration is made up of droplets which could reach the lung).

7 Conclusions and Recommendations

Because it can be demonstrated that spray drift occurs even if applications are compliant with labels, spray drift has been further examined to quantify potential risk estimates associated with exposure to chlorpyrifos downwind of a treated field. The approaches used as the basis for all calculations in this document have been thoroughly vetted through a public peer review process. In most cases, the findings indicate risks are a concern adjacent to treated fields and aerial applications, out of all application types considered, have the highest associated risk estimates. There are several actions which could be taken that would result in lower risk estimates. The impacts have been examined quantitatively where feasible. In some cases, differing combinations of possible mitigation options were also considered. These include:

- Reduce maximum application rates to those needed to achieve desired efficacy as appropriate.

- Require that buffer distances, similar to those already in place to address aquatic risk concerns, also be required to alleviate potential human health risks in situations where deemed appropriate. This would entail altering labels to expand the applicability and scope of the existing buffer distances for all areas where drift could result in human exposure.

- Implement drift reduction technologies and approaches consistent with DRTs that can alleviate risks including:

    - require larger droplets be used to alter spray patterns and reduce drift (*e.g.*, perhaps by requiring larger nozzle sizes by specifying a droplet size distribution (*i.e.*, $D_{v0.5}$) on the label))
    - require larger swath displacement from field edge

- Other possible approaches, consistent with the spirit of those used recently to manage potential risks for fumigants, may also be appropriate such as:

    - drift management plans for applicators

- specialized training for applicators on drift management issues
- use of buffer credit system consistent with the concept of DRTs, supported by appropriate data
- place restrictions around sensitive areas such as schools
- encourage use of technology to manage drift such as GPS for pilots

- Restrict aerial applications based on risk findings and available mitigation options.

Data to confirm the efficacy of any DRT measures implemented to reduce the risk estimates associated with spray drift from chlorpyrifos should be developed.

Appendix A- Summary of Label Use Patterns

Table A1: Summary of Chlorpyrifos Agricultural Uses		
Crop or Target	**Maximum (Single) Application Rate (lb ai/A)**	**Application Type/Equipment**
Alfalfa	1.0	Aerial, Chemigation, Groundboom, Tractor Drawn Spreader
Asparagus	1.0	Aerial, Groundboom
	1.5	Soil Ground Band (postharvest) (CA only) (Midwest and Pacific Northwest states – Amvac)
Beets (Table and Sugar Grown for Seed)	2	Soil Ground (preplant, at-plant) (24c)
Brassica Vegetable (Bok Choy, Broccoli, Broccoli Raab, Brussels Sprout, Cabbage, Cauliflower, Chinese Broccoli, Collards, Kale, and Kohlrabi)	1.0	Foliar Ground, Aerial
	2.25	Soil Ground Spreader or Splitter, T-Band (at-plant)
	2.4	Soil Band, Broadcast, Sidedress, & Injection (at plant, established plant, postplant)
Cauliflower	1.2 oz/1000 ft row	Soil Ground, Soil Injected (preplant, at-plant, postplant)
Broccoli, broccoli raab, etc.	1.3 - 1.4 oz/1000 ft row	Soil Ground, Soil Injected (preplant, at-plant, postplant)
Broccoli, Brussels sprout, cabbage	1.4 oz/1000 ft row	Ground (at-plant) (24c)
Cauliflower	1.3 oz/1000 ft of row	Ground (at-plant) (24c)
Bok choy, broccoli raab, Chinese broccoli	2.11	Soil Ground (at-plant) (24c)
Carrot (Grown for Seed)	1	Aerial, Groundboom (24c)
Citrus Fruit	6 (AZ and CA)	Aerial, Airblast, Groundboom, High Volume Spray (dilute), Low Volume (concentrate) Ground Sprayer, Broadcast,
	4 (States other than AZ and CA)	Aerial, Airblast, Groundboom, High volume Spray (dilute), Low Volume Ground (concentrate) Sprayer
	2.5 (24c)	Trunk Drench, Hand Held Sprayer
	2.0	Soil Broadcast, Chemigation Orchard Floors, Fertilizer Treatment; Mechanical Mixer
	1.0	Soil Ground Orchard Floors Soil Ground, Chemigation Orchard Floors, Fertilizer Treatment, Trunk Drench, Soil Broadcast, Directed Spray, Granule Applicator, Mechanical Mixer, Power

		Sprayer, Backpack Mist
Citrus orchard floors	1.0	Soil Ground orchard floors Soil Ground, chemigation orchard floors
Clover (Grown for Seed)	2.0 (24c)	Soil Ground (preplant), Broadcast Foliar Ground (postplant)
Corn (Field, Grown for Seed and Sweet)	3.0	Soil Incorporation (preplant)
	2.0	Soil In-Furrow, T-Banding, Granule Applicator (at plant; post plant)
	1.5	Aerial, Broadcast, Chemigation (postemergence)
	1.0	Foliar Aerial (postplant); Soil Band/In-Furrow Ground (preplant, at-plant, preemergence); Broadcast, Chemigation
Field, sweet	0.06 lb/lb seed (24c)	Commercial seed treatment
Field, sweet	0.5	Foliar Ground, Aerial, Chemigation (preplant, at plant, postemergence) (24c)
Cotton	2.0 (24c)	Aerial, Broadcast, Power Sprayer
Cotton	1.0	Aerial, Foliar, Broadcast, Chemigation, Groundboom
Cotton	012 lb/lb seed	Commercial Seed Treatment
Cranberry	1.5	Aerial, Chemigation, Groundboom, Broadcast Sprayer
Fig (CA only)	2.0	Soil Incorporation (dormant)
Ginseng	2.0	Soil Broadcast (preplant incorporation, postemergence)
Grapes	2.0	Soil Surface, Broadcast Ground Spray (dormant, delayed dormant) (24c, East of Continental Divide) postharvest (24c)
	1.0	Soil Drench (prebloom), Soil Broadcast, Ground, Hand-Held Sprayer (base, vine, Soil) (24c)
Legume Vegetables (Succulent or Dried, Except for Soybeans)	1.0	Soil Incorporation, T-Band, Ground (preplant, at-plant)
Mint (Peppermint and Spearmint)	2.0	Chemigation, Groundboom
Onion (Dry Bulb)	1.0	Groundboom, Tractor Drawn Spreader, Handgun
Peanut	2.0	Soil Ground (at-plant) Foliar Ground (postplant, band rescue) Soil Ground (preplant)
Peppers	1.0	Broadcast, Foliar, Groundboom (24c)
Pineapple (Non-bearing)	2.0	Broadcast, Foliar Ground (postplant) (24c)
Radish	3.0	Soil Ground Spreader or Splitter (at-plant in-furrow)
Radish (Grown for Seed)	1.0	Foliar Ground (24c)
Rutabaga	8.8	Soil T-Band (at plant)
	2.0	Soil T-Band, Incorporation (preplant; at-plant)

Sorghum	3.0	Soil Band, Incorporation (at plant)
	1.0	Aerial, Chemigation, Groundboom, Soil T-Band, Incorporation, Broadcast (at plant, postemergence, Foliar)
	0.0024 lb/lb seed	Commercial Seed Treatment
Soybean	2.0	Soil T-Band, Incorporate, Ground (at-plant, postplant)
	1.0	Aerial, Broadcast, Foliar Groundboom, Chemigation (preemergence, at-plant, postemergence)
Strawberry	2.0	Groundboom, Soil Incorporation, Broadcast (preplant spring, Foliar)
	1.0	Broadcast, Directed Spray (Foliar), Soil Incorporation (preplant); Foliar Ground (postharvest)
Sugarbeet	2.0	Soil Incorporation, Band, (preplant, at plant, postplant)
	1.0	Aerial, Chemigation, Groundboom, Broadcast, Soil Band, Incorporation, (at-plant, preplant, postemergence)
	0.5	Soil band, Incorporation (preplant, at plant)
Sugarbeet (Grown for Seed)	2.0	Soil incorporation, (preplant, at-plant) (24c)Aerial, Chemigation, Groundboom
Sunflower	2.0	Soil Ground (at-plant) Soil Ground (preplant)Aerial, Groundboom
	1.5	Foliar Ground, Aerial Broadcast (postemergence)
Sweet Potato	2.0	Soil preplant incorporation
Tobacco	2.0	Soil preplant incorporation
Almonds	4.0	Chemigation, Soil Broadcast, Foliar, Mound Treatment
	3.0 lb/100 gal	Trunk Drench, Foliar
	2.0	Foliar Ground, Aerial, Broadcast, Ground, High Volume Dilute and Low Volume Concentrate Sprays (dormant/delayed dormant)
Almond orchard floor	4.0	Soil Ground orchard floor, Soil Broadcast, chemigation, individual Mound Treatment
Filbert	2.0	Foliar Ground, Aerial, Broadcast, Ground, High Volume Dilute and Low Volume Concentrate Sprays (dormant/delayed dormant)
Pecan	4.0	Chemigation, Soil Broadcast, Foliar
	2.0	Foliar Ground, Aerial, Broadcast, Ground, High Volume Dilute and Low Volume Concentrate Sprays (dormant/delayed dormant)

	1.0	Foliar Ground, Aerial, Broadcast, Ground, High Volume Dilute and Low Volume Concentrate Spray (dormant/delayed dormant)
Pecan orchard floor	2.0	Soil Ground, chemigation orchard floors
Walnut	4.0	Chemigation, Soil Broadcast, Foliar
	2.0	Foliar Ground, Aerial, Broadcast, Ground, High Volume Dilute and Low Volume Concentrate Sprays (dormant/delayed dormant)
	1.0	Foliar Ground, Aerial, Broadcast, Ground, High Volume Dilute and Low Volume Concentrate Sprays
Apple	3.0	Ground High Volume Dilute Spray (dormant, delayed dormant)
	2.0	Ground High Volume Dilute & Low Volume Concentrate Spray (dormant/delayed dormant), Broadcast
	1.5 lb/100 gal	Trunk Spray Low Volume Handgun, Shielded Spray equipment
Cherry-sour	1.5	Foliar Ground, Aerial
Cherry	2.0	Foliar Ground, Aerial (dormant, delayed dormant)
	3 lb/100 gal	Ground trunk Spray (Foliar, postharvest)
Nectarine, Peach	2.0	Ground High Volume Dilute & Low Volume Concentrate Spray, (dormant/delayed dormant)
	3 lb/100 gal	Ground trunk Spray or preplant dip
Pear	3.0	Ground High Volume Dilute Spray (dormant, delayed dormant)
	2.0	Ground airblast speed Sprayer (postharvest); Ground High Volume Dilute & Low Volume Concentrate Spray (dormant/delayed dormant)
Plum/prune	3.0	Ground High Volume Dilute & Low Volume Concentrate Spray (dormant, delayed dormant)
	3 lb/100 gal	Trunk Drench, Foliar
Pumpkin	0.0625 lb/lb seed (24c)	Commercial Seed Treatment, preplant
Triticale	0.0024 lb/lb seed	Commercial Seed Treatment
Turnip	2.25-2.4	Soil Ground spreader or splitter ; Broadcast, Directed Spray, Soil band, Incorporation, (preplant, at-plant, postplant)
	1.4 oz/1000 ft row	Soil Ground (preplant, at-plant, postplant)
Wheat	4.0 (24c)	Soil Incorporation, Foliar
	1.0	Aerial, Broadcast, chemigation
	0.5	Aerial, Chemigation, Groundboom, Broadcast, Foliar

	0.0024	Commercial seed treatment

Table A2: Summary of Chlorpyrifos Non-Agricultural Uses

Crop or Target	Maximum (Single) Application Rate (lb ai/A)	Application Equipment
Ants (Fire Ant Mound, Carpenter)	0.080 (lb ai/gallon)	Backpack Sprayer, Handgun, Low Pressure Handwand
Cattle Ear Tags	0.0066 (lb ai/ ear tag)	Pliers (spring, summer)
Christmas Trees (Nurseries and Plantations, Stumps)	1.0	Foliar Ground nurseries & plantations Foliar Ground plantations; Broadcast, Aerial (Helicopter)
	3 lb/100 gal	Stump treatment, Drench
Golf Course Turf	4.0	Spot treatment
	1.0	Belly Grinder, Low Pressure Handwand, Push Type Spreader, Tractor Drawn Spreader, Turfgun; Soil Broadcast, Mound Treatment,
Grass Seed (Perennial Crops)	1.0	Foliar, Ground, Aerial
Greenhouse and Nursery Production (Bedding Plants, Containerized Ornamentals, Cut Flowers, Flowering Hanging Baskets, Pine Seedling Transplant, Potted Flowers, Ornamentals, Trees and Shrubs)	4.0	Soil Ground (preplant) field grown nursery stock
	3.0	High Volume Dilute Spray, Ground (dormant, delayed dormant)
	1.0	High Volume Dilute Spray, Ground (dormant, delayed dormant), Low Volume Concentrate Spray, trunk Drench, stump treatment, Foliar, preplant incorporation, Broadcast (Power Sprayer, Hand Held),
	1.0	Submerge Soil (containerized (potted) or balled & burlapped nursery stock)
Mosquitocide (Outdoor Residential, Recreational, or Other Non-Cropland Areas)	0.010	Wide Area Aerial and Ground
Non-crop Areas (Commercial Indoor/Outdoor Industrial Sites, Commercial Livestock Holding and Housing, Dumpsters/ Trash Areas, Food Processing Plants, Grown for Seed, Industrial Plant Site Perimeter Treatments, Manufacturing Sites, Power Utilities, Railroad Box Cars, Railroad Equipment, Road Medians, Ship Holds, Sod Farms, Telecommunications,	1.0	Aerial (ultra Low Volume), Belly Grinder, Groundboom, Push Type Spreader, Tractor Drawn Spreader, Soil Broadcast, band,treatment, Mound Treatment
	0.11 (lb ai/gallon)	Backpack Sprayer
	0.080 (lb ai/gallon)	Handgun, Low Pressure Handwand, Paint Brush/Roller
	0.044 (lb ai/1,000 sq ft)	Shaker Container
	0.018 (lb ai/1,000 sq ft)	Open Pour Bag

Warehouse Sites)		
Ornamentals (Cut Flowers, Industrial Buildings/Plant Sites Perimeter Treatments and Road Medians, Evergreens, Field Grown Nursery Stock, Flowers, Greenhouses, Non-bearing Fruit Trees Shrubs, Nurseries, Trees, Vines, Woody)	6.0	Belly Grinder, Push Type Spreader, Tractor Drawn Spreader, Broadcast, preharvest
	4.0	Groundboom, Preplant Incorporation
	3.0	High Volume Dilute Ground Sprayer (dormant, delayed dormant)
	2.0	Broadcast, Hand Held Sprayer, Power Sprayer
	0.16 (lb ai/gallon)	Backpack Sprayer, Handgun, High Pressure Handwand, Low Pressure Handwand
	0.020 (lb ai/gallon)	Drench/Dip
Poultry Litter	3.0	Bedding/Litter Treatment; Sprayer
Roach Control Bait Stations	0.00026 (lb ai/bait station)	Hand
Sewer Manhole Walls	0.30 lb/manhole	Airlee Spray Equipment Delivered thru a 6 ft. Spray Wand
Trees (Cottonwood and Poplar Trees Grown for Pulp, Conifer, Deciduous, Grown in Nurseries and Greenhouses)	2.0	Foliar Ground, Aerial, (dormant/delayed dormant) (24c)
Turfgrass (Sod or Seed)	4.0	Soil surface, Spot Treatment,
	1.0	Tractor Drawn Spreader, Soil Broadcast treatment, Sprayer
Wood Products (Fence Posts, Industrial Sites, Landscape Timbers, Logs, Manufacturing, Pallets, Processed Wood, Right of Way, Railroad Ties, Utility Poles, Wooden Containers)	16.65 lb/10,000 sq. ft.	Belly Grinder, Push Type Spreader, wood Surface Treatment, Brush, Sprayer

Appendix B - Lorsban 75WP Label Excerpt

Spray Drift Management

Do not allow spray to drift from the application site and contact people, structures people occupy at any time and the associated property, parks and recreation areas, non-target crops, aquatic and wetland sites, woodlands, pastures, rangelands, or animals.

Avoiding spray drift at the application site is the responsibility of the applicator. The interaction of many equipment and weather-related factors determine the potential for spray drift. The applicator is responsible for considering all of these factors when making the decision to apply this product.

Observe the following precautions when spraying Lorsban 75WG adjacent to permanent bodies of water such as rivers, natural ponds, lakes, streams, reservoirs, marshes, estuaries, and commercial fish ponds.

The following treatment setbacks or buffer zones must be utilized for applications around the above-listed aquatic areas with the following application equipment:

Application Method	Required Setback (Buffer Zone) (feet)
ground boom	25
chemigation	25
orchard airblast	50
aerial (fixed wing or helicopter)	150

Making applications when wind is blowing away from sensitive areas is the most effective way to reduce the potential for adverse effects.

The following spray drift best management practices are recommended to avoid off-target drift movement from applications.

Aerial Application

- The boom width must not exceed 75% of the wingspan or 90% of the rotor blade.
- Nozzles must always point backward, parallel with the air stream, and never be pointed downward more than 45 degrees.
- Nozzles must produce a medium or coarser droplet size (255 to 340 microns volume median diameter) per ASABE Standard 572 under application conditions. Airspeed, pressure, and nozzle angle can all effect droplet size. See manufacturer's catalog or USDA/NAAA Applicator's Guide for spray size quality ratings.
- Applications must not be made at a height greater than 10 feet above the top of the target plants unless a greater height is required for aircraft safety. Making applications at the lowest height that is safe reduces exposure of droplets to evaporation and wind.

• Use upwind swath displacement and apply only when wind speed is 3 to 10 mph as measured by an anemometer. Do not apply product when wind speed exceeds 10 mph.
• If application includes a no-spray zone, do not release spray at a height greater than 10 feet above the ground or crop canopy.

Where states have more stringent regulations, they must be observed.

The applicator should be familiar with and take into account the information covered in the Aerial Drift Reduction Advisory.

Aerial Drift Reduction Advisory
This section is advisory in nature and does not supercede the mandatory label requirements.

Information on Droplet Size: The most effective way to reduce drift potential is to apply large droplets. The best drift management strategy is to apply the largest droplets that provide sufficient coverage and control. Applying larger droplets reduces drift potential, but will not prevent adverse effects from drift if applications are made improperly, or under unfavorable environmental conditions (see Wind, Temperature and Humidity, and Temperature Inversions).

Controlling Droplet Size:

• Volume - Use high flow rate nozzles to apply the highest practical spray volume. Nozzles with higher rated flows produce larger droplets.
• Pressure - Do not exceed the nozzle manufacturer's recommended pressures. For many nozzle types, lower pressure produces larger droplets. When higher flow rates are needed, use higher flow rate nozzles instead of increasing pressure.
• Number of nozzles - Use the minimum number of nozzles that provide uniform coverage.
• Nozzle orientation - Orienting nozzles so that the spray is released parallel to the airstream produces larger droplets than other orientations and is the recommended practice. Significant deflection from horizontal will reduce droplet size and increase drift potential.
• Nozzle type - Use a nozzle type that is designed for the intended application. With most nozzle types, narrower spray angles produce larger droplets. Consider using low-drift nozzles. Solid stream nozzles oriented straight back produce the largest droplets and the lowest drift.

Boom Length: For some use patterns, reducing the effective boom length to less than 3/4 of the wingspan or rotor length may further reduce drift without reducing swath width.

Application Height: Applications should not be made at a height greater than 10 feet above the top of the target plants unless a greater height is required for aircraft safety. Making applications at the lowest height that is safe reduces exposure of droplets to evaporation and wind.

Swath Adjustment: When applications are made with a crosswind, the swath will be displaced downwind. Therefore, on the up and downwind edges of the field, the applicator should

compensate for this displacement by adjusting the path of the aircraft upwind. Swath adjustment distance should increase, with increasing drift potential (higher wind, smaller drops, etc.).

Wind: Drift potential is lowest between wind speeds of 2 to 10 mph. However, many factors, including droplet size and equipment type, determine drift potential at any given speed. Application should be avoided below 2 mph due to variable wind direction and 'high inversion potential. Note: Local terrain can influence wind patterns. Every applicator should be familiar with local wind patterns and how they affect spray drift.

Temperature and Humidity: When making applications in low relative humidity, set up equipment to produce larger droplets to compensate for evaporation. Droplet evaporation is most severe when conditions are both hot and dry.

Temperature Inversions: Applications should not occur during a temperature inversion because drift potential is high. Temperature inversions restrict vertical air mixing, which causes small suspended droplets to remain in a concentrated cloud. This cloud can move in unpredictable directions due to the light variable winds common during inversions. Temperature inversions are characterized by increasing temperatures with altitude and are common on nights with limited cloud cover and light to no wind. They begin to form as the sun sets and often continue into the morning. Their presence can be indicated by ground fog; however, if fog is not present, inversions can also be identified by the movement of smoke from a ground source or an aircraft smoke generator. Smoke that layers and moves laterally in a concentrated cloud (under low wind conditions) indicates an inversion, while smoke that moves upward and rapidly dissipates indicates good vertical air mixing.

Sensitive Areas: The pesticide should only be applied when the potential for drift to adjacent sensitive areas (*e.g.*, residential areas, bodies of water, known habitat for threatened or endangered species, nontarget crops) is minimal (*e.g.*, when wind is blowing away from the sensitive areas).

Ground Boom Application
The following mandatory spray drift best management practices are required to reduce the likelihood of off-target drift movement from ground applications.

- Choose only nozzles and pressures that produce a medium or coarse droplet size (255 to 400 microns volume median diameter), per ASABE Standard 572. See manufacturer's catalog or USDA/NAAA Applicator's Guide for spray size quality ratings.
- Apply with nozzle height no more than 4 feet above the ground or crop canopy.
- Do not apply product when wind speed exceeds 10 mph as measured by an anemometer.

Orchard Airblast Application
The following mandatory spray drift best management practices are required to reduce the likelihood of off-target drift movement from airblast applications.

- Nozzles must be directed so spray is not projected above the canopies.

• Apply only when wind speed is 3 to 10 mph at the application site as measured by an anemometer outside of the orchard/vineyard on the upwind side.
• Outward pointing nozzles must be shut off when turning corners at row ends.

The applicator should take into account the following best management practices to reduce off-site spray drift. This section is advisory and does not supersede mandatory label requirements.

• Number of nozzles, nozzle orientation and spray volume, air speed and wind direction are key factors in adjusting airblast spray delivery to match the height and density of the crop canopy. Airblast equipment should be adjusted to provide uniform coverage while minimizing the amount of spray movement over-the-top or completely through the crop canopy.
• High air volumes deliver spray more efficiently than air at high speed. Reducing forward travel speed decreases the air speed necessary to deliver the spray to the top of the crop canopy.
• Use air guides along with the number and orientation of spray nozzles to achieve the desired spray coverage and directional control.

• The following steps should be taken to minimize drift and the amount of non-target spray:
• Orient nozzles and adjust air speed/volume/direction to force the spray through the crop canopy but not allow drift past the canopy.
• Shut off spray delivery when passing gaps in crop canopy within rows.
• Spray the outside rows of orchards from outside in, directing the spray into the orchard and shutting off nozzles on the side of the sprayer away from the orchard.

• When treating smaller trees, vines or bushes, shut off top nozzles to minimize over-the-top spray movement.

Appendix C- Summary of Use Information

Chlorpyrifos usage data on all crops were compiled from a private marketing research database. For the years 2006-2010, the following information was queried at the crop level: pounds of active ingredient (a.i.) applied, total area treated (includes multiple applications to the same field), base area treated (the area treated at least once), crop area grown, percent of crop treated, average a.i. single application rate, the average number of applications per year (row crops only). Averages were calculated for each variable over the five years of data. To calculate the average number of applications per year for the specialty crops, the total area treated was divided by the base area treated. When possible, a comparison was made to the relevant data that were available from the USDA National Agricultural Statistics Service (NASS) pesticide use reports. For the following crops: beans, cucumbers, dry beans, hazelnuts, peas, peppers, plums/prunes, pumpkins, sorghum, and squash, the data are considered less robust because the values are based on a limited sample size (this is defined as a total sample size of <50 across 5 years), and the data could not be verified in NASS. While the low sample size may simply be a function of low use, sample size is also an indication of reliability and should be considered when using the data.

In addition to estimating the average application rate for chlorpyrifos, a rate distribution was generated to estimate an upper bound application rate for each crop. The data were generated using the *a.i. Rate Range* database variable (set from 0-10 pounds applied per application and at a rate interval of 0.25 pounds a.i. applied per application) along with the total acres treated database variable. These variables are part of the search functions available in the software provided along with the proprietary market database. To calculate the upper bound rate, the total area treated reported at each rate interval was divided by the cumulative total area treated across all intervals. A running total was included to show the cumulative percentile at each rate increment and the rate associated with the 90^{th} percent was reported as the upper bound rate. The upper bound rate in this analysis is the rate at which 90% (or as close to 90% as possible) of the acres treated with chlorpyrifos are treated at or below. The values obtained using this process, related to the overall use of chlorpyrifos, are provided in an accompanying spreadsheet to this document denoted as D399483-Appendix C.

The key information, excerpted from a separate memorandum, which details how ground applications play a role in chlorpyrifos use (Grube, 2012) is included below for informational purposes.

Table 1: Ground Applications of Chlorpyrifos to Agricultural Crops 2006-2010 excluding Seed Treatments and Incorporated Applications

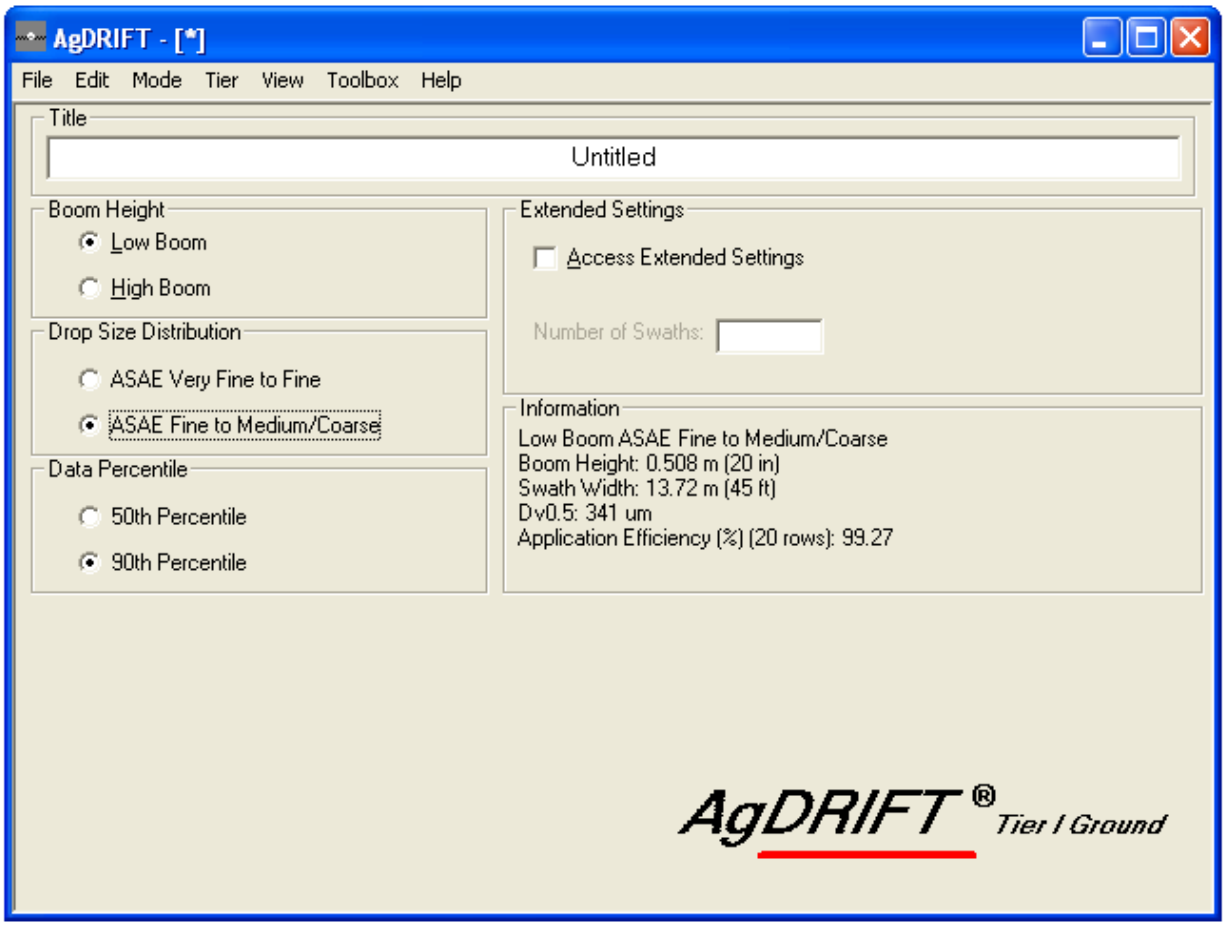
Crop	%of Total Applications Applied by Ground	%of Total Crop Acreage Treated by Ground	Average Lbs AI Year Applied by Ground	Average Application Rate for Ground Applications	Maximum Rate Observed for Ground Applications (rounded up) When 90th percentile is lower it is shown in parentheses
Alfalfa	69	2	320,000	0.6	1.0
Almonds	86	20	380,000	1.9	(2.0) 4.0
Apples	95	59	370,000	1.5	(2.0) 2.8
Asparagus	76	29	10,000		(1.0) 1.5
Beans (Snap)	99		2,000	0.5	1.0
Broccoli	100	53	100,000	1.4	(2.1) 2.3
Cabbage	100	14	10,000	1.1	(1.5) 2.3
Cauliflower	99	40	15,000	1.1	(1.5) 2.3
Cherries	98	36	80,000	1.5	(2.0) 3.0
Com	19	0	200,000	0.9	(1 .5) 3.0
Cotton	32		150,000	0.8	1.0
Dry BeanslPeas	37	0	5,000	0.5	(0.5) 0.8
Grapefruit	98	22	50,000	1.9	3
Grapes, Raisin	100	8	50,000	1.9	(2.0) 2.2
Grapes, Table	100	51	140,000	2.7	4
Grapes, Wine	100	9	120,000	2	(2.0) 4.0
Hazelnuts	68	8	4,000	1.4	2
Lemons	96	37	90,000	3.6	6
Onions	100	45	70,000	0.9	(1.1) 2.8
Oranges	93	22	600,000	2.6	6
Peaches	95	26	50,000	1.3	(2.0) 3.0
Peanuts	13		20,000	1.6	2
Pears	97	17	20,000	1.8	2
Peas (Fresh)	100		1,000	1	(1.0) 1.4
Pecans	85	24	200,000	0.9	(1.0) 3.4
Peppers	93	2	2,000	0.8	1.5
PlumslPrunes	100	11	25,000	1.8	2
Potatoes	82	0	1,000	0.8	I
Pumpkins	100	1	2,000	1	(1.1) 1.2
Sorghum	47	0	6,000	0.5	0.8
Soybeans	82	5	3,000,000	0.4	(0.8) 1.1
Squash	99	2	800		(1.5) 2.0
Strawberries	100	28	15,000		(1.0) 2.0
Sugar Beets	38	4	40,000	0.7	(1 .0) 2.0
Sunflowers	20		15,000	0.5	(0.5) 0.6
Sweet Com	57	7	70,000	1.1	(2.0) 4.0
Tobacco	83	11	80,000	2.1	(2.1) 5.0
Walnuts	94	43	320,000	1.8	(2.0) 4.0
Wheat, 'Spring	56		70,000	0.3	(0.5) 1.0
Wheat, Winter	69	2	290,000	0.4	(0.8) 1.0

Table 2. Incorporated Ground Applications of Chlorpyrifos 2006-2010

Crop	% of Total Applications Incorporated	% of Total Crop Acreage With Incorporated Treatments	Average Lbs AI Year Applied by Incorporated Applications	Averagc Application Rate for Incorporated Applications	Maximum Rate Observed for Incorporated Applications (rounded up)When 90th percentile is lower it is shown in parentheses
Com	65		800,000	1.0	(1.4) 2.0
Cotton	5	0	15,000	0.4	1.0
Dry Beans/Peas	45	0	5,000	0.5	0.5
Peanuts	85	6	140,000	1.7	2.0
Potatoes	10	0	<500	1.1	1.5
Sorghum	13	0	3,000	0.8	0.8
Soybeans		0	40,000	0.6	1.2
Sugar Beets	50	5	80,000	1.0	(1.0) 2.0
Tobacco	17	2	13,000	1.6	(2.1) 5.0

The applications reported in this table were those for which the application method clearly indicated incorporation. This level of detail
about application method was not available for fruits and vegetables so some of the ground applications for those crops may have incorporated as well as some of the ground applications for field crops not listed here. Source: Private market research data, 2006-2010.

Appendix D- Model Screenshots



Appendix E - Results of Drift Modeling

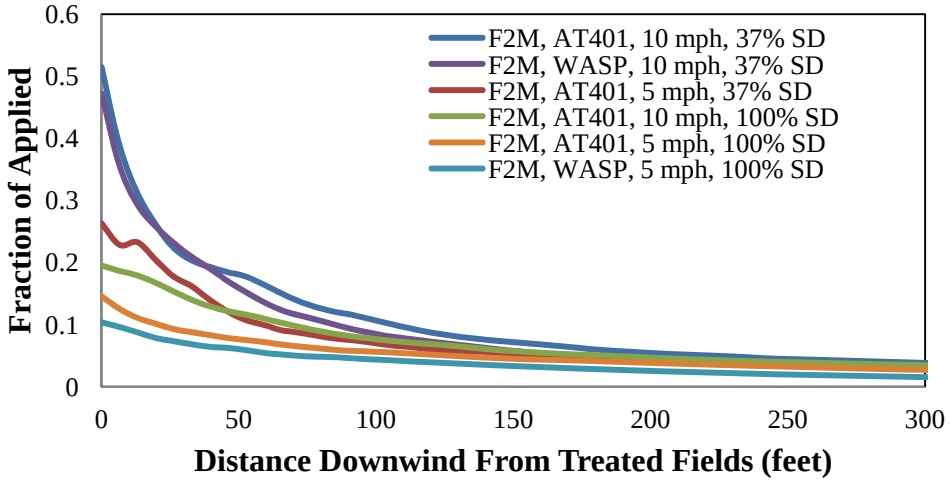


Figure E.1. Tier II Spray Drift Analysis: Deposition Curves for a Fine to Medium Droplet Size Following Chlorpyrifos Application Using Several Different Aerial Application Configurations

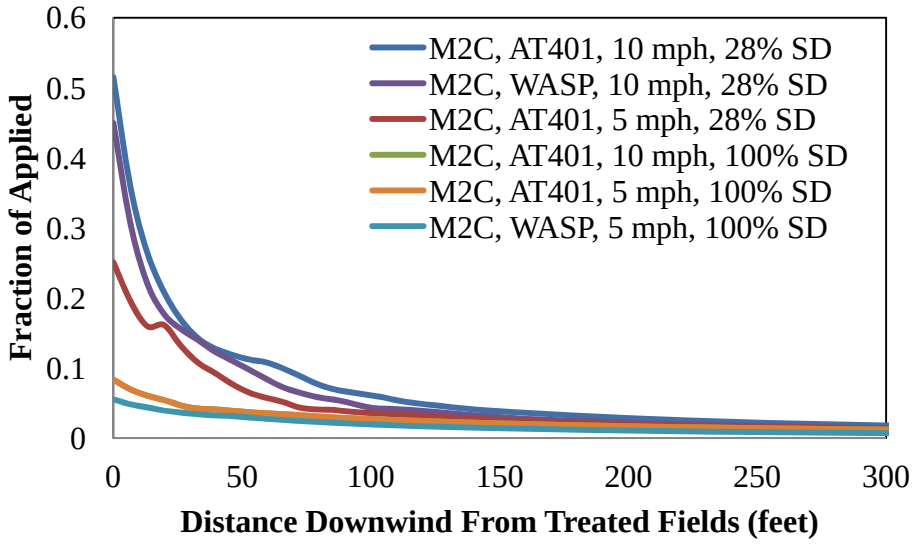


Figure E.2. Tier II Spray Drift Analysis: Deposition Curves for a Medium to Coarse Droplet Size Following Chlorpyrifos Application Using Several Different Aerial Application Configurations

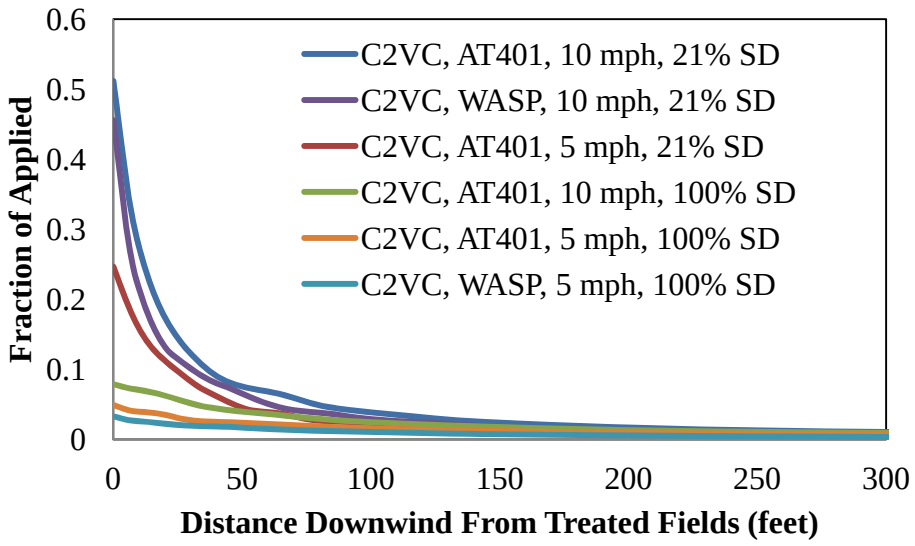


Figure E.3. Tier II Spray Drift Analysis: Deposition Curves for a Coarse to Very Coarse Droplet Size Following Chlorpyrifos Application Using Several Different Aerial Application Configurations

Detailed results and calculations are provided in an attached supplemental spreadsheet as Appendix E.

Appendix F- Inhalation Risk Calculations

This appendix is an accompanying spreadsheet to this document.

Appendix G- Risk Calculations From Deposited Residues on Lawns

This appendix is an accompanying spreadsheet to this document.

ATTACHMENT C

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON D.C., 20460

OFFICE OF
PREVENTION, PESTICIDES AND TOXIC
SUBSTANCES

December 17, 2008

MEMORANDUM

SUBJECT: Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held September 16-18, 2008 on the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos

TO: Debbie Edwards, Ph.D.
Director
Office of Pesticide Programs

FROM: Sharlene R. Matten, Ph.D.
Designated Federal Official
FIFRA Scientific Advisory Panel
Office of Science Coordination and Policy

THRU: Steven Knott, Executive Secretary
FIFRA Scientific Advisory Panel
Office of Science Coordination and Policy

Frank Sanders
Director
Office of Science Coordination and Policy

Attached, please find the meeting minutes of the FIFRA Scientific Advisory Panel open meeting held in Arlington, VA on September 16-18, 2008. This report addresses a set of scientific issues regarding the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos.

Attachments

cc:
James B. Gulliford
James J. Jones
Anne Lindsay
William Jordan
Margie Fehrenbach
Donald Brady
Janet Andersen
Steven Bradbury
William Diamond
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Heather A. Young Durick, Ph.D., MPH

SAP Minutes No. 2008-04

A Set of Scientific Issues Being Considered by the Environmental Protection Agency Regarding:

The Agency's Evaluation of the Toxicity Profile of Chlorpyrifos

September 16-18, 2008
FIFRA Scientific Advisory Panel Meeting
held at the
Holiday Inn - Rosslyn
Arlington, Virginia

NOTICE

These meeting minutes have been written as part of the activities of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), Scientific Advisory Panel (SAP). The meeting minutes represent the views and recommendations of the FIFRA SAP, not the United States Environmental Protection Agency (Agency). The content of the meeting minutes does not represent information approved or disseminated by the Agency. The meeting minutes have not been reviewed for approval by the Agency and, hence, the contents of these meeting minutes do not necessarily represent the views and policies of the Agency, nor of other agencies in the Executive Branch of the Federal government, nor does mention of trade names or commercial products constitute a recommendation for use.

The FIFRA SAP is a Federal advisory committee operating in accordance with the Federal Advisory Committee Act and established under the provisions of FIFRA as amended by the Food Quality Protection Act (FQPA) of 1996. The FIFRA SAP provides advice, information, and recommendations to the Agency Administrator on pesticides and pesticide-related issues regarding the impact of regulatory actions on health and the environment. The Panel serves as the primary scientific peer review mechanism of the Environmental Protection Agency, Office of Pesticide Programs (OPP), and is structured to provide balanced expert assessment of pesticide and pesticide-related matters facing the Agency. FQPA Science Review Board members serve the FIFRA SAP on an ad hoc basis to assist in reviews conducted by the FIFRA SAP. Further information about FIFRA SAP reports and activities can be obtained from its website at http://www.epa.gov/scipoly/sap/ or the OPP Docket at (703) 305-5805. Interested persons are invited to contact Sharlene R. Matten, Ph.D., SAP Designated Federal Official, via e-mail at matten.sharlene@epa.gov.

In preparing these meeting minutes, the Panel carefully considered all information provided and presented by EPA, as well as information presented in public comment. This document addresses the information provided and presented by EPA within the structure of the charge.

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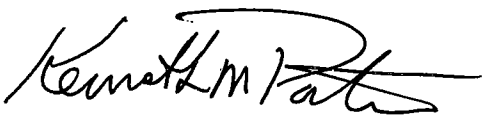
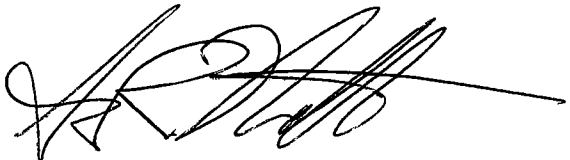
The Agency's Evaluation of the Toxicity Profile of Chlorpyrifos

September 16-18, 2008
FIFRA Scientific Advisory Panel Meeting
held at the
Holiday Inn - Rosslyn
Arlington, Virginia



Kenneth M. Portier, Ph.D.
FIFRA SAP Session Chair
FIFRA Scientific Advisory Panel

Date: 12/17/08



Sharlene R. Matten, Ph.D.
Designated Federal Official
FIFRA Scientific Advisory Panel Staff
Date: 12/17/08

Federal Insecticide, Fungicide, and Rodenticide Act
Scientific Advisory Panel Meeting
September 16-18, 2008

The Agency's Evaluation of the Toxicity Profile of Chlorpyrifos

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INTRODUCTION

The Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP) has completed its review of **The Agency's Evaluation of the Toxicity Profile of Chlorpyrifos.** Advance notice of the SAP meeting was published in the *Federal Register* on **July 9, 2008 and August 28, 2008**. The review was conducted in an open panel meeting September 16-18, 2008 held at the Holiday Inn-Rosslyn, Arlington, Virginia. Dr. Kenneth M. Portier chaired the meeting. Dr. Sharlene R. Matten served as the Designated Federal Official. Dr. Tina Levine, Director, Health Effects Division, Office of Pesticide Programs (OPP), provided opening remarks at the meeting. Presentations of technical background materials were provided by Dr. Anna Lowit and Ms. Deborah Smegal, MPH, Health Effects Division, OPP and by Dr. Ginger Moser, EPA-ORD-National Health and Environmental Effects Research Laboratory (NHEERL). Additional technical assistance was provided by Dr. John Liccione and Dr. John Doherty of the Health Effects Division, OPP.

Chlorpyrifos (O,O-diethyl-O-3,5,6-trichloro -2-pyridinyl phosphorothioate) is a broad-spectrum, chlorinated organophosphorus (OP) insecticide. Chlorpyrifos is one of the most widely used OPs in the U.S. In 2000, nearly all residential uses were voluntarily cancelled by Dow AgroSciences, LLC. However, chlorpyrifos continues to be used extensively in commercial agriculture. Since 2000, there has been extensive research on various aspects of chlorpyrifos toxicity, particularly on effects in animals and humans from gestational and postnatal exposure. Many new studies in rats investigating different endpoints including acetylcholinesterase (AChE) inhibition and adverse effects on the developing brain are now available. In addition, manuscripts have recently been published from three cohorts of pregnant women and children exposed *in utero* to organophosphates (OPs). At this time, the Agency is re-evaluating the extent to which toxicity endpoints and extrapolation/uncertainty factors for chlorpyrifos require updating based on this new information. The Agency's issue paper and associated appendices contain the proposed updates and the scientific foundation for the proposed revisions. The contents and conclusions drawn in the issue paper and appendices are preliminary. The ultimate goal of the Agency's on-going work is to improve the scientific support for the Agency's risk assessment. This will be accomplished by 1) evaluating new data on potentially susceptible subpopulations and 2) incorporating improved approaches, e.g., benchmark dose modeling instead of relying on no-observed-adverse-effect levels (NOAELs) for points of departure; and using extrapolation factors based on data instead of relying on default factors to account for differences in animals and humans and among humans. The Agency has progressed to a point in the review that feedback from the FIFRA SAP would be helpful.

PUBLIC COMMENTERS

Oral statements were presented by:

1) Daland Juberg, Ph.D., Dow AgroSciences, LLC
2) Charles Timchalk, Ph.D., DABT, Battelle Center for Biological Monitoring and Modeling
3) Carol Burns, Ph.D., Dow Chemical Company
4) Pamela Mink, Ph.D., MPH, Department of Epidemiology, Emory University
5) Michael Bartels, Ph.D., Dow Chemical Company
6) Douglas Weed, M.D., MPH, Ph.D., DLW Consulting Services
7) Robert Sielken, Ph.D., Sielken & Associates Consulting, Inc.
8) Michael Dourson, Ph.D., DABT, ATS, Toxicology Excellence for Risk Assessment (TERA)
9) Mr. Ray McAllister, Crop Life America
10) Elliot Gordon, Ph.D., Elliot Gordon Consulting, LLC
11) Jennifer Sass, Ph.D., Natural Resources Defense Council (NRDC) and on behalf of Pesticide Action Network North America (PANNA)
12) Michael Fry, Ph.D., American Bird Conservancy
13) Robin M. Whyatt, Ph.D., and Virginia A. Rauh, Ph.D., Columbia Center for Children's Environmental Health, Mailman School of Public Health, Columbia University

Written statements were provided by:

1) Torka S. Poet, Ph.D. and Charles Timchalk, Ph.D., DABT, Battelle Center for Biological Monitoring and Modeling, Battelle
2) Charles Timchalk, Ph.D., Battelle Center for Biological Monitoring and Modeling
3) David Eaton, Ph.D., DABT, FATS, Center for Ecogenetics and Environmental Health and Associate Vice Provost for Research, University of Washington on behalf of the authors of Eaton et al. 2008
4) Theodore Slotkin, Ph.D., Professor of Pharmacology and Cancer Biology and Psychiatry and Behavioral Sciences and Neurobiology and Director of Graduate Studies, Integrated Toxicology and Environmental Health Program, Duke University Medical Center
5) Scott Phillips, M.D., Department of Medicine, University of Colorado Health Sciences Center
6) Pamela Mink, Ph.D., MPH, Department of Epidemiology, Emory University
7) Douglas Weed, M.D., MPH, Ph.D., Founder and Managing Member, DLW Consulting Services, LLC
8) Kenneth D. Racke, Ph.D., Dow AgroSciences, LLC
9) Gary J. Mihlan, Ph.D., CIH and Walter Schmitt, Ph.D., Bayer CropScience
10) Michael Dourson, Ph.D., DABT, ATS, Bernard Gadagbui, Ph.D., DABT, and Lynne Haber, Ph.D., DABT, Toxicology Excellence for Risk Assessment (TERA)

SUMMARY OF PANEL DISCUSSION AND RECOMMENDATIONS

The Panel acknowledged the extent of the chlorpyrifos database and commended the Agency for preparing a comprehensive document considering the scientific evidence as a whole. Some of the more significant new data available to the Agency since the preparation of the 2000 chlorpyrifos risk assessment comes from three large prospective cohort studies of pregnant women and their children. Throughout the two-day discussion, Panel members referred to the results of these studies in an attempt to integrate their findings with the toxicology data from laboratory animal experiments. Despite the large volume of data available to address the risks of chlorpyrifos exposure from agricultural uses, several Panel members were concerned that a high degree of uncertainty is evident in the available data, particularly under low dose, chronic conditions. Uncertainty was expressed in attributing observed adverse effects to chlorpyrifos in the epidemiological studies where exposure was to metabolites of chlorpyrifos or to a mixture of chlorpyrifos and two additional anticholinesterase insecticides. The Panel agreed that the epidemiological studies have utility for risk assessment, but not as the principal basis for characterization of the point of departure (PoD). There was extensive discussion on experimental and epidemiological studies, e.g., of low-dose exposure in animals, of exposures in agricultural and pesticide handlers, and of the mode of action including the need to develop chlorpyrifos-specific PBPK models, that could provide the information needed to address critical data gaps and reduce uncertainty.

1. Metabolism & Toxicokinetics

The Panel concluded that a weight of evidence evaluation of available information supports the Agency's conclusions that 1) sensitivity to the adverse effects of chlorpyrifos is influenced by age, with young animals having lesser total ability to detoxify chlorpyrifos and many other organophosphorus compounds, and 2) that the age-dependent sensitivity observed in experimental animals is due mostly to toxicokinetic (TK) differences between juveniles and adults instead of only being the result of toxicodynamic (TD) differences.

The Panel agreed that current scientific data on chlorpyrifos uptake, transport, sequestration and excretion suggest that individual differences in metabolism and transformation will explain much of the variability seen in these factors, but that other potential TK differences between juveniles and adults should not be dismissed. For instance, the high respiratory rate of children may enhance the absorption of chlorpyrifos present in the air. Children have a greater cardiac output compared to adults, together with their less developed blood-brain barrier; facilitate some of the chlorpyrifos-oxon reaching the brains of exposed infants.

Some panel members questioned whether detoxication of the oxon plays a significant role in explaining the differential susceptibility between adults and juveniles at lower levels of chlorpyrifos exposure. Toxicokinetic differences are less likely to be relevant under low dose conditions compared to high dose conditions where enzyme systems may become saturated. Human tissue data specific to enzyme-mediated detoxication are minimal, however. While blood levels of detoxifying enzymes (e.g., PON1, carboxylesterases) have been studied, data on chlorpyrifos detoxication in specific organs (e.g., the liver) from humans are limited, making it

difficult to draw firm conclusions about the role of activation and detoxication in explaining differences in susceptibility to chlorpyrifos toxicity between human infants, children, and adults.

The Panel also agreed with the Agency's conclusion that "pregnant animals and humans may be somewhat more sensitive to chlorpyrifos than non-pregnant adults" based on the expected role of key detoxication enzymes (e.g., paraoxonase, P450 isozymes) in modulating levels of chlorpyrifos in animal studies. However, the Panel noted that there are relatively small differences in levels of these key enzymes between pregnant and non-pregnant animals. At dose levels expected from environmental exposures the importance of such small differences in enzyme levels is unknown, but the data suggest a reduced capacity to detoxify chlorpyrifos during pregnancy.

2. Cholinesterase Inhibition

Panel members agreed with the Agency's conclusion that postnatal rats are more sensitive than adult rats to increased cholinesterase inhibition and lethality following acute exposures. Increased susceptibility appears to be the result of the juvenile animals' lower capacity to detoxify either chlorpyrifos or the chlorpyrifos-oxon. The Panel was uncertain about how to interpret the data describing differences in cholinesterase inhibition between post-natal animals and adults following repeated chlorpyrifos dosing. The evidence suggests that younger animals are less sensitive than adults to cholinesterase inhibition from low dose repeated exposures, likely due to a lack of enzymes capable of converting chlorpyrifos to the oxon form in younger animals. In essence, the total acetylcholinesterase activity in the younger animal's tissues increases more effectively in between exposures, leading to lesser accumulation of inhibition over time than in adults. However, the role of the activation and detoxication pathways in producing differential cholinesterase inhibition and cholinergic effects at low dose repeated exposures is not clearly defined by the available data.

The Panel agreed with the Agency's preliminary conclusion that data showing less cholinesterase inhibition after repeated dosing during gestation may not reflect the true potential toxicity to the fetus. The data indicate, however, that the level of cholinesterase inhibition depends on the time of sampling following exposure. Developing rats would be expected to show less cholinesterase inhibition than would adults, with increasing length of time of measurements vs. dosing. Two mechanisms may explain this phenomenon: 1) a higher rate of recovery of the inhibited cholinesterase molecules and/or 2) an increase in cholinesterase enzyme synthesis, in the developing rat. There is little experimental evidence to indicate differences in enzyme reactivation following inhibition in cholinesterases from either young or adult tissues. The difference in rate of synthesis of new enzyme molecules is likely the dominant factor. A more accurate comparison of cholinesterase inhibition between the developing animal and the adult could be determined if sample collection coincided with the time of the peak cholinergic effect.

3. Laboratory Studies on the Developing Brain

In general, the Panel agreed with the Agency's conclusion that gestational or early postnatal exposures can lead to neurochemical and behavioral alterations that persist into adulthood. A number of rodent studies suggest that prenatal or postnatal exposures may lead to long-term

neurobehavioral changes in motor and cognitive behaviors. The Panel questioned the meaning of the phrases "at sufficiently high exposures" and "persisting into adulthood after any initial AChE inhibition has reversed." Some panelists stated that a "sufficiently high exposure" should be defined on the basis of a dose that induces brain cholinesterase inhibition. They noted that the majority of experimental animal studies reviewed by the Agency used doses of about 1 mg/kg. This dose (exposure level) should be sufficient to inhibit brain cholinesterase activity and at the same time causes neurobehavioral effects in most age groups.

Some members questioned the experimental methods used in some of the animal studies as well as the interpretation and application of the results of neurobehavioral testing in animals for risk assessment. Several Panel members felt that while behavioral findings are consistently reported in the literature, the types of behavioral activities reported as significant were not always the same. It was acknowledged that the study outcomes could be affected by 1) the route of administration of chlorpyrifos, 2) the developmental period of exposure, 3) the methods used to measure changes in behavioral domains, and 4) the choice of dependent variables. Panel members agreed with the Agency's expressed caution on the use of dimethyl sulfoxide (DMSO) as a vehicle because of its intrinsic toxicity and potential influence on absorption. In addition, uncertainly was expressed about potential interactions between DMSO and low doses of chlorpyrifos and the effect of this interaction on the developing animal.

The Panel agreed with the Agency's determination that insufficient data exist to clearly identify a specific mode of action for effects in the developing nervous system for use in risk assessment. Some panel members believed that studies cited by Eaton et al. (2008) were indicative of possible non-cholinergic modes of action. Other panel members disagreed with this conclusion. They stated that the effects on blood cholinesterase are considered protective for the observed effects (i.e., occurring at lower doses); however, nothing is implied about mode of action, except in the context of species and interindividual variability in toxicokinetics. Most of the Panel stated that the available information does not allow behavior to be considered as a point of departure. These panelists stated that cholinesterase inhibition should continue to be used for PoD until, at such time; an alternative mode of action is identified and validated. Collaboration between EPA and developmental neurotoxicologists would help identify enzyme inhibition occurring in discrete brain sites at critical periods of development.

4. Epidemiology Studies in Children and Mothers

The Panel agreed with the Agency's conclusions regarding the potential utility of the epidemiological data from the three cohort studies (Columbia University in New York, Mt. Sinai Hospital in New York, and the University of California at Berkeley) in broadly characterizing risk of chlorpyrifos exposure to pregnant women, fetuses, infants, and children. They also concurred that these studies should not be considered as the principal basis for characterization of the PoD. The Columbia study was considered the most epidemiologically-sound and to have adequately addressed selection and information biases to the best extent possible. It was also considered the most robust and appropriate in assessing chlorpyrifos developmental toxicity because specific measurements of exposure to chlorpyrifos in neonates and children (i.e., the study populations) were obtained. Confounding factors in the Mt. Sinai and Berkeley studies, particularly the fact that exposures were based on OP and carbamate metabolites and that

chlorpyrifos was not specifically measured, reduce their utility in a quantitative context for risk assessment. Several panel members pointed out that although the Agency classified the study population's exposure as "high" for a residential setting, it would be considered a "low" exposure for a population of agricultural workers or pesticide handlers.

The weaknesses inherent to observational studies were discussed. Given the paucity of human epidemiological studies of OP insecticides in children, the Mt. Sinai and Berkeley studies do provide useful information for risk assessment. For example, these studies provide data demonstrating abnormal reflexes in neonates that is not available from the Columbia cohort study. In addition, the Mt. Sinai cohort study considered paraoxonase activity as a factor in the analyses that may have relevance for human toxicity. There is uncertainty in the exposure estimates in the Columbia study because measurements were derived from a single time point (maternal and/or cord blood). Notwithstanding this uncertainty, the Panel felt that the results from the Columbia study raise concerns for adverse neurodevelopmental effects in children exposed *in utero* to environmental levels of chlorpyrifos although there was confounding by the presence of other anticholinesterase insecticides. The majority of issues raised by the Panel were points of clarification or issues that if addressed could strengthen the analysis and provide the Agency with better and more useful data for risk assessment.

Overall, the Agency's conclusion that chlorpyrifos could have contributed to the birth and neurodevelopmental outcomes noted in the three cohort studies was supported. The Panel agreed with the Agency that: a) exposures to multiple cholinesterase-inhibiting insecticides in combination cannot be ruled out as contributing to the birth and neurodevelopmental outcomes; b) a potential synergistic and/or additive effect of these compounds does not rule out a role for chlorpyrifos in contributing to the adverse outcomes; and c) it cannot be determined if chlorpyrifos is the sole contributor to the observed outcomes. These conclusions are supported by the Columbia study finding of a crude dose-response relationship to chlorpyrifos levels which persisted even after controlling for exposure to other pesticides.

When the results of the three cohort studies (with an emphasis on the Columbia study) are considered along with the findings from experimental studies in animals, the Panel concluded that maternal chlorpyrifos exposure would likely be associated with adverse neurodevelopmental outcomes in humans. Some panelists indicated that if the associations between chlorpyrifos exposure and the reported outcomes are causally related, then it is possible to conclude that the mechanism of action might be independent of cholinesterase inhibition. Exposures to multiple cholinesterase-inhibiting pesticides or other neurotoxicants may result in additive or interactive effects. This information might be useful when considering cumulative exposures to these pesticides.

5. Human Information Available for Risk Assessment

The Panel generally agreed with the Agency's conclusion that due to their limitations, the epidemiological data currently available are useful primarily for hazard identification. The Panel disagreed on whether the current epidemiological data provide sufficient evidence to suggest that the uncertainty factor for the cholinesterase inhibition endpoint be changed to accommodate the possibility of neurodevelopmental effects from low-level *in utero* exposures of chlorpyrifos. The

majority of the Panel agreed that the current epidemiological data do not provide sufficient evidence to increase the uncertainty factor for the cholinesterase inhibition to accommodate the possibility of neurodevelopmental effects. However, some panel members felt strongly that the current epidemiological data do provide evidence to indicate that the margin of safety should be increased. The Panel recommended that the Agency conduct a full formal weight of evidence evaluation for causality of the reported associations between exposure to chlorpyrifos and neurodevelopmental outcomes in the existing epidemiological database.

The Panel recommended that the Agency continue its collaboration with Columbia University researchers in analyzing the epidemiological data. The Agency is encouraged to continue open discussions with the study researchers (and the Centers for Disease Control and Prevention, as appropriate) to seek clarification on the level of confidence in the reported exposure levels. The Agency should then attempt to use the cohort data quantitatively to inform the risk assessment process, such as in a boundary setting exercise.

The use of physiologically-based pharmacokinetic (PBPK) modeling would enable estimation of exposure dose metric for multiple sources of exposure, e.g., air, food, water. The Panel also agreed with the concept of using a PBPK model to examine individual intra-species variations as well as inter-species differences. While the potential contribution of the chlorpyrifos PBPK model being developed was recognized, some panelists believed that the Agency should also pursue a simpler PBPK model specifically applicable to the chlorpyrifos data that would be available in a relatively short timeframe. The Panel concluded that the epidemiological data may be used for bounding exposure levels, and in conjunction with PBPK models, address current or potential human exposures and to determine the final reference dose or reference concentrations.

The Panel agreed with the Agency's approach to deriving a dermal absorption factor using data from the deliberate dosing in human studies. This approach is especially valuable if no other sources of data are available. However, the current 3% absorption factor should be further refined by considering different exposure scenarios and adjusting for expected underestimation of exposure. The Panel also agreed with the Agency's scientific analysis that these deliberate dosing studies cannot be used to directly establish a PoD or UFs, but indicated that these data might be used as bounding levels similar to what was suggested by the Panel concerning the data from the epidemiological studies.

6. Points of Departure (PoD) for Risk Assessment

The Panel was presented with three options for deriving points of departure for acute and chronic chlorpyrifos exposure from animal studies for extrapolating human risk. Panel members expressed difficulty in separating the discussion of points of departure from that of uncertainty factors with the result that the discussion of these two issues overlapped in many areas.

The Panel agreed that Option 3 was the most favorable option for deriving points of departure for acute and chronic exposure. There was also general agreement among Panel members with the Agency's proposal to use the dermal and inhalation studies from their 2000 assessment as a basis to develop points of departure for these exposure routes, taking into account sensitive life stages and adjusting for dosing regimen. Option 3 was chosen principally on the basis that the proposed point of departure for acute exposure takes into account all life stages, is based on

benchmark doses (an advantage over the NOAEL or LOAEL), and represents the results of several studies.

Based on available data, most of the Panel members (a few members disagreed) stated that the PoD presented in Option 3 is also believed to be protective for effects on the developing brain, although it is based on cholinergic effects. However, this conclusion has associated uncertainties; the lack of information on the mode of action in inducing the observed behavioral effects and evidence from *in vivo* and *in vitro* studies that show non-cholinergic modes of action are likely to be involved in the adverse developmental neurotoxicity and behavior endpoints. The Panel encourages EPA to explicitly address these uncertainties when deriving the reference dose/concentrations.

The effects on neurobehavioral development observed in the three epidemiological studies, the uncertainties surrounding the mode(s) of action following longer term exposure, and the need to account for potential cumulative effects support the lower point of departure for repeated exposures used by the Agency in its 2000 risk assessment. The Panel also recommended that the Agency investigate the possibility of conducting a benchmark dose assessment for the chronic point of departure determination.

The Panel recommended that the Agency consider development of an appropriate study using doses spanning and below those expected to produce cholinergic effects with the most sensitive administration method and the most sensitive life stage. Such a study would examine developmental neurotoxicity and behavior outcomes along with cholinesterase inhibition. This would likely help elucidate non-cholinergic modes of action for developmental neurotoxicity and behavior effects.

7. Extrapolation/Uncertainty Factors

After review of available data, the Panel concurred with the Agency that paraoxonase-1 (PON1) status cannot be ruled out as a determinant of chlorpyrifos toxicity, particularly for the fetus and the young child. Actual human exposures vary between bystanders and applicators (and everyone in between) and it would be difficult to define what constitutes a low level of exposure in humans. The Panel disagreed on whether the PON1 data alone could be used to address uncertainty. The majority of panel indicated that these data should not be used out of context until rate limiting step(s) is/are identified. The use of the PON1 data without such information was considered by some to be a misinterpretation or misuse of the IPCS guidance for determining chemical-specific adjustment factors (CSAFs).

Some of the Panel indicated that chlorpyrifos may produce effects in the fetus that would be manifest later at the juvenile stage or later on in life (e.g. adulthood). They stated that adverse neurodevelopmental effects in the fetus, neonates, and young children should be considered the most important endpoints for assessing chlorpyrifos toxicity. The Panel noted that there does seem to be a different susceptibility between fetuses and neonates compared to adults. Because specific modes of action have yet to be identified for these effects, available data are inadequate to inform inter-species and intra-species differences in TD and TK. Therefore, the application of default uncertainty factors for TD and TK considerations was recommended by the Panel.

The Panel noted the uncertainties surrounding the role of PON1 and its genetic polymorphisms in humans exposed to chlorpyrifos and other cholinesterase inhibitors. Some panel members emphasized the lack of data in animals and humans on the involvement of PON1 in detoxication pathways, especially in situations of low dose exposures. Most of the Panel encouraged the Agency to use PBPK modeling to assess the overall impact of PON1 on toxicity over a range of chlorpyrifos exposures. While other panel members recommended that the Agency gather data (both *in vivo* and *in vitro* data) on animal and human enzyme kinetics and analyze carefully the effects as related to the PON-192 Q/R polymorphism. The information on the overall contribution of PON1 Q192R polymorphism on the deactivating pathways and potentially rate-limitanting components will be very useful when examined in light of future PBPK models.

The Agency proposed two options for deriving an intra-species TK (human variation) uncertainty factor: 1) a 12-fold data-derived factor based on the PON1 data or 2) the default three-fold factor. The majority of the Panel supported the use of the default three-fold factor. These panel members disagreed with the Agency's approach to use PON1 genetic polymorphisms to derive intra-species uncertainty factor. They stressed that PON1 is only one downstream enzyme in a complex metabolic pathway and that the PON1 genotype alone is insufficient to predict human variability. One panel member added that if chlorpyrifos acts directly on certain brain targets to elicit developmental neurotoxicity without the need of activation to the chlorpyrifos oxon (the substrate for PON1), then the role of PON1 for neurodevelopmental toxicity would be irrelevant. On the other hand, several panelists stated that the available PON1 data support an uncertainty factor of 12 for the intra-species factor to account for potential developmental neurotoxicity of chlorpyrifos as shown in the epidemiological studies. These members preferred the UF_{HK} of 12-fold, rather than the default of 3-fold, because these were the only two choices proposed. However, none of the panel members endorsed the CSAF approach used by the Agency to identify the factor of "12-fold" calculated based on chlorpyrifos-oxonase and encouraged the Agency to pursue other approaches based on the mode of action (i.e., 2005 IPCS CSAF guidelines).

The Panel did not reach consensus on one specific uncertainty factor for inter-species TK differences. Most of the panel concurred with the Agency's proposal to stay with the default three-fold factor for inter-species TK differences and add no additional uncertainty factor for developmental and behavior neurotoxicity. These panel members noted that the most sensitive effect appears to be AChE inhibition with a $BMDL_{10}$ (i.e., lower confidence limit on the benchmark dose calculated to a 10 percent effect level) for the red blood cell (RBC) AChE inhibition after repeated exposure at 0.03 mg/kg/day; whereas, the lowest dose tested in the developmental neurotoxicity studies in dams (0.3 mg/kg/day) was 10-fold greater and failed to produce observable behavioral effects in the offspring. On the other hand, a few panel members disagreed with this assessment and recommended that the Agency should apply a default uncertainty factor of 100 to the points of departure based on the cholinesterase inhibition endpoint and further consider the use of an additional uncertainty factor to address the concerns for developmental and behavioral neurotoxicity as observed in both the animal and epidemiological studies.

The Panel generally favored the use of a PBPK model to integrate key TK and TD factors and evaluate their contributions to the endpoints of interest (e.g., cholinesterase inhibition) for various chlorpyrifos exposure scenarios and for various life stages. In addition, the dose response relationships used in these models need to reflect the understanding gained from animal studies, what is known about inter-species and intra-species differences, and be validated against data from deliberate dosing studies in humans and epidemiological studies. The Panel encouraged the Agency to continue examining the importance of all enzymes in the metabolic pathway for chlorpyrifos, and acknowledged the data gaps for carboxylesterases and P450 enzymes.

The Panel discussed alternative approaches for calculating the uncertainty factors in the chlorpyrifos risk assessment.

DETAILED RESPONSES TO CHARGE QUESTIONS

1. Metabolism & Toxicokinetics (Issue Paper Section 3.1, Appendix A):

The Agency has performed a literature review of *in vivo* and *in vitro* studies on the metabolic profile and toxicokinetic (TK) properties of chlorpyrifos with particular focus on age-dependent and lifestage sensitivity.

a. The Agency has concluded that age-dependant sensitivity, at least in part, is derived based on toxicokinetic (TK) differences between juveniles and adults. These TK differences lead to reduced ability to detoxify chlorpyrifos or the oxon in juvenile animals. *Please comment on the Agency's conclusion and the scientific support for or against this conclusion.*

Panel Response

The Panel supported the Agency's conclusion. A weight of evidence evaluation of the available information shows that sensitivity to the adverse effects of chlorpyrifos is influenced by age, with young animals having lesser total ability to detoxify chlorpyrifos as well as many other OPs. The Panel agreed with the Agency's conclusion that the age-dependent sensitivity is mostly based on TK differences between juveniles and adults rather than soley on TD differences.

Chlorpyrifos is activated by oxidative desulfuration to the oxon which is further detoxified by cytochrome P450-mediated dearylation. A second detoxication mechanism involves A-esterases (PON1 activity towards chlorpyrifos-oxon) and B-esterases (carboxylesterases, cholinesterases) operating only on oxon that is available to them following the bioactivation reaction, and hence these are the second enzymes in the pathway. These reactions take place mainly in the liver, although they can also occur to a minor extent in other tissues.

The Panel agreed that, based on the current scientific data, differences in metabolism and biotransformation seem to play a more prominent role than age-dependent differences in the uptake, transport, sequestration, and/or excretion of chlorpyrifos. They also indicated that other potential TK differences between juveniles and adults should not be dismissed. For instance, it

was mentioned that the high respiratory rate of children enhances their absorption of any chlorpyrifos present in the air. Children have a higher skin surface-to-weight ratio that favors dermal absorption and a higher permeability of the small intestine epithelium that facilitates absorption by the oral route. The relatively greater cardiac output of juveniles, together with their less developed blood-brain barrier may facilitate some of the circulating chlorpyrifos-oxon eventually reaching the brain. Children have a greater cardiac output compared to adults, facilitating some of the chlorpyrifos-oxon reaching the brain. The blood-brain barrier in humans is not complete until 6 months of age (Rodier, 2004), facilitating some of the circulating chlorpyrifos-oxon in reaching the brains of exposed infants

Polymorphisms in several of the enzymes involved in chlorpyrifos metabolism can lead to variability in an individual's ability to detoxify the chlorpyrifos-oxon. Some enzymes are also inducible which may also contribute to variability in response to chlorpyrifos (and its metabolites). Most of the studies evaluating biotransformation capacity with acute sensitivity are correlative in nature, however, and are not mechanistic. While isoform expression of cytochrome P450 (CYP450) can be markedly different between juvenile and adult animals, the relative roles these isoforms play in sensitivity to chlorpyrifos are not clear. Juveniles appear to be less efficient than adults at activating chlorpyrifos to its oxon metabolite using CYP450. Nevertheless, the dearylation-to-desulfuration ratio in adults and juveniles is unknown, but may differ as a function of age.

Choi et al. (2006) stated that the adult (human) liver plays an important role in detoxication of chlorpyrifos and that in the liver the oxon does not accumulate to be released into the bloodstream. Hunter et al. (1999), however, suggested that the fetal liver does not play a significant role in detoxication. Some studies suggest that chlorpyrifos oxon formed in the liver does not escape to enter the bloodstream (Sultatos et al., 1984). Poet et al. (2003) stated that at low oral doses of chlorpyrifos, CYP/PON1 in the intestine and liver may effectively remove the oxon from circulation prior to systemic exposures. By contrast, individuals with lower levels and/or activity of the detoxication enzymes may have these pathways become saturated at high exposure levels. At high acute dosages, as is frequently the case in laboratory animal experiments, the detoxication systems are operating at maximal levels and the differences between adults and juveniles are more readily apparent. However these saturating conditions are less likely to occur at environmental exposure levels. If the detoxication mechanisms are not saturated, then their effectiveness in adults and juveniles may not be as different as experimental animal data with high exposures suggest. The lack of available data on maturational differences in tissue (non-blood) detoxication capacity by A-esterases and other pathways limits extrapolation of relative sensitivity in infants based on differential detoxication.

Timchalk et al. (2006) reported that the rate of detoxication (measured as the formation of the chlorpyrifos specific metabolite, 3,4,6-trichloro-2-pyridinol, referred to as TCP) exceeded the rate of activation of chlorpyrifos (measured as chlorpyrifos oxon formation) in rats across age groups from 5 days to adulthood, with exposures ranging from 1-10 mg/kg. This would suggest that at similar exposures, inactivation processes may outweigh the net activation. At higher exposures, inactivation might become compromised. At low chlorpyrifos concentrations, the formation of non-toxic metabolites is highly favored in the fetus (Buratti et al., 2006).

In evaluating the role of toxicokinetics and more specifically the lower detoxication potential in juveniles compared to adults, the Panel proposed that the relatively inefficient bioactivation reaction, especially in juveniles, would not likely lead to high concentrations of the oxon at environmental exposure levels. The detoxication enzymes would not be at saturating substrate concentrations for efficient inactivation. Age differences in toxicokinetics are likely to be important at high dosage levels, but less likely to be relevant at lower dosages where enzymes are not saturated.

The two detoxication pathways studied the most, in relation to age-related differences in sensitivity, are carboxylesterases and A-esterases (primarily paraoxonase-1 referred to as PON1). Specific activities of liver carboxylesterases (CarbE) increased with age in male rats (Atterberry et al., 1997) which indicated the presence of more protective esterases in the adult as compared to the juvenile animal. The lower carboxylesterases activity in blood and tissues in juveniles appears to play a role in the differential sensitivity of juvenile animals to chlorpyrifos compared to adults (Morgan et al., 1994; Moser et al., 1998; Karanth and Pope, 2000). However, humans do not have carboxylesterases in the plasma (Li et al., 2005) and some data suggest relatively minimal differences in liver carboxylesterases between infants and adults (Pope et al., 2005). On the other hand, juveniles contain low serum albumin levels as compared to adults, and albumin has been reported to hydrolyze chlorpyrifos-oxon (Sogorb et al., 2008), and is capable of protecting AChE (acetyl cholinesterase) "in vitro" from inhibition at low concentrations of this toxic metabolite (~ 0.1 μM).

Although BChE -/- (butyrylcholinesterase knockout) mice showed relatively similar toxic responses as BChE +/+ mice following chlorpyrifos oxon exposure (Duysen et al. 2007), the contribution of BChE to the metabolism of chlorpyrifos-oxon in humans remains to be determined. This finding can be explained by the high carboxylesterase activity in mouse plasma. Given that carboxylesterase is lacking in human plasma, the protective role of BChE cannot be disregarded.

The Panel was not aware of data specifically demonstrating that chlorpyrifos can be bioactivated in the brain, the main target organ of this compound. However, there are data from "*in situ*" experiments in rats that show bioactivation of parathion (another insecticide of the organophosphorothioate class) in the brain in the intact organism (Chambers et al., 1989 and 1991). Buratti et al. (2005) hypothesized that independent of the chemical structures, organophosphorothioates are bioactivated by the same CYP450s. Since chlorpyrifos is bioactivated less efficiently than parathion by rat liver microsomes (Ma and Chambers, 1994, 1995), it is certainly possible that bioactivation of chlorpyrifos occurs in the brain since very low desulfuration activity of various phosphorothioate insecticides, including chlorpyrifos, has been reported in both microsomal and crude mitochondrial fractions from brain (Chambers and Chambers, 1989). The brain desulfuration activities of these phosphorothioates generally correlate well with the toxicity and may be important in determining their overall acute toxicity levels (Chambers and Chambers, 1989). Thus, extrahepatic sites of activation likely play an important role in mediating the acute toxicity of chlorpyrifos (Sultatos et al., 1984).

PON1 is differentially expressed throughout maturation with lower levels in younger animals being associated with higher acute sensitivity (Mortensen et al., 1996; Li et al., 1997; Karanth

and Pope, 2000). The Panel noted that there is less information suggesting that human infants are more sensitive than adults to chlorpyrifos toxicity because of age differences in PON1-mediated detoxication. In humans, blood A-esterase is very low at birth and increases for the first two years of life (Augustinsson and Barr, 1962; Ecobichon and Stephens, 1973; Burlina et al., 1977) or up to 4 years according to more recent data from large cohorts (Holland et al., 2006).

Errors in the metabolism section of the Agency's background materials provided to the FIFRA SAP for this review were pointed out by one Panel member.

> b. There are limited data on the metabolic capacity of pregnant animals and pregnant humans. These limited data on metabolism are supported by some toxicity data in rats. The Agency believes these studies suggest that pregnant animals and humans may be somewhat more sensitive to chlorpyrifos than non-pregnant adults to chlorpyrifos. *Please comment on the Agency's preliminary conclusion and the scientific support for or against this conclusion.*

Panel Response

The Panel agreed with the Agency's preliminary conclusion that "pregnant animals and humans may be somewhat more sensitive to chlorpyrifos than non-pregnant adults" based on metabolic capacity and their effect on levels of key enzymes in modulating chlorpyrifos toxicity. However, panel members noted that only relatively small differences in levels of the key detoxication enzymes involved in chlorpyrifos metabolism have been reported. Some panel members recommended that a direct comparison of the metabolic capacity of pregnant versus non-pregnant females be performed. Other panel members strongly disagreed with this recommendation.

The Panel noted that it is not clear why pregnant females would be more susceptible to the adverse effects of chlorpyrifos than non-pregnant females. The information on toxicity of chlorpyrifos in pregnant animals is limited and the data on biotransformation of chlorpyrifos in pregnant females are inadequate. Reports from the American Association of Poison Control Centers (AAPCC) do not suggest increased susceptibility of pregnant women to chlorpyrifos and other OPs. The physiological response to pregnancy includes an increased vascular volume resulting in relevant hemodilution, along with higher cardiac output and increase in liver weight. These physiological changes may impact liver and serum enzymes involved in chlorpyrifos metabolism that contribute to some extent in the variability of activity observed during pregnancy. Most of the gestational studies published so far report that there is greater concern for the fetus rather than the dam. These studies are inadequate to determine whether the fetus is more sensitive than postnatal pups. The sensitivity to chlorpyrifos clearly decreases when dosing occurs with increasing age in postnatal pups. However, the increased sensitivity does not seem to occur when pups are repeatedly exposed to low doses. In fact, when cholinesterase activity, muscarinic receptor binding, and motor activity changes in response to the antimuscarinic drug scopolamine were compared in juvenile and adult rats treated repeatedly with chlorpyrifos, adults showed more extensive changes (Chakraborti et al., 1993). As noted before, this may be due to more rapid synthesis of acetylcholinesterase molecules in tissues from younger animals

between exposures, thus leading to lesser accumulation of inhibition across exposures in the younger animals.

The Panel discussed the biological significance of the possible changes in metabolic activity during pregnancy relative to expected toxicology outcomes. In humans, the main three CYP450 isozymes that metabolize chlorpyrifos are CYP2B6, CYP2C19, and CYP3A4. The net effect of these isozymes on the balance of desulfuration/dearylation is unknown since CYP2B6, CYP3A4 and CYP1A2 promote the activation of chlorpyrifos to chlorpyrifos-oxon whereas CYP2C19 detoxifies the oxon by means of a dearylation reaction. Whereas, CYP3A4 increases during pregnancy, CYP2C19 decreases (Anderson, 2006) and no data on CYP2B6 in pregnant women are available. Pregnancy-induced decreases in levels of the CYP2B1/2 protein, which is the orthologous form of human CYP2B6, has been reported in rat liver but this is not accompanied by a statistically significant decrease in pentoxyresorufin O-dealkylase (PROD) activity (Czekaj et al., 2000).

Review of the literature indicates that there appears to be a reduction in the activities of several enzymes potentially important in the detoxication of chlorpyrifos associated with pregnancy, including PON1, carboxylesterases and blood cholinesterases. The Panel agreed with the Agency that the "importance of the decreases (in detoxifying enzymes) is unknown at environmental exposures." Although the importance of these decreases under low environmental exposures is not known, decreases during pregnancy may indicate a corresponding reduced capacity to detoxify chlorpyrifos. While the enzymes mentioned above are expected to have lesser (or perhaps no significant) role in modulating toxicity with low level exposures, the limited evidence does suggest a potential for an overall reduced capacity to detoxify chlorpyrifos during pregnancy.

2. Cholinesterase Inhibition (Issue Paper Section 3.2, Appendix B):

The Agency has reviewed numerous studies submitted for pesticide registration and from the literature in animals and human on the AChE-inhibiting effects of chlorpyrifos in blood and in the peripheral and central nervous system.

a. Regarding inhibition of AChE, the Agency has preliminarily concluded that post-natal studies in rat support the conclusion that juveniles are more sensitive than adults. The Agency has further concluded that sensitivity is greatest in younger pups and decreases as pups mature towards adulthood. *Please comment on these Agency's preliminary conclusions and the scientific support for or against these conclusions.*

Panel Response

The Panel agreed with the Agency's conclusion that post-natal rats are more sensitive than adult rats with respect to increased AChE inhibition and lethality following acute exposures. A number of studies have consistently shown that juvenile rats are more sensitive than adults to cholinesterase inhibition and cholinergic signs of toxicity following acute doses of chlorpyrifos exposure. In addition, several studies show a gradient of sensitivity to acute chlorpyrifos in rats during postnatal maturation. This greater sensitivity appears due to a lower detoxication capacity and as these capacities develop, the rats become more aligned with the lower adult sensitivity.

However, this greater sensitivity may be true mainly for high dosages because of the saturation of the detoxication mechanisms. This difference in sensitivity (measured as acetylcholinesterase inhibition) between juveniles and adults is less likely to be apparent at low environmentally relevant dose levels because the detoxication mechanisms will not be saturated. Some on the Panel felt that physiologically-based pharmacokinetic/pharmacodynamic (PBPK/PD) models are likely to be of greatest value in predicting such age-related differences over a span of dose levels.

The Panel, however, did not support the suggestion that post-natal rats are more sensitive than adult rats under repeated exposures. The timing of the measurement of cholinesterase inhibition, as well as, the timing between exposures can be pivotal in the degree of inhibition observed. Results in juveniles can be confounded by their higher capacity to synthesize of new AChE molecules. For example, Chakraborti et al. (1993) reported that juvenile rats are less sensitive than adults under subacute chlorpyrifos dosing (i.e., 40 mg/kg/day, every four days for 4 doses). Four days after the last exposure, adult rats showed 90-92% inhibition of brain cholinesterase while juvenile rats show only 54-59% inhibition. Greater inhibition of brain cholinesterase in adults was correlated with more extensive reductions in muscarinic receptor binding. These receptor changes were correlated with enhanced locomotor responses to the antimuscarinic drug scopolamine in adults only, out to eight weeks after the end of dosing. Thus, under these conditions, adults appear markedly more sensitive to cholinesterase inhibition and cholinergic effects of chlorpyrifos. Liu et al., (1999) report that repeated, daily chlorpyrifos exposures (5 mg/kg/day, for 14 days) were associated with relatively similar degrees of brain AChE inhibition between juveniles and adults 1 day after either the 7th or 14th dose, but more extensive inhibition was noted in adults 8 days after termination of dosing. Relatively similar changes in muscarinic receptor binding were also noted between age groups following daily exposures. Together, these studies illustrate the ability of brain cholinesterase to more effectively recover between repeated doses in pups and the possibility of relatively more extensive cholinergic responses in adults following repeated chlorpyrifos dosing under the same conditions (repeated, "low" dose exposures).

Overall, the Panel decided that a conclusion of greater sensitivity to cholinesterase inhibition in juvenile rats compared to adult rats only holds for acute high exposure situations. When acute exposures occur at environmental concentrations and/or under repeated (low-level) exposures; however, many on the Panel believed that higher sensitivity in younger animals under "environmental concentrations" would be unlikely. Other members stated that this conclusion cannot be made without having a better idea of the meaning of "high exposure level situations" or "environmental concentrations."

b. There are multiple gestational studies available which provide AChE data in dams and/or fetuses. These gestational studies have consistently shown that AChE inhibition observed in the dam is greater than in the fetus. These studies suggest that the dam serves to protect the fetus. However, TK gestational studies have shown that fetal tissues have similar or higher levels of chlorpyrifos and/or its metabolites than the dam. In addition, multiple studies have shown that recovery from AChE inhibition is more rapid in juveniles compared to adults. This rapid recovery combined with production of the AChE enzyme as the rats mature leads to less AChE inhibition observed in the juveniles. The Agency has preliminarily concluded that AChE inhibition data from repeated dosing gestational studies showing less inhibition in fetuses may

not reflect the true potential toxicity to the fetus. *Please comment on the Agency's preliminary conclusions and the scientific support for or against this conclusion.*

Panel Response

The Panel agreed with the Agency's preliminary conclusion regarding observations of lower brain cholinesterase inhibition in pups (rats) compared to dams following gestational OP exposures. However, as noted in the previous question on the findings from the juvenile versus adult studies, the cholinesterase activity measured depends on the time of sampling relative to time of exposure. Whether one considers gestational or postnatal exposures, the longer the time from dosing to measurement of cholinesterase inhibition, the lesser the inhibition expected in younger animals. If the time between dosing and measurement is held constant and the same for dams and pups, one would expect lesser inhibition in pups compared to the dam. This is thought to be due to the more rapid synthesis of new proteins including cholinesterase in tissues of younger animals. Thus, the addition of newly synthesized (uninhibited) cholinesterase molecules in younger individuals contributes to more effective and rapid recovery of activity.

The Panel also agreed with the Agency's preliminary conclusion that "AChE inhibition data from repeated dosing gestational studies showing less inhibition in fetuses may not reflect the true potential toxicity to the fetus." The Panel based its decision after review of those studies indicating the sensitivity to cholinesterase inhibition at dosages of chlorpyrifos sufficient to elicit AChE inhibition. One Panel member cautioned about equating AChE inhibition to toxicity, as is implied in the Agency's question. Inhibition is a sensitive biomarker of exposure and is a precursor to at least some, if not all, of the adverse acute effects of AChE organophosphates. In addition, there can be no cholinergic toxicity without previous AChE inhibition. Lassiter et al. (1998) confirmed less inhibition of brain cholinesterase of pups treated daily with chlorpyrifos (7 mg/kg/day) from GD14-18, when assayed from 2 to 48 hours after the last dose compared to peak inhibition measured at 5 hours after a single dose of chlorpyrifos (10 mg/kg) on GD18. Relatively similar inhibition was noted in both dam and fetus (approximately 40% in the fetus and 50% in the dam) for the acute dose treatment. Hunter et al. (1999) also found that peak AChE inhibition was 5 hours for the dam and fetus and this paralleled the time of peak TCP levels in the brain.

Lassiter et al. (1998) considered several mechanisms for differential ChE inhibition, but concluded that more effective recovery of enzyme activity following each daily exposure in the fetal tissues prevented accumulation of ChE inhibition, while slower recovery of activity in the maternal brain allowed increased ChE inhibition throughout the dosing period. This is essentially the same mechanism described earlier in postnatal pups to explain the lesser cholinesterase inhibition and effects on other cholinergic markers in pups as compared to adults that were treated at subactute doses with chlorpyrifos (Chakraborti et al., 1993). The recovery of inhibited AChE, i.e., the apparently lower level of AChE inhibition in juveniles compared to adults, is very likely due in large measure to greater synthesis of new AChE molecules during growth. In animal studies, it would be unlikely that significant "synthesis of new AChE" would occur in a 5 hour window to a level such that it would alter AChE activity and give the impression of less inhibition. In fact, AChE activity in control animals changed very little between 2, 5, and 10 hours following exposure as reported in Lassiter et al. (1998). Thus, AChE measurements prior

to 5 hours may be an accurate reflection of the amount of AChE inhibition, but AChE measurements beyond that point are subject to question.

The Panel agreed that the timing of the AChE assay following exposure may be less important when evaluating the degree of cumulative inhibition in dams following repeated dosing, but critical when evaluating the more rapid inhibition and recovery in the fetus or pup. A more accurate assessment of AChE inhibition in the growing juvenile could be determined if the timing of the sample collection were closer to the time of peak effect and if more interim sacrifices were used in these experiments. Some on the panel recommended more studies to examine AChE inhibition resulting from lower doses of chlorpyriphos in dams and developing neonates/pups, at more time points, as well as more carefully quantifying AChE protein levels as well as AChE inhibition.

Regardless of whether there was recovery of AChE inhibition or increased AChE synthesis, this event is expected to be of little consequence at levels of exposure that are below those inhibiting the target enzyme. It was pointed out that the human fetus is slower developing than the rat, so rapid synthesis would not be present. Some Panel members felt that in the human fetus there would be no inhibition at all at environmentally relevant levels of exposure. But another member indicated that inhibition might be a concern for exposures experienced by agricultural workers. Many agricultural workers are women and some of those may be pregnant when exposed to higher levels. One Panel member stated that predicting the toxicological effects in humans based on the available data is too risky based solely on animal studies.

3. Laboratory Studies on the Developing Brain (Issue Paper Section 3.3, Appendix C):

The Agency has performed a literature review of *in vivo* and *in vitro* studies on the effects of chlorpyrifos on the developing brain.

a. From a review of laboratory animal studies, the Agency has preliminarily concluded that gestational and early postnatal exposure at sufficiently high exposures to chlorpyrifos can lead to neurochemical and behavioral alterations persisting into adulthood after any initial AChE inhibition has reversed. The Agency has put particular emphasis on the behavioral data because studies are available from multiple laboratories. *Please comment on the Agency's preliminary conclusions and the scientific support for or against this conclusion.*

Panel Response

In general, the Panel was in agreement with the preliminary conclusions made by the Agency. The preliminary scientific findings and conclusions are supported by the review of the *in vivo* neurobehavioral data from multiple laboratories on two mammalian species (i.e., rat and mice) of gestational exposure and/or early postnatal exposure to chlorpyrifos. The Panel concluded that independent of the exact time frame of the gestational or postnatal exposure, only doses of 1mg/kg or greater demonstrate significant effects on behavior. However, only two studies to date (Maurissen et al., 2000; Jett et al., 2001) have examined developmental effects at the lower dose level of 0.3 mg/kg, and neither study is considered sufficient (see below). The Panel noted

that the inclusion of some more recent studies would add to the number of laboratories involved, as well as increase the number of neurobehavioral endpoints examined. One panel member provided a table of studies that supplements the information provided in Table 3 of the Agency's issue paper (see Appendix 1).

Two areas of concern were identified and discussed. The first involved methodological issues in the studies reviewed. The second was conceptual, and involved the use of the phrase "sufficiently high exposures" in the Agency's charge question.

The methodological issues centered on the consistency of behavioral endpoints reported as significant. The Panel noted that although behavioral findings were consistently found, (refer to Table 3, Agency Issue Paper), a somewhat inconsistent set of behavioral changes were reported. In some cases, motor activity is increased and in others, it is decreased. In learning and memory tests, error rate can be increasing, decreasing or not affected under similar treatment conditions. Increased and decreased habituation was reported with different gestational exposure paradigms. Gender differences are also reported in some studies. In studies comparing exposure level, lower dosages sometimes caused greater effects. These inconsistencies likely reflect differences in route of administration, developmental period during exposure to chlorpyrifos, method used to measure changes in the same behavioral domain, and/or choice of dependent variable. For example, motor activity can be measured by distance traveled in an open field and electronically by beam breaks. Moreover, different studies use different types of testing chambers, or number of crossings in an elevated plus maze. The use of several detection methods for motor activity may present difficulty in comparing across studies, especially with respect to younger animals. Concerns were also raised about the high level of variability of measurements and its effect on such endpoints as the mean startle response (see Maurissen et al., 2000).

While the Agency put particular emphasis on the behavioral data, primarily because it is available from multiple laboratories, all related data were considered in identifying long-term changes in the nervous system. With respect to the Maurissen et al. (2000) study, the Panel raised concerns on the use of static morphometrics to evaluate a dynamically changing brain structure. Measurement of the hippocampal dentate granule cell layer at postnatal day 12 would potentially capture a structural assessment of a region with poorly defined landmarks at a time when the neurons are undergoing an active period of migration. In the adult brain, morphometric measurements of the cortical regions displayed about 10% variability, a level expected to be within the normal variability for such crude measurements. Unbiased stereology is recommended by the Panel to address questions of regional volume or cell number.

Several Panel members specifically noted methodological shortcomings in the Jett et al. (2001) study. Although it is only one of two studies to investigate behavioral changes after a dose of 0.3 mg/kg of chlorpyrifos, there were significant problems with respect to the training paradigm i.e., the Morris water maze [MWM]. Animals were not tested to asymptote and only 60% learning was evident in the control group. Neurochemical data indicated that AChE inhibition was not observed at a dose of 7 mg/kg, a finding at odds with other published studies. In addition, behavioral changes were noted for the 0.3 mg/kg/dose only when dosing continued during testing. This represents a confounding factor in determining the acute versus development effects of chlorpyrifos.

The Panel agreed with the Agency's expressed caution about the use of DMSO as a vehicle because of its intrinsic toxicity and influence on absorption. In addition, the "observed signs of discomfort" in pups injected with DMSO reported by Marty et al. (2000), and more recently by Carr and Nail (2008), raised concerns about the use of this vehicle in developmental studies. An additional concern was raised by the Panel concerning the influence of DMSO on the toxicity of the compound under study. Ballough et al (2008) reported that DMSO, administered at levels below those used in many developmental chlorpyrifos studies, enhanced the toxicity of soman by exacerbating the neuropathology caused by soman-induced seizures. The interaction of DMSO and soman, an OP, raised concerns about a possible DMSO-chlorpyrifos (also an organophosphate) interaction in developmental studies. Additional concerns were raised regarding both DMSO and ethanol as vehicles used not only in the *in vivo* studies, but also in the *in vitro* studies.

There was also discussion about the varied routes of administration (ROA) of chlorpyrifos used for developmental exposure. The Panel noted that each ROA used presented drawbacks and most if not all were stressful. It was generally agreed that the subcutaneous ROA was valid for gestational exposure. Moreover, despite a variety of routes used in the literature, there are consistent reports of long-term behavioral changes in offspring after recovery of cholinesterase inhibition.

The Panel noted that many of the papers cited by the Agency do not link exposure periods to chlorpyrifos with structural changes occurring in the developing brain. While the EPA developmental neurotoxicity guideline studies defined a broader period of developmental exposure, more recent studies from the peer-reviewed literature have attempted to demonstrate that there are distinct developmental windows of vulnerability. While effects of chlorpyrifos can be detected in the older animal a direct linkage between the developmental window with regard to the stage of brain development and endpoints examined was ill-defined. It was suggested that a more specific examination of the critical events occurring at the time of exposure and the specific targeted endpoints of interest would be a valuable contribution to the literature. Such data would be particularly important to distinguish cholinergic versus non-cholinergic modes of action for specific adverse effects. Clancy et al. (2007) and the website, http://www.translatingtime.net, were suggested as potential resources for the design of such studies with respect to timing of brain development.

The Panel indicated that the phrase "at sufficiently high exposures" is open to interpretation. Overall, it was interpreted by the Panel in this meeting to refer to studies in which the exposure level was within a dose range expected to inhibit brain AChE. The majority of studies reviewed by the Agency focused on an exposure near 1 mg/kg/day, a level generally sufficient to inhibit brain acetylcholinesterase in most treatment/age groups studied, and which produce behavioral alterations. However, measurements were often obtained at a single time point post-dosing and at a time when the period of peak inhibition may be missed. The availability of dose-response data was limited and in some studies the calculated levels of inhibition were not consistent with the predicted levels of inhibition for high dose exposure. Studies examining critical developmental periods for exposure were primarily focused on dose levels expected to produce AChE inhibition. Given the nature of the gestational exposure studies, the use of fetuses for

neurochemical analysis is limited. Given the critical role of the cholinergic system for brain development, the lack of sufficient data to support the absence or presence of enzyme inhibition is a significant problem in the interpretation of the results from many of these developmental studies. The inherent problems in assessing brain AChE inhibition during development and the type of measurements needed for the assessment were noted in the Agency's document. The Panel noted that it is possible that the morphogenic role of AChE may alter aspects of brain development and neurogenesis, even in the absence of detectable levels of inhibition.

The critical point stressed was that dosages considered "sufficiently high" to elicit neurobehavioral disruption into adulthood would also be anticipated to affect the target enzyme for the common mechanism of toxicity for chlorpyrifos and other organophosphate insecticides. Whether any such changes were initiated by early inhibition of acetylcholinesterase remain unclear.

Errors in the Agency's Issue Paper described below were noted.

1) Section 3.3.1, page 44, paragraph 3. The behavioral effects in the Maurissen et al. (2000) study are only limited to an increased latency to peak response in the auditory startle response that was independent of the dose of chlorpyrifos. The Agency interpreted the data to mean that there was both a decreased amplitude and increased latency at PND 22.
2) Section 4, page 16, paragraph 4.There is no dose-response for effects of ketanserin on reference memory since increased errors were observed in the low and high dosages but not the medium dosage.

b. Consideration of the mode of toxic action is an important component of risk assessment. The International Programme of Chemical Safety (IPCS) and International Life Sciences Institute Risk Science Institute (ILSI RSI) have developed a Mode of Action (MOA)/Human Relevance Framework which provides structure and transparency to MOA analyses (Meek et al., 2003; Seed et al., 2005 and Boobis et al., 2006). IPCS have combined and extended these components to produce a unified Human Cancer Relevance Framework (IPCS HRF). In this approach, involvement of a series of key events in the MOA is established on weight-of-evidence, using criteria based on those described by Bradford Hill, taking account of factors such as dose-response and temporal concordance, biological plausibility, coherence and consistency. Other MOAs that logically present themselves also should be considered. Once an MOA is established, qualitative and quantitative comparison of each key event between the experimental animal and humans enables a conclusion as to likely relevance of the MOA for human risk. In the case of chlorpyrifos, the Agency has considered the available mechanistic data but has not evaluated these data in the context of MOA/human relevance framework. It has been initially determined that there are insufficient data to develop a series of supportable key events (as in a mode of action analysis[1]) for neurodevelopmental toxicities other than AChE inhibition. The Agency notes a particular lack of data on dose response and temporal concordance that are critical in a MOA framework analysis. There may be other mechanisms which lead to effects on the developing brain but a supportable mode of action(s) can not be elucidated at this time. *Please comment on the Agency's preliminary conclusions and whether there is sufficient*

[1] For information on the Mode of Action Framework, see U.S. EPA ,1999, 2005; Sonich-Mullin et al., 2001; Meek et al., 2003 ; Seed et al 2005 and Boobis, et al, 2006

scientific information to merit a full mode of action framework analysis. If a mode of action framework analysis is pursued, what would be the biologically plausible hypotheses to evaluate?

Panel Response

There was a consensus of the Panel that available data were inadequate to support a weight of evidence evaluation for non-cholinergic mode(s) of action for the behavioral alterations following gestational and early postnatal exposure to chlorpyrifos that persisted into adulthood. The Panel agreed that the available information does not allow for behavioral endpoints to be considered as a point of departure and recommended, based upon currently available data, that cholinesterase inhibition be used as the PoD.

Pointed out at the beginning of the discussion of this charge question was the fact that a mode of action/human framework analysis requires consideration of testable hypotheses with identified key events in a causal pathway. It then requires comparison of systematic analyses of traditional criteria for weight of evidence for each of these hypotheses including consistency, dose-response and temporality among early key and end events, biological plausibility and coherence; all of which requires expert multidisciplinary input. The framework, therefore, focuses attention early on key events that can be compared between species and how they are quantified, ultimately critical points in driving the dose-response relationship. While the structured analysis contributes to increased transparency in weight of evidence in risk assessment, it is also helpful in framing research relevant to risk assessment and permits iterative dialogue between the research and risk assessment communities, as a basis to generate more appropriate data.

The data presented in Appendix C of the Agency's issue paper summarizes the studies on persistent behavioral changes in adults following gestational and early postnatal exposure as well as selected *in vivo* and *in vitro* studies on the interaction of chlorpyrifos with several neurochemical parameters. The Agency noted in its issue paper that during development of the brain, acetylcholine plays a role in morphogenesis, and AChE may alter aspects of neurogenesis in the absence of a detectable level of inhibition using current methods. The Panel indicated that there is a large amount of variance presented in many of the studies, a lack of dose response for many of the proposed targets, and a lack of reports of altered targets from more than one lab. The Panel concluded that these studies were not inclusive, coordinated, nor conducted in the context of hypothesis testing for mode of action. Rather they were conducted principally for generating testable hypotheses. Without clearly defined and testable hypotheses we are left with uncertainty about whether the long term behavioral changes occur downstream from AChE inhibition or independently of it.

Specific points were raised regarding the question of mode of action. In Figure 5 of the issue paper, the Agency cites Slotkin et al. (2006) to suggest possible mechanisms for how chlorpyrifos may elicit neurodevelopmental effects. The Panel did not find enough information from the Slotkin et al. (2006) paper to determine if chlorpyrifos directly interacts with the molecular components of the proposed pathways that could then lead, in a causal manner, to subsequent neurobehavioral changes. Panel members determined that it was generally unclear in what way the animal studies exhibit "qualitative similarities" to findings reported in children. The animal studies do report neurobehavioral changes in rats and mice following prenatal or

postnatal chlorpyrifos exposures at levels that would be expected to inhibit cholinesterase. These "sufficiently high" exposures may lead to persistent neurobehavioral effects under some conditions, but "qualitative" similarities to neurodevelopmental findings reported in children are difficult to equate.

There are several facets of data evaluation that may need to be examined with respect to effects on serotonin, G-proteins, macromolecules, neurotrophic factors etc. by developmental exposure to chlorpyrifos. For the most part, the effects seen on these systems following chlorpyrifos exposure are modest, and many represent less than a 25% change from control values/levels. These changes were often sex specific and dependent upon the time of exposure and measurement. Although there is always the possibility that such modest changes in neurochemistry and biochemistry will have a significant impact on behavior, replication and validation across laboratories are required. Some Panel members agreed with the Agency's conclusion that, given the effects on learning and memory, characterization of glutamate N-methyl-D-aspartic acid (NMDA) and alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors would be important endpoints in assessing the developmental neurotoxicity of chlorpyrifos.

The use of morphometric measurements of brain regions was questioned with regard to age, i.e., regional measurements were conducted during an active period of brain development resulting in the lack of discrete structures due to neuronal migration. The diffuse structural features at early postnatal ages, e.g., PND12, would confound the ability to obtain an accurate measurement. In addition, in the mature animal a change within the range of 10% for such a crude measurement would be expected to be within the normal variability of control tissue. One panel member indicated a preference for the use of unbiased stereology in obtaining structural measurements of discrete brain regions. However, this approach would also be hindered during early postnatal development. Unbiased stereology is a well established method for determining cell number and tissue volume and has been used in developmental, aging, and pharmacological studies. An additional concern was raised in that changes often occur in the network organization of the brain rather than gross cell death. A number of different neural and vascular cell types can be influenced by acetylcholine during development and such effects may not involve cell death but rather a structural or functional alteration. While standard histological assessement of hemotoxylin and eosin stained sections accurately detect cell death and general organizational features of the brain, neither it nor morphometric measurements would detect less than gross cell loss occurring prior to such assessment. Nor would such a standard approach detect changes in the non-neuronal cells within the brain, such as the various glial populations. Additionally, developmental neurotoxicity can be associated with changes in the synaptic organization of the various brain regions and the normal temporal pattern of apoptosis and synaptic pruning, differentiation of radial glia, neuronal migration, synapse stripping, maturation of the resident brain monocytes, the microglia, and the brain vascular system. Thus, to determine low-dose effects for any chlorpyriphos will require more sophisticated methods of analysis.

One Panel member took issue with the material in Section 9: *Problems with studies from the literature*, and in particular with Paragraph 5, Page 28 (see Agency's issue paper) which states "An aspect of some of the *in vivo* studies discussed above is that no AChE inhibition was detected during the windows of exposure at the dose level used. This is especially true for

gestational exposures. This suggests that some other target was affected by chlorpyrifos or its oxon." As stated repeatedly in Appendix B, AChE activity increases rapidly in gestational and neonatal animals and, in many studies, especially the gestational studies, the time of assay does not coincide with the time of peak inhibition. Thus, unless a time course of AChE inhibition is performed and clearly demonstrates that there is no AChE inhibition, the claim of no inhibition or low levels of inhibition may not be accurate.

Some panel members suggested that the recent review of chlorpyriphos toxicity by Eaton et al. (2008) provided a basis for considering possible non-cholinergic modes of action. The serine hydrolase enzyme KIAA1363 is important in an ether lipid signaling network involving platelet activating factor, and is highly expressed in cancer cells. *In vitro* studies demonstrated that this enzyme was inhibited by chlorpyrifos-oxon ($IC50^2$ = 8 nM) within the range of AChE inhibition. i*n vivo*, a 98% inhibition of AChE and lethality due to chlorpyrifos-oxon administration resulted in a 39% inhibition of brain KIAA1363 (Nomura et al., 2006). The relative changes have not been reported for lower dose levels thus, a direct causative role in the toxicity of chlorpyrifos has not been identified. So while KIAA1363 may serve as an important detoxication mechanism, as is supported by the increased toxicity in knockout mice (Nomura et al., 2006), it does not necessarily indicate that this enzyme is a target for "mode of action."

At dose levels significantly lower than any previously reported effects, chlorpyrifos induced the phosphorylation of cAMP response element binding (CREB). This transcription factor plays a role in synaptic plasticity and in cell survival and differentiation. In primary cortical or hippocampal neurons, pCREB was elevated following dosing with chlorpyrifos, 0.06 nM and 1-10nM, respectively (Schuh et al., 2002). No effect was observed in primary cortical astrocytes. Schuh et al. (2002) speculated that, rather than indicative of neurotoxicity, the elevation in pCREB represents a neuroprotective response to metabolic stress in neurons. Some Panel members felt that the studies cited in Eaton et al. (2008) (specifically Table 14 and related text) could be useful to the Agency in evaluating potential alternative modes of action.

Collaboration between EPA and developmental neurotoxicologists may help define research needs for identifying alternative neurotoxic processes involved in the developmental neurotoxicity of chlorpyrifos. At a minimum, such studies would require testing multiple doses of chlorpyrifos between 0.05 and 1.0 mg/kg to set the high dose for expected AChE inhibition. Such data can then be used to determine level of cholinesterase inhibition and related changes in brain development. This may require additional methods development for detection of targeted brain regions or cellular localization. The data set can then be used to identify cholinesterase inhibition or an alternative mode of action for developmental neurotoxicity of chlorpyrifos. Low dose level effects reported for *in vivo* developmental effects require replication as well as the extremely low-dose *in vitro* activation of CREB. Inclusion of data on multiple carboxylesterase activities and in localized brain regions are major requirements for future studies.

[2] IC50 (Isolated Cortical) refers to the concentration of chlorpyrifos in cell culture that produced the observed effect (in this case an increase in CREB) in 50% of the neurons.

4. Epidemiology Studies in Children and Mothers (Issue Paper Section 3.4, Appendix D):

The Agency has evaluated epidemiology studies from three major cohorts: the study sites are: (1) Columbia University, NYC, (2) Mt Sinai, School of Medicine, NYC, both with multi-ethnic urban low income women and infants, and (3) University of California at Berkeley (Center for Health Assessment of Mothers and Children of Salinas, CHAMACOS) with women and their children from farm worker populations.

a. The Agency believes that all three studies provide valuable information on the effects in children of high exposures to pesticides, particularly OPs. For purposes of evaluating human health effects of chlorpyrifos, the Columbia University studies provide more robust information for evaluating the human health effects of chlorpyrifos because it measured chlorpyrifos rather than a metabolite in both environmental (air) and biologic media (maternal and cord blood) and showed that chlorpyrifos was significantly associated with birth outcomes (low birth weight and length) and neurodevelopmental outcomes that were no longer present when the residential uses were cancelled (i.e., conducted a pre- and post-residential cancellation analysis). Although the results reported by the Mount Sinai group are informative with regard to evaluating the relevance of PON1 status in health outcomes, this study is limited because the neurodevelopmental outcomes were linked to non-specific OP maternal urinary metabolites (DAP, DEP and DMP), rather than the chlorpyrifos-specific metabolite TCP. The exposure of the CHAMACOS to many OPs reduces its usefulness in the chlorpyrifos risk assessment because the outcomes can not be specifically linked to chlorpyrifos exposure. *Please comment on the Agency's preliminary conclusions on each of the three cohorts regarding the degree to which the data informs the chlorpyrifos human health risk assessment. Please also comment on the scientific support for or against these preliminary conclusions.*

Panel Response

Overall, the Panel agreed with the Agency's conclusion that although each of the three studies (Columbia, Mt. Sinai, and Berkeley) provide valuable information, the Columbia study is the most robust and appropriate for informing risk assessment with respect to the exposure (chlorpyrifos), outcomes (birth and neurodevelopmental), and population (neonates and children) being addressed by the Agency's evaluation. To be precise, the Columbia study was not necessarily a more robust epidemiological study, but it directly measured chlorpyrifos exposure as opposed to the Mt. Sinai and Berkeley studies where metabolites not specific to chlorpyrifos exposure were measured and reported, including those of other OPs and carbamates. The use of the term "high exposure" in the phrasing of the question was questioned because although the exposures in these studies may be high for residential settings, they are not high when compared to agricultural worker and pesticide handler populations.

The Panel noted that the conclusions of the Agency are supported by the following information:

- All three studies are prospective cohorts with relatively low loss to follow-up and are conducted by respected researchers using widely accepted protocols with results published in peer-reviewed journals.

- The Columbia study has the strength of having used blood measures (both maternal and cord) of chlorpyrifos specifically as opposed to the non-specific urinary metabolites (e.g., diethylphosphate (DEP) and dimethylphosphate (DMP)) utilized in the other two cohorts. There is also high correlation ($r=0.76$; $p<0.0001$) between maternal and cord blood chlorpyrifos levels noted in the Whyatt et al. (2003) study. In addition, the Columbia study has exposure measurements from air monitoring which correlate with both maternal and cord blood (Whyatt et al., 2003).

- A particularly convincing strength of the Columbia cohort is that it spans the residential cancellation date allowing the researchers to evaluate changes in birth and neurodevelopmental outcomes pre and post chlorpyrifos residential cancellation. It was noted that although the pre and post analysis validates exposure measurements, the overall results are more interesting epidemiologically because it looks at the entire spectrum of exposure.

- The initial reference group for the birth outcomes (those whose chlorpyrifos levels in cord blood were below the level of detection (LOD) with similar demographic and socioeconomic characteristics) in the Columbia study was also judged to be appropriate.

- The methodology for exposure measurement was considered good by the Panel and the quality assurance and quality control procedures used by the Centers for Disease Control (CDC) are extensive. The Columbia study included both environmental and biological measurements of chlorpyrifos and biological measurements were made in both maternal and cord blood. It was noted that there are concerns about values near or below the detection limits; however, the precision of the data are acceptable while accuracy is reduced. In other words, the Panel felt confident that the chemical was present when detected in a sample although the reported value may be a poor measure of the actual concentration. The difference between the true and measured amount could be appreciable.

- Rauh and Whyatt presentations to the Agency in 2007 and 2008 (presentations found in the docket, EPA-HQ-OPP-2008-0274) showed intelligence quotient (IQ) results at five years of age and indicated intent to extend the follow-up for this cohort until age 7. This work provides the longest follow-up to date for a population of children who were exposed to chlorpyrifos *in utero*.

The Panel concluded that the Columbia study is epidemiologically sound and that there is minimal selection and information bias. Additionally, any information bias would likely be non-differential since any misclassification of outcome is unlikely to depend on exposure status and misclassification of exposure is unlikely to depend on outcome status. However, even the best-designed epidemiology studies are susceptible to weaknesses inherent to observational studies, and a few of these possible weaknesses are addressed below:

- The possibility of residual confounding in the analysis of the Columbia study data on birth outcomes (Whyatt et al., 2004) was discussed. In particular, variables such as

presence of prenatal care, sexually transmitted diseases, history of substance abuse and of cigarette smoking, low pre-pregnancy body mass index (BMI) and inadequate weight gain are potential confounders. Review of the model covariates found that a complete list of standard variables which are commonly available were controlled for in the analysis including passive smoking, ethnicity, parity, maternal pre-pregnancy weight, weight gain, newborn gender, and season of delivery. In addition, the Columbia cohort was restricted to exclude women who smoked during pregnancy, used illicit drugs, had human immunodeficiency virus (HIV), or had their first prenatal visit after the 20th week of pregnancy (Whyatt et al., 2004). The Panel agreed that these factors limit the possibility of residual confounding. In addition, the authors of the Columbia study further addressed the potential for residual confounding by including a variety of other variables in the analysis. The results as presented in the Rauh et al. 2008 paper are stable and not sensitive enough to eliminate the various confounders. Although there may be other variables not included in the analyses that are associated with either exposure or outcome, in order for a variable to be a confounder it must be associated with both exposure and outcome.

- The Panel also noted that the infants in the Whyatt study were of normal weight. The Panel questioned the significance of lower birth weights and lengths in infants more highly exposed to chlorpyrifos. Low birth weight is defined as a weight of 2500 grams (5 pounds 8 ounces) or less. Low birth weight infants are known to be at increased risk for serious health problems as newborns, lasting disabilities and death (March of Dimes; http://www.marchofdimes.com/professionals/14332_1153.asp. Accessed September 21, 2008). Although infants most affected by chlorpyrifos exposure in the Whyatt study fell within normal parameters for birth weight, there could still be cause for concern. Infants who are born smaller than expected by their genetic potential would have increased risk of perinatal mortality, neurological morbidity and morbidity in general as compared to infants born at their predicted birth weight (Figueras et al., 2007). Reductions in birth weight are also associated with marked increase in chronic diseases in adulthood including diabetes, cardiovascular disease, stroke, and hypertension (Barker, 2004). One issue of concern is whether chlorpyrifos' effect on reducing birth weight would result in more infants falling into the category of low birth weight.

- Concerns raised by public commentators before the Panel about neurobehavioral testing, including the specific tests used, the method of administration, and the training of the individuals administering the test were discussed. The Panel concluded that the Columbia study authors used neurobehavioral tests that are widely accepted in the scientific community and used experienced and well-trained examiners to administer the tests. The Panel also concluded that the results obtained were probably as good as could be obtained given all of the limitations associated with neurobehavioral testing.

- Concerns with the three year period between the sampling of cord blood and the neurobehavioral findings in Rauh et al. (2006) were discussed. The exposure classification is based on a one-time measurement that provides a snapshot of exposure and should not be taken as an absolute representation of total chlorpyrifos exposure throughout pregnancy. Many researchers have argued that chlorpyrifos in blood and

adipose tissue are in steady state (see Appendix I, p. 25, Eaton et al., 2008) so that cord blood provides a reasonable dosimeter for the amount transferred to the fetus. This assertion does not take into account the changes produced in lipid metabolism during pregnancy. In spite of maternal fat stores accumulated in early and mid pregnancy, in late pregnancy human chorionic somatomammotropin (HCS) promotes lipolysis and fat mobilization (Butte, 2000) to meet the increased fetal demand at that time; therefore, fat reservoirs are gradually emptying of the accumulated chemicals. Accordingly, the single measurement of cord blood chlorpyrifos may not be representative of the total exposure during pregnancy, but only reflects exposure happened in the few days before delivery. The Panel noted that exposure events, whether chemical or environmental, that could have occurred in these households over the three year time period are not known. These unmeasured exposure events could influence the neurobehavioral test results, especially given the fact that neurodevelopmental deficits may be multifactorial in origin. It would be important to ascertain whether there are statistically significant differences in neurodevelopmental outcomes at 3 years of age between children with chlorpyrifos levels in cord blood below the LOD and those with a chlorpyrifos level of 6.17 pg/g. Rauh et al. (2006) stated that the Bayley Scales of Infant Development scores (Mental Development Index (MDI) and Psychomoter Development Index (PDI)) were similar in both groups of children. If so, chlorpyrifos levels in cord blood at the time of delivery would not be expected to be associated with neurodevelopmental outcomes.

- It is difficult to compare exposure levels in the Columbia study with the US population (as measured in the National Health and Nutrition Examination Survey (NHANES) or with other biomonitoring studies since urinary metabolite levels were not measured.

- Although the data on post-ban declines in exposure are compelling, limitations must be kept in mind when using these results in the weight of evidence. The study was not designed to assess the effect of the ban, so data are essentially cross-sectional (i.e. exposures among the same women were not measured over time). Additionally, the data presented for exposure levels by year have been crude in nature. It is not likely to make a large difference, but an analysis should be conducted looking at exposure level as the dependent variable and year (an ordinal variable) as the primary independent variable, while also adjusting for any factors that may be associated with both exposure levels and year of study.

- There was discussion about the cut-off values and exposure groupings used in the Columbia studies (Rauh et al., 2006; Whyatt et al., 2004). There was also discussion about the numbers in the various exposure groups in 2004 compared to 2006. Concern was expressed that loss to follow-up may have occurred in more severely affected children or less severely affected children as opposed to a non-differential loss. Some felt that more attention should be given to the sensitivity of the results to the selected "highly exposed" group. For example, the authors said these groups were first based on tertiles, but according to Rauh et al. (2006) the groups were unequal in size. The authors did clarify via personal communication on September 18, 2008 (found in the docket, EPA-HQ-OPP-2008-0274) that these groups were unequal in the 2006 study because only children who reached three years of age were analyzed, suggesting that the original exposure groupings were not balanced. Some panel members noted that the Agency

should explore this further as the loss of children in the high exposure group could not be due to length of follow up as all the children in the high exposure group (bar one) had to have been born prior to 2001 (and hence should have been at least three years old) as after the ban there was only one child in the high exposure group. The Panel felt that it would be useful if the authors could provide additional details on the loss to follow-up in each of the four exposure groups and provide additional details on how the exposure groups were combined. However, it was also noted that the inclusion of both the children below LOD and with low exposures values into the reference group would be expected to attenuate any association, if indeed there is an effect at such low exposures. However, in Rauh et al. (2006), there is no suggestion of such an effect as the most highly exposed group and the group with values below LOD had lower mean MDI and PDI scores than did the two middle levels. There was disagreement on the Panel about the mode of action; therefore, there is no compelling reason to believe that the epidemiologic data should reflect either a linear association or a threshold effect.

- The post-ban declines in effect are based on only one subject being classified as highly exposed in post-ban years. This analysis lacks statistical power and may be sensitive to the cutoff value chosen to classify subjects as highly exposed. It would be interesting to see what the relationships looked like among more equally sized exposure groups that separate between the lower exposure levels.

- The Panel felt that it might be helpful to have a longitudinal component to the Columbia study to determine if the adverse effects persisted in those children who exhibited poor birth outcomes or neurobehavioral deficits.

- It would be useful to examine the results of a statistical analysis that includes all three AChE-inhibiting insecticides in the analysis model as dichotomous variables (above or below LOD) in combination with continuous measurements for these variables. This type of analysis would likely not change the results, but it could be helpful in illustrating threshold or dose response effects.

Despite the questions and discussion noted above, it was affirmed that the Columbia study was indeed quite strong and provided extremely valuable information. Most of the points addressed above are points of clarification or points that if addressed could strengthen the analysis and provide the Agency with better data to use in the risk assessment process.

Given the paucity of human epidemiological studies of organophosphates in children, the other two cohorts (Mt. Sinai and Berkeley) should not be completely discounted. Both studies provide information on abnormal reflexes in neonates, and this data is not available in the Columbia cohort. Additionally, the Mt. Sinai cohort considers PON1 as a factor in the analyses, something that may have some relevance to risk assessment (Engel et al., 2007). More information may be gleaned from the Berkeley cohort (Eskenazi et al., 2004). A close comparison of OP metabolite concentrations in this study with those reported in NHANES may be useful. It is interesting to note that median 3,5,6-trichloro-2-pyridinol (TCP, a specific metabolite of chlorpyrifos) levels in this cohort were similar to those reported in NHANES; whereas, DEP (diethylphosphate, one of the non-specific urinary metabolites) levels were much higher. This suggests chlorpyrifos

exposures in the study are similar to "background level" and that exposures to other OPs (besides chlorpyrifos) that are metabolized to DEP are proportionally much higher in this cohort (relative to chlorpyrifos) than the general population. Other OPs may very well be driving the associations between DEP/DMP (non-specific organophosphate urinary metabolites) levels and adverse outcomes. However, because chlorpyrifos is likely to contribute very little to the overall DAP levels in the Berkeley cohort TCP and DAP levels may be poorly correlated. Thus, in the TCP analysis, it may (with some caution exercised) be possible to additionally adjust for DAPs in an attempt to assess more chlorpyrifos (and/or chlorpyrifos-methyl) specific associations. The presence of these other OP pesticides may overshadow any TCP associations, but may still be worthwhile to investigate.

The Panel acknowledged that there are potential confounders and issues that reduce the utility of both the Mt. Sinai and Berkeley cohorts for risk assessment. For example, both studies measure organophosphate metabolites in urine but chlorpyrifos is not specifically measured. The Berkeley study, in particular, has the least relevance to chlorpyrifos risk assessment because only a small percentage (10%) of the pesticides applied in the Salinas Valley are chlorpyrifos; therefore, the Panel assumed that chlorpyrifos would make only a small contribution to the non-specific metabolites measured in the study, and as a result might be expected to have small impact on the study outcomes, although this assumption has not been verified. As such, it is difficult to ascribe the effects seen to chlorpyrifos, in particular, rather than OPs in general.

A recently published study by Samarawickrema et al. (2008) entitled *Fetal effects of environmental exposure of pregnant women to organophosphorus compounds in a rural farming community in Sri Lanka* was brought to the attention of the Panel and suggested for use in clarifying the issue of whether maternal and cord blood are differentially distributed to the fetus. It was suggested that the results from this study could be compared to the results observed in Rauh et al. (2004) and Mattsson et al. (2000). Not having seen the Samarawickrema et al. study, the Panel could only recommend that the Agency consider this study when this issue is revisited. The Panel also encouraged the Agency to identify and review any other published studies on this topic, even if of lesser size and scope, since they may also contribute to the total weight of evidence analysis of the potential neurobehavioral effects of chlorpyrifos.

b. Data from Whyatt et al. (2003) show that 100% of air samples detected three AChE inhibiting pesticides (chlorpyrifos, diazinon and propoxur). Similarly, all three pesticides were found in 48-49% of umbilical cord samples at lower levels than chlorpyrifos. The investigators reported that chlorpyrifos was significantly associated with decreased birth weight and length, even after statistically controlling for these two OPs; a similar analysis has not been conducted for the neurodevelopmental outcomes. The Agency can not rule out that exposures to all three AChE-inhibiting pesticides in combination resulted in the neurodevelopmental health outcomes reported in the studies. However, this possibility does not rule out the potential role of chlorpyrifos in contributing to the reported health outcomes, particularly given the reported findings pre- and post-voluntary cancellation. In balance, given, that 1) measured levels of chlorpyrifos have been statistically associated with multiple birth and neurodevelopmental outcomes; 2) these associations are correlated in time prior to the cancellation of indoor uses of chlorpyrifos when exposures were much greater (and thereby show some degree of dose-response); and 3) there are animal data which support neurobehavioral effects resulting from

gestational exposure, the Agency has preliminarily concluded that chlorpyrifos likely played a role in these outcomes. *Please comment on the Agency's preliminary conclusion and the scientific support for or against this conclusion.*

Panel Response

Overall, the Panel agreed with the Agency's conclusion that chlorpyrifos likely played a role in the birth and neurodevelopmental outcomes noted in the three cohort studies. The Panel agreed with the Agency on the following points: a) exposures to all three AChE-inhibiting pesticides in combination cannot be ruled out as contributing to the birth and neurodevelopmental outcomes, b) the potential of combination and/or additive effects of these three compounds does not rule out the role of chlorpyrifos in contributing to the outcomes, and c) it cannot be stated that chlorpyrifos is the sole contributor to the observed outcomes. These conclusions are supported by the effects observed at various age groups across the three cohorts as summarized in Table 1.

1) **Strength and significance of association** as reflected by the following results from the Columbia cohort:

a. Extremely large odds ratios (OR) for attention disorders (OR=11.26; 95% CI: 1.79-70.99), attention deficit hyperactivity disorder (ADHD OR=6.50; 95% CI: 1.09-38.69), and pervasive developmental disorder (PDD OR=5.39; 95% CI: 1.21-24.11) were seen when comparing high to low chlorpyrifos exposure groups (Rauh et al., 2006). Although limited sample sizes resulted in fairly large confidence intervals, the magnitude of these results is so large that they are unlikely to be affected by residual confounding.

b. There were statistically significant deficits in birth weight of 150 grams when comparing high exposure to exposure that was below the level of detection (LOD); and, decreases of 43 grams in birth weight per log unit increase in chlorpyrifos in cord blood. Statistically significant deficits in birth length were also noted (Whyatt et al., 2004).

c. There were statistically significant deficits of 6.5 points on PDI at 3 years of age when comparing high to low exposure groups (Rauh et al., 2006).

d. There were 2.4 times increased odds (95% CI: 1.1-5.2) of mental delay and 4.5 times increased odds (95% CI: 1.6-12.7) of psychomotor delay when comparing high to low exposure groups at age 3 (Rauh et al, 2006).

e. There were deficits of 5.6 points on verbal IQ and 5.1 points on full IQ at age 5 (Whyatt, EPA presentation, 2007).

2) **Consistency of association** between the three cohorts is that they all found some developmental effect in a population exposed to elevated levels of OPs. The Berkeley and Mt. Sinai cohort studies measured non-specific urinary DAP metabolites (e.g., diethylphosphates (DEPs), dimethylphosphates (DMPs) produced by many OP pesticides including chlorpyrifos. However, urinary levels of 3,5,6-trichloro-2-pyridinol (TCP) were not measured in these two studies, but only in the Columbia cohort study. Therefore, the conclusions below are based on the non-specific DAP metabolites rather

than the more specific indicator, TCP. The Panel noted, however, that urinary TCP does not absolutely correlate with exposure to chlorpyrifos, but could be an indicator of exposure to other pesticides including methyl chlorpyrifos and the herbicide trichlopyr (3,5,6-trichloro-2-pyridinyloxyacetic acid, Remedy®) or even to TCP itself (as an environmental degradate of chlorpyrifos, methyl chlorpyrifos or trichlorpyr). However, developmental effects were consistently observed in children whose mothers exhibited biomarkers of exposure to OPs in each of these studies.

a. There were increased abnormal reflexes in neonates associated with maternal urinary DAP measurements in both the Berkeley and Mt. Sinai cohorts (Young et al., 2005; Engel et al., 2007).

b. There were increases in MDI associated with increases in DEP metabolites at 1 year (Eskenazi et al. 2007). Although Eskenazi and colleagues found decreases in MDI associated with prenatal DEP metabolites at 2 years, they were not statistically significant (see Table 4, Eskenazi et al., 2007).

c. Results from cohort studies conducted by Eskenazi. et al. (2007) indicated total urinary prenatal DAP metabolite measurements were associated with increased odds ratios of pervasive developmental disorder (PDD) at 2 and 3.5 years (Eskenazi et al., 2007) and child urinary DAP metabolites were associated with increased odd ratios of PDD at 2 and 3.5 years (Eskenazi et al., 2007; Eskenazi et al., in preparation as cited in the Whyatt presentation made to EPA in 2007).

d. Results indicated prenatal urinary DAP measurements were associated with decreased verbal IQ and full-scale IQ at 3.5 years and decreased verbal IQ at 5 years (Eskenazi et al., in preparation, Whyatt presentation to EPA in 2007). However, since DMP is definitely not a metabolite of chlorpyrifos, the associations stated here and in all of the previous points must be viewed with caution so as to not overinterpret the significance of the finding

3) A **crude dose-response relationship** as evidenced by the following results from the pre and post cancellation analyses in the Columbia cohort:

a. There were statistically significant deficits in birth weight (211 grams when comparing high to LOD groups) in children born before the residential cancellation, but there were no significant deficits in these outcomes in children born after the residential cancellation (Whyatt, EPA presentation, 2008).

b. Statistically significant decreases in birth weight and birth length per log unit increase of cord blood chlorpyrifos were evident in children born before residential cancellation but the association was not significant for children born post-cancellation (Whyatt et al., 2004).

c. There were statistically significant reductions in maternal personal air samples and cord blood chlorpyrifos levels pre- and post-cancellation and a statistically significant

reduction in the numbers of newborns who were categorized in the highly exposed group (Whyatt et al., 2004).

d. There were statistically significant increases in maternal personal air samples and cord blood chlorpyrifos levels in 3 year MDI and PDI scores when comparing pre-cancellation to post-cancellation periods (Rauh et al., 2006).

4) <u>Persistence of strong statistically significant associations</u> after controlling for the effects of other compounds as evidenced by the following:

a. Additional analyses on birth weight and neurodevelopmental outcomes (MDI and PDI) controlling for diazinon did not reduce the effect of chlorpyrifos (Whyatt, August 12, 2008, personal communication).

b. Association between birth weight and length remained statistically significant after controlling for diazinon and isopropoxyphenol (Whyatt et al., 2004). The authors noted that blood concentrations for chlorpyrifos were higher than those for diazinon and propoxur. The Panel had some questions about these calculations.

Table 1. Neurodevelopmental Effects with Chlorpyrifos and Its Metabolites and the Metabolites of OP Insecticides in Three Epidemiological Human Cohort Studies
Source: Appendix D, USEPA; Eaton et al., (2008), referenced studies

Age	CHAMACOS (Non-specific OP metabolites + TCP measured)	Mt. Sinai (Non-specific OP metabolites measured)	Columbia (chlorpyrifos, TCP, propoxur, diazinon)
Neonate	Abnormal reflexes (maternal urinary DAPs): During pregnancy, total DAPs (summed DEP metabolites and DMP metabolites) with increased number of abnormal reflexes and presence of 3 or more abnormal reflexes. (BNBAS: Brazelton Neonatal Behavioral Assessment Scales) (Young et al., 2005)	Abnormal reflexes (maternal urinary DAP metabolites): Increased total DAPs, (summed DEP and DMP metabolites) with increased proportion and number of abnormal reflexes (BNBAS) (Engel et al., 2007)	
6 months	No association with prenatal TCP and MDI (Motor Development Index)/PDI (Psychomoter Development Index) (BSID-II: Bayley Scale of Infant Development – II) (Eskenazi et al., 2007)		
1 year	• Child DEPs associated with increases in MDI (BSID-II) (Eskenazi et al., 2007) • No association with prenatal TCP and MDI/PDI (BSID-II) (Eskenazi et al., 2007)		
2 years	• Total urinary prentatal DAP metabolites associated with increases in PDD (Psychomoter Development Delay) (CBCL: Child Behavior Checklist)) (Eskenazi et al., 2007) • Child DAPs (measured DMPs, and DEPs)associated with increase in PDD (CBCL) (Eskenazi et al., 2007) • Prenatal DAPs (measured DMP, one DAP) associated with decrease in MDI (BSID-II) (Eskenazi et al., 2007) • Child DAPs (measured DMPs) associated with increases in MDI (BSID-II) (Eskenazi et al., 2007) • No significant association of TCP with MDI or PDI. (BSID-II) (Table 6, Eskenazi et al., 2007)	Prenatal DAPs associated with decrease in MDI (BSID-II) (Engel et al., in prep)	
3 years	IQ deficits associated with prenatal DAPs at 3.5 years (Ezkenazi et al., in prep)		Increased chlorpyrifos with decreased mean PDI, increased percent with cognitive (MDI) and motor problems (PDI), increased risk of mental and motor delay, increased problems with attention and ADHD (Attention Deficit Hyperactivity Disorder) and PDD. Decreased levels of chlorpyrifos after January 2001 associated with improved scores on MDI and PDI. (BSID-II and CBCL) (Rauh et al., 2006)
5,7 years	IQ deficits associated with prenatal DAPs (Ezkenazi et al., in prep)		IQ deficits associated with chlorpyrifos (Rauh 2008, presentation at SAP meeting, see Docket: EPA-HQ-OPP-2008-0274)

Some caveats and areas for further analysis and clarification were noted by various panel members:

1) In the Berkeley cohort, there was a finding of increased head circumference with DEP levels (3 DEPs were measured) in maternal urine and increased length and head circumference with DMP levels (3 DMPs were measured) (Ezkenazi et al., 2004). This could be a chance finding or it may be due to the exposure profile in this cohort. However, the Mt Sinai study found an association between decreased head circumference and maternal urinary TCP levels above the limit of detection (Engel et al., 2007). Due to the nature of the exposure in the Berkeley cohort, it was noted that perhaps these urinary levels do not reflect significant exposures to chlorpyrifos given that DAP metabolites have two potential origins: breakdown of products arising from OPs in general and direct absorption from environmental sources and diet. As measurements of DAP metabolites are non-specific in nature, the particular chemical involved may be one or more of diethyl phosphate insecticides in use. With regard to the Mt. Sinai study, the results indicated that maternal urinary TCP levels reflected not only direct chlorpyrifos exposure, but also exposure to TCP residues directly from the environment or diet. In addition, the Panel noted that PON1 polymorphism may potentially affect those urinary metabolite levels because women with the higher activity phenotype will eliminate higher levels of TCP in urine as compared to those with the lower activity phenotype. However, this statement is not true if exposure levels are low and below enzyme saturation in the case of carriers of a low activity phenotype.

2) In interpreting the Columbia studies, the Agency should bear in mind that diazinon and propoxur as well as other unmeasured potential developmental neurotoxicants were present along with chlorpyrifos, and the exact roles of all of these components in the mixture cannot be determined. In Table 3 of Whyatt et al. (2004) it is seen that the beta coefficients for the association with birth weight are -42.6 for chlorpyrifos and -44.2 for diazinon. The beta coefficient for chlorpyrifos is statistically significant while the coefficient for diazinon shows large variability and is not statistically significant. One panel member questioned whether selecting chlorpyrifos over diazinon is biologically justified although it may be statistically justified. When the effects of chlorpyrifos and diazinon are summed using U.S. EPA methodology (U.S. EPA, 2001), there is a slightly greater reduction in birth weight (b= -49.1). This may indicate that the effect of the combined chemicals is slightly greater than the individual chemicals alone and that there could be potential interaction between the two chemicals with respect to the association. Given these results, some on the Panel questioned whether one should single out just one of the two chemicals when both are present and there is a potential interaction. Rauh et al. (2006) did not consider diazinon or mixtures in the neurobehavioral study but justified the selection of chlorpyrifos based on the birth weight data, ignoring the similarities of diazinon with chlorpyrifos and the significance of the mixtures. There was disagreement among the Panel with this interpretation. Confidence limits as well as the beta coefficients must be considered when interpreting statistical analysis results. The Panel was in agreement that there was potential for interaction between chemicals, but this did not preclude chlorpyrifos as a contributor. The Panel also noted that Rauh did perform an

analysis of the neurodevelopmental effects while controlling for diazinon and the mixture of diazinon and chlorpyrifos (Rauh personal communication to the Agency dated August 12, 2008, found in docket, EPA-HQ-OPP-2008-0274). In this analysis, the authors stated that the combination of chlorpyrifos and diazinon produced slightly more significant effects for MDI than were seen with chlorpyrifos alone. Diazinon alone did not produce significant changes in MDI. This interaction is critically important in interpreting the results.

3) More analysis controlling for multiple exposures and examining additive effects would help to elucidate the role of other organophosphates, but these analyses were not necessary for the panel to affirm that chlorpyrifos likely played a role in the outcomes that were observed. These results are important for AChE inhibition to be considered as a mode of action.

4) Whyatt and Rauh (Columbia University) in their comments to the Panel (September16, 2008) described how they separated the neurodevelopmental effects of chlorpyrifos exposure from those of diazinon exposure. Some panelists questioned whether the biological effects of these anticholinesterase insecticides could be separated.

5) The Agency's use of the word "likely" at the end of the paragraph needs to be considered carefully. The Agency should ensure that the use of the word is not based heavily on the Columbia study in the absence of intermediate markers of biologically effective dose and early effect that further link exposure and outcome and or a clearer idea of mode of action from experimental studies.

6) It is unclear whether the blood levels of the three AChE-inhibiting insecticides are highly correlated. If there is a lack of correlation, it is less likely that they would confound or be responsible for the observed relationships between chlorpyrifos and neurological outcomes. However, it would take a much larger and more detailed study to elucidate the more complex issues involving chemical mixtures (the possibility of synergism, antagonism, potentiation, activation, etc.) Some panelists suggested that chlorpyrifos (or chlorpyrifos oxon) may inhibit certain xenobiotic metabolizing CYP isoforms which may make some individuals more susceptible to exposure to other chemicals (e.g. other OPs, carbamates, pyrethroid insecticides, or other environmental contaminants). This is may be due to the resulting declined efficiency in CYP metabolism in response to these exposures when preceded by exposure to chlorpyrifos (Note: work by Ernest Hodgson's group at North Carolina State University was mentioned). Organophosphorothiate insecticides, including chlorpyrifos, act as "suicide substrates" leading to inhibition of CYP450 (DeMatteis1974; Halpert et al. 1980). Thus, exposure to these other compounds may also affect the metabolism of chlorpyrifos.

7) Other issues with the Columbia cohort study (Whyatt et al., 2004) were identified. The level of diazinon in the homes was 8 times higher than that of chlorpyrifos suggesting that the exposure level to diazinon was 8 times higher. Diazinon is less toxic than chlorpyrifos, and chlorpyrifos is more lipophilic than diazinon. With the difference in lipophilicity, it was not surprising to see more chlorpyrifos than diazinon detected in the

cord blood. There was an additional OP insecticide (methyl parathion) as well as 5 carbamates (including propoxur) present in the homes. These anticholinesterases could be interacting with each other.

8) The following are some clarifications provided by one Panelist on the material presented in Appendix D:

   a. The Agency stated that exposure is underestimated by use of blood levels. The Panelist agrees that this is true for peak exposures and a single blood sample would likely miss the peaks. However, because exposure is multi-media and multi-route, for most women in the Columbia cohort (as well as in the general population) single high insults are likely limited, and exposure may more likely be a series of contact events that are lower in magnitude over time via various routes and media. Thus, spot samples may be a useful way to estimate average exposure for women in the study since there is much lower within-home variability of pesticide levels as compared to between-home variability (Whyatt et al., 2007).

   b. Description of blood levels and underestimation of exposure for diazinon and propoxur on page 50 of the Agency's issue paper should be edited for clarity in the document. As it appears now, the document reads as though these pesticide levels would be more heavily underestimated as compared to chlorpyrifos levels in the same blood samples.

In conclusion, notwithstanding the fact that the three studies are not totally comparable, the Panel found that there were more similarities than discrepancies across them. When considered along with animal data, chlorpyrifos is likely associated with adverse neurodevelopmental outcomes. Although the Mt. Sinai and Berkeley cohorts are less specific than the Columbia study, they support the overall findings of the latter. The Panel supported the statement that exposures to all three AChE-inhibiting insecticides may act in combination to produce the observed effects. The Panel agreed that there may, in fact, be additive effects or effects generated by a mixture of the agents. Although the authors of the Columbia studies have attempted to isolate the effects that would be associated with chlorpyrifos, the Panel noted it is difficult to quantify the contribution of other neurotoxic compounds in such simultaneous exposures. It was also noted by one panelist that other OPs, as well as other chemicals, may play a role in neurotoxic effects in light of a recent study by Eskenazi et al. (2008). The Panel concluded that although a mixture of chemicals may produce the observed effects, this did not rule out chlorpyrifos as playing a role. However, the fact that mixtures were involved in the measurements of all three studies does limit the conclusions relative to the specific role of chlorpyrifos in observed outcomes.

The majority of the Panel agreed that although the exposures in the Columbia study occurred at levels below the U.S. EPA reference dose and likely underestimate true exposures, the results of this study are of concern regarding adverse effects on normal neurodevelopment of children exposed *in utero* to chlorpyrifos. This is particularly true in light of evidence demonstrating that low levels of exposure to toxicants once thought to have adverse neurodevelopmental effects only at high levels (i.e. lead, mercury, and PCBs) are now known to produce significant effects at

lower levels. Organophosphates are known neurologic poisons designed to kill insects. People exposed to high levels of OPs may suffer seizures, coma and death (Etzel, 2003). There was disagreement among the Panel as to what conclusions can be drawn from observed neurodevelopmental outcomes regarding the cholinesterase inhibition mechanism of action. Many other associations have been found between DAP, DMP, DEP and neurodevelopmental and neonatal outcomes that indicate a role for other organophosphates. This can be supported by a cholinesterase inhibition mechanism of action or perhaps some different mechanism. Some panel members believed more strongly that there was likely a different mechanism of action other than acetylcholinesterase inhibition mediating these neurodevelopmental effects.

5. Human Information Available for Risk Assessment:

Ultimately, the Agency will assess potential risk to humans from current exposures to chlorpyrifos. Thus, data in humans provide a valuable tool for considering human outcomes, metabolism, and dose response. Under this context, the Agency has considered the extent to which data in the epidemiology studies and the deliberate dosing studies can be used quantitatively in the chlorpyrifos risk assessment (See Issue Paper, Section 2.3).

a. The epidemiology studies provide important information about potential human outcomes related to the potential effects of OPs on the developing brain. Moreover, they provide data which supports the human relevance of outcomes observed in animal studies. However, at this time, they have not been proposed for use in directly deriving the PoDs or UFs.

Each of the cohorts has been exposed to chlorpyrifos to some extent. However, in addition to chlorpyrifos, each cohort has been exposed to multiple pesticides, including other OPs. Determining the quantitative contribution of chlorpyrifos to the reported outcomes separate from the other OPs is challenging. This determination would be highly uncertain given the current state of the science on the dose response relationships for mechanisms (other than AChE inhibition) leading to effects on the developing brain. As the science evolves in this area, the understanding of TK and TD factors which impact toxicity to the developing brain will improve as will the dose response information in animals. With this improved understanding, the Agency may, in the future, be able to better characterize the linkage between blood or urinary levels of chlorpyrifos and/or its metabolites with health outcomes. At this time, the Agency has used the reported levels of chlorpyrifos and its metabolites simply as markers of exposure without an attempt to estimate actual exposure or dose to the tissues. The Agency is aware of an effort by Drs. Dale Hattis and Robin Whyatt to develop a physiologically-based pharmacokinetic (PBPK) model which includes a placental compartment for assessing tissue dosimetry to the fetus and which accounts for intra-species TK variability. The investigators then plan to use that model to estimate a human PoD from the blood biomarker reported in Whyatt et al (2003). This work has only just begun and will likely take several years. *Please comment on the Agency's conclusion to use the epidemiological studies primarily for purposes of hazard characterization and not for dose response assessment. Please also comment on the scientific support for or against this preliminary conclusion.*

Panel Response

The Panel concurred with the Agency that there are key limitations to the three epidemiological studies and that they should be used primarily for hazard identification. The Panel disagreed on whether the current epidemiological data provide sufficient evidence to suggest that the uncertainty factor for the cholinesterase inhibition endpoint be changed to accommodate the possibility of neurodevelopmental effects from low-level *in utero* exposures of chlorpyrifos. The majority position was that current epidemiological data do not provide sufficient evidence to increase the uncertainty factor for the cholinesterase inhibition to accommodate the possibility of neurodevelopmental effects. However, some panel members felt strongly that the current epidemiological data do provide evidence to indicate that the margin of safety should be increased. The Panel recommends that the Agency conduct a full formal weight of evidence evaluation for causality of the reported associations between exposure to chlorpyrifos and neurodevelopmental outcomes in the existing epidemiological database.

One of the key limitations of the epidemiological studies is that the exposure data were collected at single time point and lack information on the long-term exposure level and duration. A second limitation is that the subjects in two of the cohort studies (conducted by the Mt. Sinai and University of California-Berkeley researchers) had multiple chemical exposures including multiple AChE-inhibiting insecticides (see discussion for previous question). The Columbia cohort study (Whyatt et al., 2002) also had multiple chemical exposures,for example, to the insecticides, DDT (detected at 68% of the samples) and chlordane (detected in 78% of the samples). One panel member added that it may be more difficult to use neurobehavioral endpoints in dose-response assessment as they are qualitative in nature. Another limitation is that neurodevelopmental changes are caused by multiple factors, precluding an accurate dose-response relationship. The Panel disagreed on whether the epidemiological data, and in particular the neurodevelopmental findings, provide sufficient data to support changes to the default uncertainty factor

The Panel stated that data from human studies, in general, are of great value for assessing risks to human health. Besides epidemiological studies and deliberate dosing studies potential sources for data on the effects of chlorpyrifos in humans may come from poison control information on accidental and intentional exposure (decreased since 2001 when indoor uses of chlorpyrifos were voluntarily cancelled) as well as from occupational exposure data from workers in manufacture, formulation, transportation, and application of chlorpyrifos (e.g., pest control operators, pilots, flagmen, pickers).

Confounders are unavoidable in epidemiological studies (see Panel response to Charge Question 4). Confounders regarding the exposure agent may include impurity, mixture (synergism/antagonism with other pesticides or agents), vehicle etc. Confounders regarding the exposure are most commonly related to dose, route, and duration of exposure. Confounders associated with the subjects enrolled in the study may include age, gender, health/disease status, personal environment, and genetic factors such as isoforms and transporters in key toxicological events. Within the chlorpyrifos epidemiological datasets, determining the quantitative contribution of chlorpyrifos in exposures to mixtures may be possible by accounting for differences in chemical potency, pharmacokinetics and possible chemical interactions. Further

resolution of dose-response relationships may also come from the on-going data analysis by the Columbia University study group and any other long-term studies with improved exposure information and more quantitative measure of chlorpyrifos toxicity.

The Panel recommended that the Agency conduct a full weight of evidence evaluation for the neurodevelopmental outcomes. This generally requires a much wider scope of review that encompasses all available data; not only data for the given chemical, but also data for chemicals of similar structure and activities, e.g., OPs. To increase transparency and defensibility of the conclusions concerning the potential role of chlorpyrifos in contributing to neurodevelopmental outcomes, the weight of evidence analysis should specifically address criteria for causality including consistency, and take into account the results not only of the studies considered here but others and the supporting biological data. Such an exercise requires explicit consideration of criteria such as strength, consistency, specificity related to chlorpyrifos or to its anti-cholinesterase effects common to OPs as a whole, dose-response, temporal concordance and biological plausibility in a framework analysis similar to that which is conducted currently for hypothesized modes of action. This allows comparative analysis across assessments of consistency of weight of evidence determinations. The weight of evidence analysis might increase confidence in this case and potentially identify additional relevant analyses to address uncertainties such as the role of other pesticides in the observed associations. To the extent possible, once a hazard is established, an array of dose-response data in both animals and humans in the context of their relative associated uncertainties may also be helpful, recognizing that the magnitude of these uncertainties for the dose-response may generally be greater for animal species, owing to interspecies differences. Outliers need to be excluded, and the relative magnitude of uncertainties explicitly weighted in considering the relative contribution of epidemiological versus other types of data.

The Panel agreed with the Agency that there were limitations in the three epidemiological studies that precluded them from being used to directly derive the PoD or the uncertainty factor.The Columbia University cohort study could be used to determine bounding values for the levels of chlorpyrifos that might cause a measurable effect. In a similar way, data from epidemiological studies can also be used in risk assessment. .The use of a PBPK model would enable estimation of an exposure dose metric for multiple sources of exposure, e.g., air, food, water.

For example, one Panel member estimated the blood concentrations of chlorpyrifos from the PBPK model provided by Dr. Timchalk (Battelle Center for Biological Monitoring and Modeling -Public comments dated Aug 28, 2008, submitted to docket EPA-HQ-OPP-2008-0274)) at the 50th, 75th, 95th percentile at the maximum of the California air concentrations of chlorpyrifos as stated in oral public comments provided by Dr. Sass of the Natural Resources Defense Council (NRDC and representing the Pesticide Action Network – North America (PANNA), September 16, 2008). Based on this analysis, the simulated chlopryrifos blood concentration was approximately $1x10^{-4}$ µmol/L, or 35 ng/L, when calculated using a peak air concentration of 1.34 ug/m^3. Some panel members raised concerns about this computation.[3] In

[3] One panel member provided some additional notes to clarify the vague concerns expressed at the SAP meeting. This information will be useful to the Agency. Review of public comments in the docket (EPA-HQ-OPP-2008-0274-0056,) indicated that the 24-hour average concentrations were below the adult RELs (recommended exposure levels). This finding raises a concern about the calculations made by one panel member. A lag time is needed to

this illustration, potential exposures to a blood concentration of 35 ng/L chlorpyrifos would be nearly 6 times greater in comparison with the measured chlorpyrifos blood concentration, of 6.17 ng/L reported in the Columbia University study. In the Columbia University study, levels of 6.17ng/L chlorpyrifos in blood were associated with neurobehaviornal effects. The Panel urged the Agency to more carefully analyze these current data to determine whether individuals exposed to potentially higher levels of chlorpyrifos in air would show signs of neurodevelopmental effects. In a similar fashion, data from the epidemiological studies could be used in risk assessment as bounding values to evaluate exposure standards, i.e., reference concentrations or doses. Based on the level of analysis that can be performed, Panel members suggested that the Agency might be able to use the cohort data in a more quantitative fashion as part of the risk assessment, such as in the aforementioned boundary setting exercise.

While the potential contribution of the chlorpyrifos PBPK model being developed was recognized, some panelists believed that the Agency should also pursue a simpler PBPK model specifically applicable to the chlorpyrifos data that would be available in a relatively short timeframe. Some panel members urged the Agency to not delay in studying whether the level of chlorpyrifos in air, as noted in the NRDC and PANNA comments, poses a concern for human health.

The Panel recommended the consideration of a fetal or placental compartment in a PBPK model should there be any indication from the paired maternal and cord blood data that chlorpyrifos and its active metabolite(s) are differentially distributed to the fetuses. In addition, the PBPK model can also simulate inter-individual (intraspecies) TK variations as well as inter-species differences. Furthermore, the Panel noted that additional work as a basis to consider more robustly interspecies differences (using central estimates) may be more fruitful over the shorter term.

b. Three deliberate dosing studies in adult (non-pregnant) humans are available which measure AChE activity and urinary levels of chlorpyrifos and/or its metabolites (See Appendix G). The Agency has determined that the deliberate dosing studies in adults are not appropriate for use in PoD or UF derivation in the current proposal. This determination is based on several factors.

- There are experimental laboratory animal data that indicate that the susceptibility of the developing nervous system to chlorpyrifos may be related to cholinergic and noncholinergic mechanisms. Findings in epidemiology studies in children support the animal studies. The human studies do not include the potentially susceptible populations being evaluated in the current effort, namely pregnant woman and children and thus do not consider toxicity endpoints other than AChE inhibition (and related clinical signs).

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reach a steady-state between air concentration of chlorpyrifos and blood levels, so that peak concentrations lasting one hour should not be used for calculating blood concentrations. A time-weighted average must be used instead, which would lead to lower blood levels.

- Nolan et al. (1982) and Griffin et al. (1999) only include a single dose group for a particular route (a study design issue previously criticized by the Human Studies Review Board (HSRB) with respect to other human studies).

- Griffin et al. (1999) report no changes in AChE inhibition and in Kisicki et al. (1999) changes were only seen in one person leading to the characterization of these studies as NOAEL only studies (a type of study not supported by the HSRB for use in risk assessment since absence of an effect (LOAEL) raises questions about whether the investigators were able to detect an effect or that it was possible given the study design).

However, the Agency has determined that the human studies do provide valuable information on correlating oral or dermal exposure with levels of chlorpyrifos and/or TCP in blood and urine. In addition, these studies also provide information on time course of absorption, metabolism, and excretion. Kisicki et al. (1999) also includes PON1 genotype information. Due to the availability of quality TK information, these studies have been used in the past by the Agency to aid in interpreting biomonitoring data. Specifically, results from Nolan et al. (1982) have been used previously by the Agency in estimating (i.e., back-calculating) chlorpyrifos exposure based on urinary levels of TCP. Nolan et al. (1982) has also been used to derive a dermal absorption factor in humans. If the Agency wishes to continue this use of the human studies to assist in characterizing and interpreting epidemiology and biomonitoring data, the studies will be brought to the Agency's HSRB for review of their scientific and ethical conduct. *To assist the Agency in preparing for this review by the HSRB, please comment on the clarity and completeness of the Agency's scientific analysis of the human studies. In particular, please focus on whether the Agency has identified the key scientific issues and whether other information or studies are available that should be considered in formulating the Agency's preliminary conclusion to use these studies for purposes of characterizing and interpreting the epidemiology and biomonitoring data and not for deriving PoDs or UFs.*

Panel Response

The Agency's issue paper clearly laid out EPA's scientific analysis of the human studies and identified desirable components that are missing in these human studies. One panel member noted that to enhance clarity for readers not intimately familiar with EPA procedures, information about human subjects and the conduct of the study should accompany the synopses of the studies provided to the Panel.

Overall, the Panel agreed that the human deliberate dosing studies contain scientifically useful information for risk assessment, but not for directly establishing PoD or uncertainty factors. The following major limitations of these data were noted by the Panel. Collectively, these limitations preclude their direct use for establishing PoDs.

- These studies do not include the potentially susceptible populations (e.g., women of childbearing age, pre- and post natal stages, teenagers and young adults before brain development is complete in the mid-20's).
- The single dose regimen does not provide information to evaluate dose-response relationship and span the range of environmental exposure levels.

- These studies do not provide information on any other toxicological endpoints besides cholinesterase inhibition.
- These studies do not include the inhalation route, a major route of exposure for workers and bystanders alike.

One panelist provided the following information to the Panel from his unpublished study conducted on a small cohort of children (n = 50) examining cholinesterase activity and PON1 genotype as a result of chlorpyrifos exposure (hand wash measurements for exposure). Two children with comparable exposures showed a 100-fold cholinesterase difference at the highest hand rinse concentrations, but different (slow versus fast) polymorphisms in the PON-1 genotype. On the other hand, two other children showed a 10-fold variation in cholinesterase activity even though they had the same PON-1 genotype. These studies used a single dose that may not reflect environmental concentrations and examined only dermal exposure. A significant portion of the observed differences in cholinesterase activity may be due to differences in inhalation or dietary exposures rather than dermal exposure (the route of exposure measured in this study). Given the small population size, use of two isolated comparisons, and the uncertainty associated with exposures, this study does not provide enough information to influence a choice of uncertainty factor. However, these unpublished results do confirm that genetic differences between individuals in a population may alter the exposure to toxic chlorpyrifos metabolites.

In principle, the Panel agreed with the Agency's scientific approach to derive a dermal absorption factor from the human studies. These human studies contain valuable scientific information especially if no other source of data is available. However, the current 3% absorption factor should be further refined by considering the different exposure scenarios. Taken together, these studies provide more robust database than what each single study can offer. They span a wide range of exposures, between 0.06 mg/kg (5 mg for 67-81 kg subjects in the Meuling et al. (2005) study) and 5 mg/kg (subjects B-F in the Nolan et al. (1982) study), different wash-off durations (as short as 4 hours in the Meuling et al. (2005) study), and using different vehicles.

The Panel recommended that if the Agency wishes to use these studies to derive dermal absorption factors for use in risk assessment, then they should adjust for any underestimation using the apparent inverse relationship between dermal absorption and the concentration of exposure (see Nolan et al., 1982 and Meuling et al., 2005.) The Panel also noted that when deriving the dermal absorption from each study, the estimated oral absorption in the same study should be used as a reference point instead of the 100% oral absorption that was assumed in extrapolating data from oral and dermal routes. When necessary, material balance and elimination kinetics from each study should also be accounted for. Other limitations of these studies should be considered as uncertainties, e.g., exposure only on forearms and shorter duration of skin contact than the anticipated human exposures. The Panel was familiar with how the 3% dermal absorption factor used by the Agency (see EPA's 2000 chlorpyrifos risk assessment) was derived based on the Nolan et al. (1982) study. The Agency compared this factor to the 0.03 ratio between the oral LOAEL of 0.3 mg/kg/day from the rat DNT study to the 21-day dermal LOAEL of 10 mg/kg/day in rats. Because it is not likely that the dermal absorption factor derived in humans would closely coincide with the values for rats, and given

that the comparison of toxicity at the LOAELs is crude, the support from this comparison would not be crucial.

The Panel agreed with the Agency scientific analysis that these deliberate dosing studies cannot be used to directly establish a PoD or UFs. The Panel indicated that these data might be used as bounding levels similar to what was suggested by the Panel concerning the data from the epidemiological studies (see Panel response to Question 5(a)).

The Panel appreciated the Agency's scientific analysis to compare the blood levels in the deliberate dosing and epidemiological studies, and considered it critically important to maximally use the information from these studies. These studies could be used for purposes of interpreting (at least crudely) the nature of dose-response in the epidemiological data and as a basis to "bound" the reference doses/concentrations. The Panel encouraged the Agency to consider the use of a PBPK model to widen the application of these bounding data for current or potential human exposures and for the final reference dose or reference concentrations. In addition, these human study data may contribute to an array of the dose-response data in both animals and humans mentioned under question 5a, but in the context of their relative associated uncertainties.

6. Points of Departure (PoD) for Risk Assessment (Issue Paper, Section 2.5):

a. Based on the results of the extensive literature review, the Agency has proposed updated PoDs derived from the laboratory animal studies for extrapolating human risk. The Agency has posed three options for the PoDs for chlorpyrifos in its issue paper. When the Agency derives PoDs for assessing the risk from exposures to pesticides, it needs to consider all relevant routes (oral, dermal, inhalation), durations (ranging from acute to chronic), and all exposed populations (including adult, pregnant women of child bearing age and children).

The first option proposes to use the PoDs which were based on rat RBC and plasma cholinesterase inhibition in the 2000 risk assessment for acute oral exposures and blood AChE for chronic oral exposures. The 2000 risk assessment included a weight of the evidence discussion primarily on adult rat and dog AChE guideline studies and adult data from Zheng et al. (2000). This option would involve application of the no-observed-adverse-effect-levels (NOAELS) for blood AChE inhibition from route specific studies (oral, dermal, inhalation) in rats or dogs to all populations. The acute oral PoD would be 0.5 mg/kg/day and the repeated oral PoD would be 0.03 mg/kg/day. The dermal and inhalation NOAELs would be 5 and 0.1 mg/kg/day, respectively.

The second option proposes to use a value of 0.1 mg/kg/day derived from multiple studies and lifestages. This proposed PoD would be applied to all populations and all durations. The proposed value of 0.1 mg/kg/day was derived using benchmark dose estimates from brain and RBC AChE in young pups (PND1 and 12) following acute dosing and from peripheral (heart) AChE following repeated gestational studies with dams. As such, multiple lifestages are considered in the proposed PoD: pregnant dams, PND1 pups, and PND12 pups. Furthermore, the proposed value is 3 to 10 fold lower than causing effects on the developing brain reported in other laboratory animal studies and thus is expected to be protective for those effects.

The third option is a blend of options 1 and 2. This option proposes to use a value of 0.1 mg/kg/day derived from the acute post-natal rat brain and RBC AChE data for all populations but only for the acute duration for oral exposures. Exposure scenarios involving repeated exposures would use the PoD of 0.03 mg/kg/day from the 2000 risk assessment for oral exposures. The value of 0.03 mg/kg/day was derived for the 2000 risk assessment based on blood AChE from multiple adult rat and dog studies. The lower PoD for repeated exposures in option 3 is proposed to account for potential accumulation of toxicity which can occur following repeating doses of chlorpyrifos in adult studies. *Please comment on the strengths and weaknesses of each proposed approach. Is there another option or a variation of one of the three options that the Agency should consider?*

Panel Response

General comments:
The principal focus of the questions from the Agency on PoD relates to whether PoD used in the 2000 chlorpyrifos risk assessment will be altered given more recent data on neurodevelopmental effects associated with early exposure to chlorpyrifos. Recommendations here are predicated on some general principles in selecting PoDs for risk assessments, including making maximal use of route specific data as a basis to minimize uncertainty, to rely to the extent possible on benchmark doses (BMDs) versus no- or lowest-observed-(adverse)-effect levels (NOAEL or LOAEL) and to take into consideration the sensitivity of all life stages.

Members of the panel also noted that it was difficult to separate fully the discussion of points of departure from that of uncertainty factors so that the content here has relevance to the responses to Question 7 and vice versa (i.e., the adequacy of proposed uncertainty factors is necessarily related to the nature of the point of departure). As a result, comment is also included herein on residual uncertainty associated with proposed or preferred points of departure including that related particularly to potential lack of adequate consideration of all life stages.

The Panel agreed with the Agency's decision not to use the human deliberate dosing studies to establish PoDs due to their limitations, in particular their lack of account of potentially sensitive life stages.

Specific Comments:
There was consensus that of the three options proposed by the Agency, Option 3 is preferred, based on the available data. This is predicated principally on the basis that the proposed PoD for acute exposure takes into account all life stages, is based on benchmark doses which offer an advantage over the NOAEL or LOAEL, and represents the results of several studies, for which results converge around the same value. Based on available data, most of Panel members (a few disagreed) stated that the PoD presented in Option 3 is also believed to be protective for effects on the developing brain, although it is based on cholinergic effects. However, this conclusion is highly uncertain, given the lack of information on the mode of induction of the observed behavioral effects and in light of evidence from *in vivo* and *in vitro* studies that non-cholinergic mode of action(s) are likely involved in the adverse developmental neurotoxicity (DNT) and

behavior endpoints. The Agency is encouraged, therefore, to explicitly address this uncertainty in their additional deliberations in derivation of the reference dose/concentrations.

The Panel suggested that if the Agency wanted to test whether the cholinergic pathways are an appropriate endpoint for the PoD then an appropriate temporal dose-response study using the most sensitive administration method and the most sensitive life-stage comparing DNT/behavior with cholinesterase inhibition would be needed. This would test the hypothesis that cholinergic pathways are an appropriate endpoint for the PoD, even though such a study would likely not elucidate specific non-cholinergic mode of action(s) for DNT/behavior effects. Relevant design would need to take into account differing sensitivity in various species to detect effects on the developing brain.

The neurobehavioral developmental effects seen in the three epidemiological studies supported the retention in Option 3 of the lower PoD for repeated exposures used in EPA's 2000 chlorpyrifos risk assessment to account for potential cumulative effects. For repeated exposures, PoD of 0.03 mg/kg/day is supported by a $BMDL_{10}$ of 0.03 mg/kg/day for RBC AChE inhibition in pregnant dams (see Agency issue paper, p. 18, Table 2). The Panel also recommended that the Agency additionally investigate developing appropriate benchmark doses (BMDs) for the chronic PoD determination.

More detailed comments:

Option 1: Option 1 uses the rat RBC and plasma ChE inhibition in the Agency's 2000 risk assessment (Table 1 of the Agency's issue paper), weight of evidence (WOE) approach based primarily on adult rat and dog AChE guideline studies, NOAELs for various routes and durations of exposure with an acute oral dose of 0.5 mg/kg/day and a repeated oral dose of 0.03 mg/kg/day.

Option 1 is the status quo used in the Agency's 2000 chlorpyrifos risk assessment based on studies conducted solely in adults (blood AChE for acute and chronic oral exposures). The additional data on neurodevelopmental effects would have no impact at this time. As per other options, it has the advantage that it avoids uncertainties associated with interroute extrapolation. However, it is based on no-or lowest-observed-advese-effect-levels rather than benchmark doses; moreover, the dermal and inhalation NOAELs were determined in mature animals and did not include protection for pre- and post-natal sensitivity.

Option 2: Option 2 uses studies that provide data amenable to benchmark dose (BMD) modeling; RBC AChE studies in repeated gestational studies in dams (heart) and acute post-natal pups (brain and RBC AChE) provide BMDs (BMD and $BMDL_{10}$) in the same range of 0.06 – 0.12 mg/kg/day. The Agency proposes to use the weight of evidence approach, a PoD of 0.1 mg/kg/day for oral exposure, all age groups and all durations.

For Option 2, a value of 0.1 mg/kg/day is being proposed for all populations and durations. It is based on benchmark doses/concentrations which offer some advantage over NOAELs/LOAELs though it needs to be recognized that these advantages can be small in relation to relative uncertainties associated with interspecies and intraspecies differences. It does draw from

multiple studies covering several life stages (brain and RBC) AChE in young pups following acute dosing – PNDs 1, 7 and 12 and peripheral AChE from repeated gestational studies with dams; also pups). The Panel was reassured that the values for several studies fall within the same range and the approach offers considerable advantage in drawing maximally on the available data in animals at several life stages. Some panel members stated that 0.3 mg/kg/day was protective because it is 3- to 10-times lower than levels causing effects on the developing brain. The lowest dose tested for learning/memory was 0.3 mg/kg/day so proposed PoD was 3-fold lower and 10-fold lower than lowest dose used in many gestational and post-natal studies which evaluated toxicities other than AChE inhibition. Nevertheless, the degree of protectiveness is uncertain because the dose-response relationship cannot be obtained from these studies reporting substantial effects at 0.3 to 1.0 mg/kg/day. Another consideration is that the 0.1 mg/kg/day dose does not take into account potential for cumulative effects following repeated exposure.

Option 3: Option 3 separates the PoD for acute and repeated exposures. A PoD of 0.1 mg/kg/day was used for acute and short term exposures. For chronic dietary exposure, a value of 0.03 mg/kg/day was used. The value for repeated exposure is based on NOAELs and LOAELs for plasma and RBC ChE inhibition in 5 studies. This option is also supported by the BMD in pregnant dams.

These PoDs (0.1 and 0.03 mg/kg/day) are 3 and 10 times, respectively, lower than the lowest dose tested across all neurodevelopmental studies (0.3 mg/kg/day). The latter dose was administered to dams through gestation and even several postnatal days without observing behavioral effects at any time in the offspring exposed to this dose level (see Agency issue paper, p. 19). The PoDs for acute and repeated exposures might be considered 3-fold and 10-fold higher than the uncertainty factor (TD), respectively, for neurodevelopmental effects. Option 3, is essentially Option 2, but introduces a lower value for repeated exposures from the 2000 assessment to account for potential accumulation of toxicity which can occur following repeated doses. This option was preferred by the Panel, on the basis of potential for different or perhaps multiple actions of chlorpyrifos at repeated low level exposures and results of the recent epidemiological studies. Most of panel members (a few disagreed) stated that this option was protective of effects on the developing brain, although it is based solely on cholinergic effects. However, this conclusion has associated uncertainties; the lack of information on the mode of action in inducing the observed behavioral effects and evidence from *in vivo* and *in vitro* studies that show non-cholinergic modes of action are likely to be involved in the adverse developmental neurotoxicity and behavior endpoints. The Panel encourages EPA to explicitly address these uncertainties when deriving the reference dose/concentrations.

b. Route specific data are preferred because such data accounts for potential differences in absorption, distribution, or metabolism. In the case of chlorpyrifos, dermal and inhalation studies are available which identify NOAELs for these routes in adult rats. With respect to inhalation exposure, there are two nose only studies with vapor chlorpyrifos which provides a NOAEL of 287 ug/m3 or 20 ppb (0.1 mg/kg/day). Similarly, there are two dermal studies which together provide a dermal NOAEL in adult rats of 5 mg/kg/day. These studies do not include pregnant dams, fetuses or post-natal pups and therefore do not consider potentially susceptible populations. In the absence of data in these groups, the Agency will continue to use route specific studies, as appropriate. An alternative for dermal exposure is to use an oral Pod derived

from susceptible populations (as discussed above) with a dermal absorption factor. Specifically, the Agency could use a dermal absorption of 3% from human subjects (Nolan et al., 1982). *Please comment on the strengths and weaknesses associated with use of the adult dermal and inhalation studies in the chlorpyrifos human health risk assessment. The Agency also requests the SAP to provide suggestions on potential toxicity and/or toxic kinetic studies (if any) in pregnant dams, fetuses, and/or post-natal pups which could be conducted to better inform the dermal and inhalation risk assessments.*

Panel Response

The Panel was in general agreement with the Agency's proposed option to use the dermal and inhalation studies as a basis for development of the points of departure for these routes for repeated exposures, as per the 2000 chlorpyrifos risk assessment given the uncertainty associated with route to route extrapolations based on available data, though residual uncertainty concerning sensitive life stages would need to be taken into account. The Panel also noted that the proposed oral, inhalation and dermal PODs are roughly equivalent (0.1 mg/kg/day), taking into account estimated absorption from the Agency's 2000 chlorpyrifos risk assessment based on the Nolan et al. (1982) study. However, proper adjustment of the dermal absorption factor for use in oral to dermal extrapolation is needed when the in-study oral absorption factor is less than 100%. Other considerations for deriving and using the dermal absorption factor in response to question 5(b). Inhalation Pod of 0.1 mg/kg/day should also be adjusted for repeated exposures based on the 5 days per week dosing regimen.

Also, rather than relying on the potentially proposed value of 3% for dermal exposure derived in the deliberate dosing study in humans by Nolan et al. (1982), the Agency is encouraged to review the entire database from all doses and all studies to obtain quantitative understanding of age sensitivity specific to the endpoint of choice, as a basis to apply it across all routes of exposure consistently. Physiologically based pharmacokinetic modeling may be helpful in this context. While the Agency is pursuing development of a PBPK model that includes the fetal compartment, the Panel also encourages consideration of simpler models which might additionally inform comparison of age- and route sensitivity in the near term future. In relation to dermal exposure, the Panel suggested a more relevant species, e.g., minifies, may be helpful.

Detailed Comments:

Given the magnitude of the uncertainties to extrapolate between routes, use of route specific data for inhalation and dermal exposure as a basis for points of departure was supported. However, the residual uncertainty for the lack of relevant data in pregnant dams, fetuses or post-natal pups exposed by these routes needs to be additionally addressed.

Inherent in calculating the absorbed dose from the inhalation NOAEL is the recognition that the dose level should be somewhat comparable between routes for the same systemic endpoint, although rout-specific pharmacokinetic differences can be a significant factor. Some comparisons between dermal or inhalation NOAELs and the oral PoD are presented below.

Dermal NOAEL of 5 mg/kg

Using the Agency's suggested 3% dermal absorption factor derived from the deliberate dosing study in humans of Nolan et al (1982), the absorbed dose for the dermal NOAEL is 0.15 mg/kg/day, not significantly different from the 0.1 mg/kg/day value in Option 2. However, because the extent of oral human absorption estimated from the same study (i.e., Nolan et al., 1982) is 73%, the oral NOAEL of 0.1 mg/kg/day would need to be adjusted by 73% of its original value for this comparison. Thus, the acute dermal NOAEL of 5 mg/kg is approximately 2-fold higher than the oral BMD.

The Panel commented that it is important to recognize that when used in human risk assessment, the magnitude of the absorption factor is dependent on the dermal exposure conditions. For example, in the Nolan et al (1982) study, the absorption factor was 3% at 0.5 mg/kg, but only 1% at 5 mg/kg. The Panel noted that it is important to consider other factors that could affect dermal absorption, e.g., variation of rate of penetration on different skin surfaces, moisture on the skin under the cover of clothing, contact duration before wash off.

Inhalation NOAEL of 0.1 mg/kg/day

According to the Agency's Issue Paper, the NOAEL of 20 ppb from the two inhalation studies in rats resulted in an absorbed dose of 0.1 mg/kg/day, the same value as in Option 2. The value, as calculated, is therefore applicable to acute exposures. For intermediate and chronic inhalation exposure, the absorbed dose of 0.1 mg/kg/day should be adjusted down by a factor of 5/7 to account for the 5 days/week dosing regimen. This is slightly lower than the 0.1 mg/kg/day value as proposed in Option 2.

Overall, the Panel encourages the Agency to review the entire database from all doses and all studies (not just the Nolan et al. (1982) study) to obtain quantitative understanding of age sensitivity specific to the endpoint of choice and apply it across all routes of exposure consistently. Physiologically-based pharmacokinetic modeling may be helpful in this context. While the Agency is pursuing development of a PBPK model that includes the fetal compartment, the Panel also encourages consideration of simpler models which might additionally inform comparison of age- and route sensitivity in the near term future.

7. Extrapolation/Uncertainty Factors (Issue Paper, Section 2.6, Appendix E):

In risk assessment, once PoDs are selected, extrapolation from animals to humans (inter-species) and within human variability is performed. Historically, the Agency has used default 10-fold factors to account for inter- and intra-species extrapolation. More recently, emphasis on the derivation of extrapolation factors from TK and toxicodynamic (TD) data instead of default factors has increased. With the intent of improving the scientific basis for the chlorpyrifos risk assessment, in this issue paper the Agency has considered the availability of current PBPK models, TK, and TD data for chlorpyrifos to use in animal to human and within human extrapolations. Overall, the available PBPK models, although well-developed and supported for non-pregnant adults, do not include calculations for dose during pregnancy (e.g., no placental compartment) and for young children less than 5 years old and thus cannot be used in a quantitative manner for this effort. As such, the Agency has used the 2005 IPCS guidance on Chemical-Specific Adjustment Factors to evaluate available TK and TD data in animals and

humans and to determine the extent to which such data support data-derived or chemical-specific extrapolation factors.

a. Inter-species and Intra-species Toxicodynamic Extrapolation: The Agency has preliminarily concluded that with regard to TD characteristics, due to the likelihood of several possible modes of action of neurodevelopmental toxicity of chlorpyrifos and lack of identifiable and quantifiable key events for MOAs not related to AChE inhibition, the Agency cannot confidently refine the TD component of the animal to human and within human variability factors (i.e., UF_{AD} and UF_{HD}). *Please comment on the scientific support for or against the use of default factors of inter- and intra-species TD extrapolation.*

Panel Response

In response to the question of whether the Agency can refine the toxicodynamic (TD) component with respect to uncertainty of the animal to human and within-human variability, the Panel was in agreement with the Agency that there is not sufficient information to confidently refine the TD component of the animal to human and within human variability factors (i.e., UF_{AD} and UF_{HD}). The Panel agreed that neurodevelopmental effects in the fetus and neonate are important endpoints for chlorpyrifos toxicity, but that there are several possible modes of action of neurodevelopmental toxicity of chlorpyrifos and there is a clear lack of identifiable key events for mode of actions not related to AChE inhibition. One panel member commented that the Agency should also consider AChE inhibition rather than behavioral outcomes, 0.03 mg/kg/day versus 0.3 mg/kg/day, respectively, because of its higher level of sensitivity. Overall, the lack of identified mode of action, lack of correlation of acetylcholinesterase inhibition with possible non-cholinergic mechanisms and developmental outcomes, taken together, do not support a data-derived inter-species and intra-species TD extrapolation. All panel members concurred that the application of uncertainty factors is linked to the effect and dose at the PoD and the issues related to PoD described in Question 6 are relevant here.

Based on consensus around sensitivity discussed in Questions 1 and 2, panel members agreed that animal and human sensitivities are greater for fetuses and neonates, although the current data do not allow for determination of the magnitude of the sensitivity. Until this is known, the Panel recommended application of default factors for the TD uncertainty extrapolation. A few members of the panel suggested that the Agency search the open literature for specific information and perhaps seek additional data regarding xenobiotic chemical effects on the developing nervous system. This information might show variations in response among developing brains due to genetics, time of exposure, pre-existing health status and other factors and support the adequacy of a 3-fold uncertainty factor (UF) for intra-species variation for these endpoints.

In conclusion, until a mode of action is identified and supported (i.e., primary target other than AChE activity) for neurodevelopmental toxicity is identifiable and consistently documented *in vivo*, the default uncertainty factors for the toxicodynamic component should be used. That is, 3-fold and 10-fold, for acute and chronic exposures, respectively, for neurodevelopmental effects.

b. Inter-species and Intra-species Toxicokinetic Extrapolation: As discussed in detail in Appendix E, the Agency evaluated the extent to which data on carboxylesterases, P450s, and paraoxonase (PON1, or A-esterase) support development of DDEFs of inter- and intra- TK extrapolation (i.e., UF_{AK} and UF_{HK}). Based on differences in rat and humans with regard to maturation of metabolic processes, there are uncertainties surrounding appropriate metabolic parameters for animal to human extrapolation of juveniles. This uncertainty in combination with limited data precludes the development of a DDEF for inter-species TK extrapolation (i.e., UF_{AK}). Thus, the Agency proposes to apply the default 3X for UF_{AK}. Data on carboxylesterases are not sufficiently robust for intra-species TK extrapolation (i.e., UF_{HK}). Data on P450s are complicated by multiple enzymes each with its own maturation profile. Others have evaluated the P450 literature for use in derivation of child specific UFs with poor success (Ginsberg et al., 2004a). *Please comment on the scientific support for or against the use of default factors of inter-species TK extrapolation. Please further comment on the Agency's preliminary conclusions on the utility of carboxylesterase and P450 data to refine the intra-species extrapolation factor.*

Panel Response

The Panel concurred with the Agency that there is scientific support to use a default factor for inter-species TK extrapolation. The Panel agreed with EPA's decision to apply the default UF_{AK}. The Panel unanimously encouraged the Agency to continue to consider the importance of all the enzymes in chlorpyrifos' metabolic pathway; the P450s, the carboxylesterases and PON1 as chlorpyrifos' toxicity appears to be dependent on the active metabolite, chlorpyrifos-oxon. The PON1 activity towards the chlorpyrifos-oxon, i.e., chlorpyrifos-oxonase is involved in the catalytic activity of the enzyme, but displays genetic polymorphism. There are important differences in the rates of hydrolysis (enzyme activity) across genotypes (192RR > 192QR > 192QQ), with PON1 R192 allele hydrolyzing chlorpyrifos with a higher catalytic efficiency than PON1 Q192 allele (Furlong, 2007). However, PON1 activity and polymorphism at position 192 (referred to as PON1 status) is not the only determinant of chlorpyrifos toxicity, since other metabolic pathways may modulate potential deficits in detoxication capacity. Thus, at least theoretically, the level of chlorpyrifos-oxon present in serum will protect against the circulating active metabolite. However, at low dose exposures, most active metabolites generated in the liver are also detoxified immediately in the liver, either catalytically (PON1 and CYP450) or stoichiometrically (carboxylesterase and butyrylcholinesterase, BChE)). One panel member commented that research by Sogorb et al. (2008) indicated that even if a small fraction of chlorpyrifos-oxon leaks from the liver without being detoxified, it is rapidly bound and inactivated by serum BChE and also by albumin, demonstrating the hydrolytic activities of BChE against chlorpyrifos oxons at concentrations lower than 0.5 μM (Sogorb et al, 2008). Mattsson et al. (2000) detected chlorpyrifos-oxon in blood of fetuses at a concentration of 1 ng/g (that equals to 0.003 μM) only after the administration of high doses of chlorpyrifos (5mg/kg/day) by gavage to dams at gestational day 20. Such a low concentration of chlorpyrifos-oxon can be efficiently detoxified by serum albumin regardless of PON1. On the other hand, no chlorpyrifos-oxon was detected in blood samples from human volunteers exposed to single oral doses of chlorpyrifos ranging from 0 to 2 mg/kg (Kisicki et al., 1999). Even in the case that some chlorpyrifos-oxon escapes from binding to blood proteins or enzymes, and from being hydrolyzed by PON1 in the blood; the possibility that it reaches the brain is scarce since chlorpyrifos-oxon is a highly reactive metabolite and undergoes spontaneous hydrolysis (its half-

life is less than one minute in blood, see Brzak et al., 1998), so there is not enough time to complete that journey intact. Thus, PON1 probably fails to play a relevant role at low environmental concentrations (nM to low μM). The maximum daily intake of chlorpyrifos is 0.11 μg/kg/day for infants and 0.24 μg/kg/day for toddlers (see review by Eaton et al., 2008; Table 21). At middle and higher concentrations, chlorpyrifos-oxon may be formed in the brain because of the low desulfuration activity of brain microsomes and mitochondria (Chambers and Chambers, 1989). However, the brain has no PON1 activity and, therefore, the oxon produced "*in situ*" interacts with its target molecules unless it undergoes stoichiometric binding to BChE or other proteins acting as "scavengers".

Lassiter at al. (1998) demonstrated that maternal blood chlorpyrifos-oxonase activity is variable before birth, and the placenta has about 20% the activity of the liver, which is consistent during late pregnancy. Fetal liver exhibited minimal activity during gestation, but increased after delivery. Mortensen et al. (1996) showed that maternal and fetal brains had no detectable chlorpyrifos-oxonase activity (Mortensen et al, 1996). Although, the Km of chlorpyrifos-oxonase activity is high (Km=210-380 μM) , chlorpyrifos-oxonase in plasma and liver from adult animals has be shown to be capable of hydrolyzing physiologically relevant concentrations of chlorpyrifos-oxon (nM to low μM) whereas the young animal has, in turn, less capacity to detoxify such concentrations of chlorpyrifos-oxon via chlorpyrifos-oxonase (Mortensen et al., 1996).

One panel member pointed out that the lack of tissue specific data for both species under similar dose and timing considerations e.g., liver, is a major gap in the ability to extrapolate from animal data to humans. Others pointed out that there are human data on human liver carboxylesterases during maturation, but these data were not considered "sufficiently robust" to be useful in TK extrapolations.

c. Intra-species Toxicokinetic Extrapolation (Within Human Variability): There are extensive data on PON1 from many populations worldwide and for different age groups. Using these data, the Agency has performed a preliminary analysis for within TK human variability for PON1 activity. These calculations were done in a manner consistent with the IPSC CSAF guidance. The calculations show that the largest variability in PON1 activity is between newborns and their mothers and is thus likely related to age-dependant maturation.

There is some debate as to the extent to which PON1 status plays a role in toxicity at low environmental concentrations. Some have suggested that significant amount of OP (active oxons, not the parent components) must be present in the blood or brain for PON1 activity to affect toxicity based on generally low affinity (Km, 0.1-10 mM). Others believe that PON1 status is a key determinant in chlorpyrifos toxicity. The Agency has evaluated the available *in vivo* and *in vitro* data from animals and humans relevant to this issue. The Agency has preliminarily concluded that the available data suggest that PON1 status can not be ruled out as a determinant in chlorpyrifos toxicity, particularly for the fetus or young child. However, uncertainties remain, particularly regarding the degree to which other metabolic pathways modulate potential deficits in detoxication capacity. *Please comment on the science which does and does not support PON1 status as a determinant in toxicity at low environmental concentrations.*

Panel Response

The Panel unanimously concurred with the Agency, based on the data, that PON1 status cannot be ruled out as a determinant in chlorpyrifos toxicity, particularly for the fetus or young child. Several panel members stated that actual human exposures vary between bystander and applicators (and everyone in-between) and it is difficult to define what exactly a low level of exposure is for humans. Some in the Panel suggested that the PON1 dataset could be used to address data uncertainty. However, many in the Panel cautioned that these data should not be used out of context until the rate limiting step is identified based on a PBPK model. Such a model does not yet exist, but is under development. These panelists believed that the use of the PON1 data set without such information would be a misuse of the IPCS guidance for determining a CSAF (chemical-specific adjustment factor).

Several members of the Panel encouraged the Agency to obtain an independent peer review for the Timchalk PBPK model in order to assess the overall impact of PON1 relative to the toxicity of a range of CPF, including what may be the choice of dose metric for such comparison, e.g., BChE inhibition or blood or tissue level chlorpyrifos, chlorpyrifos-oxon, or TCP.

One panel member reminded the panel that there have been no direct experiments looking at what we might consider to be low dose exposures in animals in reference to the involvement of PON1. This panel member discussed two published model systems that might be of use to answer this question: the PON1 knockout (ko) and the PON1-Q192 humanized mice. In the PON1 ko model if there any effects of PON1 at low dose exposures then the panelist suggested that these effects would be manifested in this model. The Shih et al. (1998) paper shows that the presence of PON1 prevents the inhibition of brain acetylcholinesterase at a dose of chlorpyrifos of 300 mg/kg (see discussion in Appendix A (Metabolism), p. 26, Figure 8, Panel E of the figure reprinted from Shih et al., 1998). However, the magnitude of the difference between the wild type and the PON1 ko mice is on further reflection not that great. By this panel member's calculations the loss of acetylcholinesterase activity is ~60% in the ko mice and 40% in wt mice. This would then suggest that at lower chlorpyrifos exposures and in people with varying levels of PON1 activity (and not a complete lack of activity) there might be very little effect.

In the second model system, the PON1 humanized mouse model, if there are higher chlorpyrifos-oxon doses then there would be a significant difference in the inhibition of brain acetylcholinesterase in mice with either the R192 or Q192 polymorphism (see Appendix E, figure 4, p21). At low doses, it may not be so clear, particularly as there are no data for the R192 mice at the lowest dose used. The question then remains whether this results in a functional effect that can be identified. The original paper by Cole et al. (2005) also presents morbidity data in the same mice following these chlorpyrifos-oxon doses (Panel C of the same figure). Again there would appear to be missing data (namely there does not appear to be any data for the R192 mice below 1.5 mg/kg. However at 1.5 mg/kg there is no morbidity in the R-192 mice and with the Q192 there is no morbidity at doses below 1.0 mg/kg. Again this would suggest at "low" doses there might not be significant differences between the R/Q forms in preventing chlorpyrifos-oxon induced toxicity. This panel member concluded that further data are needed

but that current evidence is suggestive that PON1 is not a determinant of toxicity at low environmental concentrations.

d. The Agency's PON1 calculations have focused on the PON-192Q/R polymorphism based largely on the extensive data available. No calculations have so far been performed on other genotypes. *Please comment on scientific support for or against focusing on the PON-192Q/R polymorphism.*

Panel Response

The Panel concluded that the use of the PON192Q/R polymorphism in the Agency's calculations for PON1 intra-human variability is appropriate. This conclusion was based on the available data (Furlong et al., 2005; Brophy et al., 2001). However, the analysis of the individual contribution of each polymorphism in the promoter region on serum paraoxonase activity/levels is complicated because of the pronounced linkage disequilibrium between the promoter and the coding region polymorphisms (Draganov and La Du, 2004). Thus, it cannot be ruled out that the variation observed for one of the promoter polymorphisms (PON1 C-108T) may be in part due to linkage disequilibrium with the PON1 Q192R polymorphism (Brophy et al., 2001; Sirivarasaia et al., 2007). Other studies have actually excluded a significant effect of this polymorphism on serum PON1 (Phuntuwate et al., 2005).

The 2005 IPCS CSAF guidelines state that the sensitive subgroup should be evaluated as a distinct population (bi-modal distribution). The data from Holland et al. (2006) on mothers and infants are the two important sensitive subgroups (infants and those with the Q/R polymorphism). Intra-individual variability could be underestimated because this population is of similar ethnic descent (i.e., Latina mothers and infants, primarily of Mexican descent).

In the case of PON1, the sensitive group is the neonates with the QQ genotype. They should be (and are) compared back to the mothers of the QR genotype. If data from other studies exist (could be done now for adults with the data from Brzak et al. (1998), it would be appropriate to combine geometric means and geometric standard deviations by a weighted average approach. This would provide a more diverse group to be considered as the reference group (i.e. QR genotype, Latina mothers, white women and white men).

Several panel members made a strong recommendation that the Agency gather and analyze the data (*in vivo* & *in vitro*) from animal and human enzyme kinetics to look more closely at the PON-192Q/R polymorphism. Once a PBPK model is developed then the impact of activating and deactivating pathways and potentially rate-limiting components will be identified and information regarding PON1 Q/R polymorphism could be put into context. Such information, if attainable, might be used to modify the Agency's proposed approach.

e. The Agency's calculations conducted on PON1 activity follow the 2005 IPCS CSAF guidance for developing intra-species extrapolation factors for TK. The preliminary analysis suggests that within human variability is larger than the default 3X when newborns and adults are considered together. Specifically a value of 12X has been calculated for chlorpyrifos-oxonase activity. The Agency has proposed two options for these calculations: 1) use the value calculated

for chlorpyrifos-oxonase derived from newborn and mother values in Holland et al. (2006) and 2) use the default factor of 3X. *Please comment on the strengths and weaknesses of each proposed approach. Please include comments on the statistical approach used in the analysis as a component of your response.*

Panel Response

The panel reviewed the two approaches and the majority of the panel concluded that the currently available data indicate that the UF_{HK} should be the default value of 3-fold when newborns and adults are considered together. On the other hand, a few panel members concluded that the currently available data indicate that the overall UF (UF_{HK} x UF_{HD}) should be greater than 10 based on PON1 data (not other enzymes involved in the metabolism of chlorpyrifos) and the potential for developmental neurotoxicity of chlorpyrifos as noted in the epidemiological studies (Rauh et al). These members preferred the UF_{HK} of 12-fold, rather than the default of 3-fold, because these were the only two choices proposed. However, none of the panel members endorsed the CSAF approach used by the Agency to identify the factor of "12-fold" calculated based on chlorpyrifos-oxonase and encouraged the Agency to pursue other approaches based on the mode of action (see 2005 IPCS CSAF guidelines). Many panel members did not endorse the 12-fold uncertainty factor because it focused strictly on PON1 as the focal point of toxicity, i.e., focused on only one enzyme out of a very complex system. Similarly, one panel member stated that use of the 12-fold uncertainty factor would be a complete misuse of the 2005 IPCS CSAF guidelines in which there is a clear need to define uncertainty factors based on the endpoint linked to the mode of action, neither of which is defined in this case.

Members of the panel expressed concern about lack of data for non-plasma enzymes, no biological reason or data given support the notion that maturational differences in one tissue relate to another. The data on the relative contributions of P450, carboxylesterases and butyrylcholinesterase to the ultimate concentration of oxon in the target tissue(s) is insufficient to derive a data-derived uncertainty factor, as EPA has concluded. Members of the panel stated that the information on PON1 polymorphisms should not be used as the sole factor in a data-derived uncertainty factor for two main reasons: 1) it is only one enzyme in a complex pathway, and is subsequent to the bioactivation reaction; therefore, it can only function on the amount of bioactivation product (i.e., chlorpyrifos-oxon) that is delivered to it by CYP450): and 2) the genotype of PON1 alone is insufficient to predict vulnerability because the overall level of enzyme activity is ultimately what determines detoxication potential from that pathway; thus, it is better to use PON1 status because it provides information regarding both PON1 genotype and activity. Some of the data from laboratory animal studies in PON knockout animals are using an unrealistic animal model and frequently very high dose levels, and do not reflect what might happen in humans.

When all of the kinetic parameters including those for PON1 genotype were placed into the Timchalk and Poet PBPK model, PON1 genotype only influenced chlorpyrifos-oxon concentration at the higher doses, but did not appreciably influence chlorpyrifos-oxon levels at the lower more environmentally relevant levels. Therefore several panel members concluded that

it is not reasonable to base an uncertainty factor on PON1 genotype alone and the suggested factor of 12 is unreasonable.

One panelist stated the EPA-derived CSAF for UF_{HK} underestimated the UF HK and that a statistical approach is recommended to correct for this problem. This panelist suggested an alternative approach to calculate UF_{HK} would be to use the 99th (or 1st) percentile rather than the 95th (or 5th) to protect a greater portion of the population, and correct for the bias in estimating the GSD from the arithmetic summary statistics. The justification and approach to correcting the bias introduced when using arithmetic summary statistics is available in a manuscript currently in preparation by this panelist (Lynch et al., (2008)). An example of how this method can be applied is provided in Table 2 below. The Panel recognized that the justification for using the 99th rather than the 95th percentile is a policy decision.

Table 2: Comparison of UFHK Factors (Lynch et al. 2008)

	Default	50^{th} %tile QR Mothers/ 5^{th}%tile QQ neonates	50^{th} %tile QR Mothers/ 1st%tile QQ neonates
Current Method	3	11.6	16.5
Corrected Method	3	12.2	17.6

Additional Recommendations Regarding UFs

Following discussion of Question #7 (a-e), several panel members recommended the following:

1. Establish the UFs for chlorpyrifos based on the PoD as given in any of the three options presented in question 6, and the concern for developmental neurotoxicity as indicated in the epidemiological studies.

2. Because the Agency's choices of PoD are all based on cholinesterase inhibition (AChE inhibition), the Panel stated that it would be most appropriate that the UFs be established for this endpoint and not for the developmental neurotoxicity endpoints. Separate from establishing a set of UF for AChE inhibition endpoints, the Agency is encouraged to establish an UF specific for addressing the concerns for developmental neurotoxicity based on the evidence in the epidemiological studies.

3. Given the complexity of inter-relationship among all the TK and TD components leading to AChE inhibition, it is difficult to derive individual UFs (i.e., UF_{AD}, UF_{HD}, UF_{AT}, UF_{HT}) in a distinct and separate manner. The rate-limiting step in the formation/removal of the oxon is necessary to adequately predict the concentration of active metabolite in target tissues at environmental doses. Because of the complexity of the pathway, focusing on one enzyme that is likely not the rate-limiting step won't allow for the best, nor entire picture, of intra-

human variability estimates. Instead, the Agency is encouraged to pursue the use of a "simple" PBPK model that can integrate all key toxicokinetic and toxicodynamic factors and evaluate their contributions to the overall outcome of endpoint (e.g., AChE inhibition) or related dose metric of interest (e.g., profiles of chlorpyrifos, chlorpyrifos-oxon, and the chlorpyrifos-specific urinary metabolite, TCP) at various chlorpyrifos exposure scenarios and for various life stages. The Panel noted that the Agency has made substantial progress in exploring the use of PBPK model with a few remaining pieces yet to be completed for a preliminary review (Timchalk, 2008; comments to the docket, EPA-HQ-OPP-2008-0274). The Agency is encouraged to bring this model to a stage ready for independent peer review in the very near future, without waiting for the newly funded model. Such a model is expected to be revised as needed, and as new information becomes available in the future.

Data-derived extrapolation factors (i.e., uncertainty factors, UFs) are predicated on at least some understanding of the mode of action for the critical effect. DDEFs (also called, CSAFs) need to be based on quantitative data. At a minimum, we need to understand the rate-limiting step in delivery to the target tissue and the active metabolite. One panel member suggested this approach should be done particularly in two tissues, liver (before any amount of chlorpyrifos-oxon reaches the blood stream) and brain. There is uncertainty about whether the effect is mediated by the AChE inhibition pathway and even if so, a PBPK model is needed to characterize the relevant activation and detoxication at relevant concentrations or doses, as a basis to replace defaults.

One panel member stated that the replacement of default uncertainty factors is more often achievable for interspecies differences because of the attempt to capture ratios of central estimates for the relevant animal species and humans vs. population variability in the human population, which requires considerably more data. This panel member emphasized how misleading it is use the PON 1 activity data as a basis for replacement of default, in this case, given that it is an inappropriate surrogate for human variability.

Several panel members recommended "bounding" the reference doses developed on the basis of the animal studies, taking into account the dose-response information from the human deliberate dosing and epidemiological studies, but not in relation to the points of departure. This is because the dose-response information needs to be interpreted in the context of what we know about interspecies and intraspecies differences, such that the bounding exercise using animal studies can be tested using human data.

One panel member expressed overall concern about using PBPK models due to lack of transparency. This panelist believed that there is a blurring of inputs/outputs based on what is known (i.e., data are missing) versus what is predicted. Only when the predictions of the models can be verified with empirical data should they be used.

In the interim, the majority of the Panel recommended that the Agency apply a default UF of 100 to the PoD based on AChE inhibition; i.e., 10-fold for interspecies, 10-fold for inter-individual variation of sensitivity (i.e., intra-species differences). Some panel members recommended that the Agency should consider the use of additional UFs to address the concerns for developmental neurotoxicity as indicated in epidemiological studies. The

default UF may be modified in the future when information on the mode of action for the developmental neurotoxicity becomes available.

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APPENDIX 1: Summary of Neurobehavioral Effects of Developmental Exposure to Chlorpyrifos in Rodents: 2000-2008

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	Pup Toxicity	AChEI		Task	Age at Test	Results
1 Maurissen et al, 2000	Rat (S-D)	GD6 - LD 10	0.0	Gavage (corn oil)	5 mg/kg: ↓ Wgth Gain (End of Gest.)	5 mg/kg: Delay in vaginal opening	GD 20 Dams: 1 & 5 mg/kg	↓ Brain, Plasma, RBC	Delayed Spatial Alternation: Repeated Testing		
			0.3		Dams: High dose: muscle twitches, hypernea, hyperactivity Controls & low dose:sporatic hyperactivity	Decreased BW: Males PND 22-65	0.3 mg/kg	↓ Plasma	7 x 12	PND 23-24	Acquisition: No Rx Effect
			1.0 5.0						4 x 12 6 x 10 6 x 6	PND 24 PND 72-83 PND 85-90	Retention: No Rx Effect Acquisition: No Rx Effect Retention: No Rx Effect
								Apparatus:Wire bottom cages	Motor Activity (Habituation) (Beam Breaks) 1 Hr No Gender Diff	PND 13 PND 17 PND 21 PND 60	No Rx No Rx No Rx No Rx
									Auditory Startle 50 trls: 120dB	PND 22 PND 61	↑ lat to peak response (p=.03) [overall effect] No Rx effects
Slotkin's Group											
2 Levin et al, 2002	Rat (S-D)	GD 17-20 Pk period neurogenesis	DMSO(1 ml/kg) 1.0 5.0	sc (DMSO)	No change in maternal care taking		N.D.		Spontaneous Alternation A. (T-Maze)		
									Percent Alternation Latency to Choice 5 x 5	4-6 wks 1 & 5 mg/kg	No Rx Effect ↑ during Trails 1 and 2 (Hyperactivity)
									B. Fig 8 Locomot. Act 3 x 1 hr Motor Activity Habituation	4 - 6 wks No Rx Effect 1 & 5 mg/kg	 ↓ (Females)
									C. RAM (16 arms) 18 sessions Working Memory Errors Reference Memory Errs. Response Latencies	8-13wks 1 mg/kg 1mg/kg 1 & 5 mg/kg	 ↑ (Females) ↑ (Females) ↑
									Drug Challenge(RAM) Scopolamine Mecamylamine	14-17wks ↑ in WME & RME No Rx Effect	 (Difference in WME bet 1 & 5 mg/kg gps)

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	Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	Pup Toxicity	AChEI
3	Icenogle et al, 2004	Rat (S-D)	GD 9-12 Form/Clos Neural Tube	1.0 5.0	sc (DMSO)	None	No effects of growth/viability	N.D.

	Task	Age at Test	Results		
	Spontaneous Alternation				
A.	**(T-Maze)**	4-8 Wks			
	Percent Alternation	Not Reported			
	Latency to Choice	CPF 1 & 5	↓ (Trial 1)		
B.	**Fig 8 Locomot. Act**	4-8 Wks PND 1-4, & PND			
	SMA	11-14			
	Habituation	CPF 5	↑		
C.	**RAM (16 arms)**	8-13 wks			
	18 sessions				
	Working Memory Errors	CPF 5	↑ (Early Trials)		
	Reference Memory Errs.	CPF 5	↑ (Early Trials)		
	Drug Challenge(RAM)	14-17wks			
	Scopolamine	CPF 5	Attenuated response		
	Mecamylamine		No Effects		
D	**Startle/ PPI**		No Rx Effects		
	65 Db Bckg: 110dB SS				
	PP 68,71,77dB				
E	**Elevated Plus Maze**				
	Center crosses	CPF 5	↑		

	Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	AChEI
4	Levin et al, 2001	Rat (S-D)	PND 1-4	1.0	sc (DMSO)	None	N.D.
			PND 11-14	5.0	sc (DMSO)		

	Task	Age at Test	Results		
	Spontaneous Alternation				
A.	**(T-Maze)**	4-6 Wks PND 1-4, PND 11-			
	Percent Alternation	14	No Rx Effect		
	Latency to Choice	PND 1-4, & PND 11-14	↓ (Males)		
B.	**Fig 8 Locomot. Act**	4-6 Wks PND 1-4, & PND			
	SMA	11-14	No Rx Effect		
	Habituation	PND 11-14	↓ (For first session)		
C.	**RAM (16 arms)**	8-13 wks			
	18 sessions				
	Working Memory Errors	PND 1-4(CPF1)	↓ (Fems)	↑ Males	
	Reference Memory Errs.	PND 1-4	↓ (Fems)	↑ Males	Initial phase
		PND 11-14	No Rx Effect		
	Drug Challenge(RAM)	14-17wks			
	Scopolamine	No scop impair (CPF1 Fems) WME			
	Mecamylamine	No RX effect			

ATTACHMENT C

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal/Pup Toxicity	AChEI	Task	Age at Test	Results
5 Aldridge et al, 2005	Rat (S-D)	PND 1-4	1.0	sc (DMSO)	None		A. Elevated Plus Maze	PND 52-53	
							Time in Open Arms		↑ (Males)
							Center crosses		↑ (Males)
							B Choc Milk Consumpt	PND 54	
							2 choice/ 2 hrs	Preference	↓(M & F)
							C RAM	PND 64-97	
							Working Memory Errors		↓ (Fems) ↑ Males
							Reference Memory Errs.		↓ (Fems) ↑ Males
							Drug Challenge(RAM)	16-17wks	
							Ketaserin	WME	↑ (M & F)
							5-HT2 antagonist	RME	↑ (M & F)
						Acute Study			
						2 hr aft first Rx PND 1	Reflexes		
6 Dam et al, 2000	Rat (S-D)	PND 1-4	1.0	sc (DMSO)	None		↓60% (CBM, BS, FBr), Rec. 4 hr	Righting Reflex	PND 3-4
						1 mg/kg Males	↓20% (CBM, BS, FBr)	Time to Right	CPF 1 ↑ Females
		PND 11-14	5.0	sc (DMSO)		1 mg/kg Females			
						2 hr aft CPF Rx PND11	Negative Geotaxis	PND 5-8	
							↓20% (CBM, BS, FBr) M & F.	Pups meeting criteria	CPF 1 ↓ Females
						5 mg/kg		Open Field	PND 21
									SMA ↓ Males
									Rearing ↓ Males
									PND 30
									SMA ↓ Males
									Rearing ↓ Males

Calamandrei's Group

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal/Pup Toxicity	AChEI	Task	Age at Test	Results
						20-40% I serum AchE (at 24 but not 48 hrs after			
7 Venerosi et al, 2006	CD-1 Mice	GD 15-18 and PND 11-14	0.3.0, or 6.0 & 0, 1.0, 3.0	Gavage (peanut oil)	None	GD 19: CPY 3 & 6	Rx) Social Recognition Test	4 months	↑ Rate of USV
							Retest Same Partner	GD CPF6	↑ Rate of USV
							Retest Different Partner.	GD CPF6	↑ Rate of USV
							Social Investigation Test	GD CPF6	↑ Social investigation

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal/Pup Toxicity	AChEI	Task	Age at Test	Results
8 Venerosi et al, 2008	CD-1 Mice	PND 11-14	3.0	sc (peanut oil)	Maternal Behavior: PND 1: Decreased latencies to first lick PND 2:Light-Dark Test: Decreased lat. to enter light comp PND 7: Dec aggression; Inc investigation of intruder PND 1-7: Nest Building: Increases Latencies PND7: Maternal Agression		Social Approach Social Novelty	PND 45	↓ Social contact (Session 1) No Rx Effect
9 Ricceri et al, 2006	CD-1 Mice	GD 15-18 and PND 11-14	0,3.0, or 6.0 & 0, 1.0, 3.0	Gavage (peanut oil) sc (peanut oil)	None	Pups GD 19: Dose dependent decrease(Serum) No Rx effect: Brain PND 15: Same as GD 19	Open Field (SMA) Isolation- induced aggression Maternal Behavior Elevated Plus Maze	PND 70(Males) PND 75-80 (Males) PND 90 (Fems) PND 120 (M&F) Time in Open arms Head Dipping	↑ CPF 6 ↑ Dur Attack (GD CPF3) ↑ Off posture (CPF6) Def post: No Rx effect ↑licking dura & crouching (PN CPF 1&3) ↓Licking freq (PN CPF 1&3) ↑ PN CPF3 Fems ↓ PNCPF 3 Males
10 Ricceri et al, 2003	CD-1 Mice	PND 1-4, or PND 11-14 PND 32-35	1.0, 3.0 1.0, 3.0 1.0, 3.0	sc (DMSO)	No overt signs of chlinergic intoxication at either age	PND 4: CPF 1 & CPF 3 Non dose-depend. AChei (total & soluble AChE) 1hr but not at 4 & 24 hrs	**Neonates** USV (Social) Homing (Olfactory) Locomotor Activity **Adolescents** Novelty Seeking Social Interaction Agonist Behavior **Adult** Passive Aviodance	PND 5,8,11 PND 10 PND 25 PND 35 PND 45 PND 45 Males PND 60 Males	No Rx Effect: CPF 1-4 or CPF 11-14 No Rx Effect: CPF 1-4 or CPF 11-14 No Rx Effect: CPF 1-4 or CPF 11-14 CPF 1-4 ↑ Activity No Rx effect: novelty seeking CPF 11-14 ↑ Activity No Rx effect: novelty seeking CPF 1-4 ↓ Self Grooming CPF 11-14 ↑ Soliciting resp. CPF 1-4 ↑ Agonistic resp. CPF 11-14 ↑ Agonistic resp. CPF 1-4 No Rx effect CPF 11-14 No Rx effect

Individual Studies

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	Pup Toxicity	AChEI	Task	Age at Test	Results
11 Moser, 2000	Rat (Long-Evans)	PND 17 PND 27 PND 70	4, 10, 20 mg/kg 10, 25, 50 mg/kg 10, 50, 100 mg/kg	Oral gavage (corn oil)	N/A	Weight losss in all groups	N.D.	**FOB (Functional Observational Battery)** Neuromuscular Activity/reactivity Sensorimotor Physiological	PND 70 PND 17 PND 27 PND 70 PND 17 PND 27 PND 70 PND 17 PND 27 PND 70	Forelimb Grip: ↑ (F) (CPF 50); Foot Splay: ↑ (F)(CPF 100) Total Activity / Rearing / Arousal / Handl. React ↓ (M & F) (CPF 10) / ↓(F) (CPF 20) ↓ (M & F) (CPF 25) / ↓ (M & F) (CPF 25) / ↓(M & F) (CPF 50/25) / ↓(F) (CPF 50) ↓(M & F) (CPF 10/50) / ↓ (M & F) (CPF 50) / ↓(M & F) (CPF 50/100) / ↓(M) (CPF 100) Tail-pinch Response / Click Response / Touch Response ↓ (M & F) (CPF 10/4) / ↓ (M) (CPF 20) ↓ (M & F) (CPF 50/25) / ↓ (M) (CPF 25) / ↓(F) (CPF 25) ↓ (M & F) (CPF 10/4) / ↓ (M & F) (CPF 50) / ↓ (M) (CPF 50) Weight / Temperature ↓(F) (CPF 25) / ↓ (M & F) (CPF 25) ↓ (M & F) (CPF 50) / ↓ (M & F) (CPF 50)

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Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	Pup Toxicity	AChEI		Task	Age at Test	Results
12 Carr et al, 2001	Rat (S-D)	PND 1-21	LOW: 3/mg/kg (Every 2nd Day)	Gavage (corn oil)	N/A	↓ Body Weight, PND 13-21 (HIGH Dose)	PND 6, 10, 16 20,25, 30: ~ 50% Inhibition in forebrain and hindbrain in all 3 Rx groups Skeletal Muscle: ChEI on PND 6 (Low Dose); PND 10, 16(Low & Med Doses) Lung: ChEI on PND 6 (Low Dose); PND 10, 16, 20 (Low & Med Doses)	Heart: ChEI (Low Dose: PND 6,10,16,20). (Med Dose: PND 10, 16, 20, 25). (High Dose: PND 16, 20, 25). Diaphragm: (Low Dose: PND 6,10,16). (Med Dose: PND 10, 16, 20). (High Dose: PND 16, 20, 25). Serum AChEI: (Low Dose: PND 6,10). (Med Dose: PND 10, 16, 20). (High Dose: PND 16, 20).	Open Field Activity	PND 10-20	No Rx Effect
		PND 1-5, 7-21	MED: 3 mg/kg/day, 6 mg/kg/day							PND 25 & 30	↓ High & Med Doses M & F(Dose-related)
		PND 1-5, 7-13, 15-21	HIGH: 3 mg/kg/day, 6 mg/kg/day, 12 mg/kg/day								

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	Pup Toxicity	AChEI		Task	Age at Test	Results
13 Jett et al, 2001	Rat (Long-Evans)	PND 7, 11, 15	0.3 or 7.0	s.c. (peanut oil)	N/A	No overt signs of cholinergic toxicity	Cortex, hippocampus, cerebellum: PND 7,8 16, 28 (Pre/Post)	No ChEI due to CPF Rx	Morris water maze	PND 24-28	
		PND 22, 26	0.3 or 7.0							Acquisition	Preweaning CPF 7.0: ↑escape latency Day 5
							Mucarinic Receptor Binding	No Rx Effects			↓ percentage of rats finding platform by Day 5
										Probe	Preweaning CPF 7.0:↓ time in platform training quadrant
										PND 24-28	
										Acquisition	Postweaning CPF 0.3 & 7.0: ↑escape latency Day 5 ↓percentage of rats finding platform by Day 5
										Probe	Postweaning CPF 0.3 & 7.0: ↓ time in platform training quadrant
14 Abou-Donia et al, 2006	Rat (S-D)	GD 4-20	1.0	Dermal in 70% Ethanol	No Overt signs of tocicity in dams or pups		PND 90:			PND 90	
					No difference in litter size	No differences in body weight			Beam Walking (Coordination)	Walk time: No Rx Effect	
							Midbrain	No Rx Effect			
							Cerebellum	↑ in AChEI (F)	Inclined Plane (Sensorimotor Reflexes)	↓ in fall angle (F)	
							Brainstem	↑ in AChEI (F)			
									Forepaw Grip Time (Motor Strength)	↓ in grip time (M & F)	

Article	Species	Exposure Period	Dose (mg/kg/day)	RoA	Maternal Toxicity	Pup Toxicity	AChEI	Task	Age at Test	Results
15 Laviola et al, 2006 Only data from wild type mice are presented. N.B.:	Wild-type Reeler mice	GD 14, 15, 16	5 mg/kg CPF-oxon	Osmotic mini pump (DMSO): Inserted under neck skin of dam	None (WT only)	None (WT only)	PND 10-12 WT type valuse used as the 100% level	Ultrasonic Vocalizations (Social Assessment)	PND 3, 7, 11	No Rx effect
								Grasping Reflex (Sensory/Motor)	PND 3, 7, 11	↑ in fall angle
								Righting Reflex (Motor)	PND 3, 7, 11	No Rx effect
								Locomotor Activity: Open Field	PND >70	No Rx effect
								Scopolamine Challenge	PND >70	↓ in Scop-induced locomotor stimulation
								Amphetamine Challgene	PND >70	↓ in Amph. induced locomotor stimulation

ATTACHMENT C

ATTACHMENT D, Pt. 1

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

DATE: February 25, 2010

SUBJECT: Transmittal of the Meeting Minutes of the FIFRA SAP Meeting Held December 1-3, 2009 on the Scientific Issues Associated with "Field Volatilization of Conventional Pesticides"

TO: Steven Bradbury, Ph.D.
Acting Director
Office of Pesticide Programs

FROM: Sharlene Matten, Ph.D.
Designated Federal Official
FIFRA SAP Staff
Office of Science Coordination and Policy

THRU: Laura Bailey
Executive Secretary, FIFRA SAP
Office of Science Coordination and Policy

Frank Sanders
Director
Office of Science Coordination and Policy

Please find attached to this memorandum the meeting minutes of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP) open meeting held in Arlington, Virginia on December 1-3, 2009. This report addresses a set of scientific issues associated with "Field Volatilization of Conventional Pesticides."

Attachment

cc:
Stephen Owens
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Betsy Shaw
Vicki Dellarco, Ph.D.
William Jordan
Margie Fehrenbach
Donald Brady, Ph.D.

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William Diamond
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Tina Levine, Ph.D.
Lois Rossi
Richard Keigwin
Jack Housenger
Enesta Jones
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Vanessa Vu, Ph.D. (SAB)
OPP Docket

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Steven Heeringa, Ph.D. (FIFRA SAP Chair)
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Kenneth Portier, Ph.D. (SAP Session Chair
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Michael Yost, Ph.D.

SAP Minutes No. 2010-02

A Set of Scientific Issues Being Considered by the Environmental Protection Agency Regarding:

Field Volatilization of Conventional Pesticides

December 1-3, 2009
FIFRA Scientific Advisory Panel Meeting
held at
One Potomac Yard
Arlington, Virginia

NOTICE

These meeting minutes have been written as part of the activities of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), Scientific Advisory Panel (SAP). The meeting minutes represent the views and recommendations of the FIFRA SAP, not the United States Environmental Protection Agency (Agency). The content of the meeting minutes does not represent information approved or disseminated by the Agency. The meeting minutes have not been reviewed for approval by the Agency and, hence, the contents of these meeting minutes do not necessarily represent the views and policies of the Agency, nor of other agencies in the Executive Branch of the Federal government, nor does mention of trade names or commercial products constitute a recommendation for use.

The FIFRA SAP is a Federal advisory committee operating in accordance with the Federal Advisory Committee Act and established under the provisions of FIFRA as amended by the Food Quality Protection Act (FQPA) of 1996. The FIFRA SAP provides advice, information, and recommendations to the Agency Administrator on pesticides and pesticide-related issues regarding the impact of regulatory actions on health and the environment. The Panel serves as the primary scientific peer review mechanism of the Environmental Protection Agency (EPA), Office of Pesticide Programs (OPP), and is structured to provide balanced expert assessment of pesticide and pesticide-related matters facing the Agency. FQPA Science Review Board members serve the FIFRA SAP on an ad hoc basis to assist in reviews conducted by the FIFRA SAP. Further information about FIFRA SAP reports and activities can be obtained from its website at http://www.epa.gov/scipoly/sap/ or the OPP Docket at (703) 305-5805. Interested persons are invited to contact Sharlene R. Matten, Ph.D., SAP Designated Federal Official, via e-mail at matten.sharlene@epa.gov.

In preparing these meeting minutes, the Panel carefully considered all information provided and presented by EPA, as well as information presented in public comment. This document addresses the information provided and presented by EPA within the structure of the charge.

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SAP Minutes No. 2010-02

A Set of Scientific Issues Being Considered by the Environmental Protection Agency Regarding:

Field Volatilization of Conventional Pesticides

December 1-3, 2009
FIFRA Scientific Advisory Panel Meeting
held at
One Potomac Yard
Arlington, Virginia

Kenneth M. Portier, Ph.D.
FIFRA SAP Session Chair
FIFRA Scientific Advisory Panel

Date: 2/25/10

Sharlene R. Matten, Ph.D.
Designated Federal Official
FIFRA Scientific Advisory Panel
Staff

Date: 2/25/10

Panel Members for the Meeting of the Federal Insecticide, Fungicide and Rodenticide Act Scientific Advisory Panel (FIFRA SAP) to consider and review Field Volatilization and Conventional Pesticides

December 1-3, 2009

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INTRODUCTION

The Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP) has completed its review of the Agency's analysis of **Scientific Issues Associated with Field Volatilization of Conventional Pesticides**. Advance notice of the SAP meeting was published in the *Federal Register* on **September 16, 2009**. The review was conducted in an open Panel meeting December 1-3, 2009 held at One Potomac Yard, Arlington, Virginia. Materials for this meeting are available in the Office of Pesticide Programs (OPP) public docket or via Regulations.gov, Docket No. EPA-HQ-OPP-2009-0687. Dr. Kenneth Portier chaired the meeting. Dr. Sharlene Matten served as the Designated Federal Official. Dr. Stephen Bradbury, Deputy Office Director for Programs, OPP, and Dr. Tina Levine, Director, Health Effects Division, OPP provided opening remarks at the meeting. Presentations of technical background materials were provided by Mr. Jeff Evans, Mr. Charles Smith, Dr. Judy Facey, and Dr. Elizabeth Mendez from the Health Effects Division, OPP; Dr. Faruque Khan, Mr. Chuck Peck, and Mr. Gabe Rothman from the Environmental Fate and Effects Division, OPP; and Ms. Annie Jarabek, National Center for Environmental Assessment, Office of Research and Development.

The *Standard Operating Procedures for Residential Exposure Assessment, i.e.*, Residential SOPs, is a set of standard instructions for estimating residential exposure resulting from various direct, labeled pesticide uses. Individuals in residential settings can also be potentially exposed via indirect exposure to conventional pesticides. These types of exposures can occur through a variety of means including field volatilization of conventional pesticides, spray drift, and take-home exposure. Methodologies for assessing indirect exposures are not currently included in the Residential SOPs.

Recently, the Agency has been exploring the development of an approach for assessing inhalation exposure resulting from the field volatilization of conventional pesticides. The following issues have been identified as key elements for this exposure scenario:

1) Use of the Agency's Reference Concentration (RfC) methodology to calculate Human Equivalent Concentrations (HECs) when inhalation toxicity studies are available.

2) Comparison of the use of inhalation vs. oral toxicity studies.

3) Development of a tiered approach to determine the level of complexity and refinement needed to estimate exposure, including:

    a) Use of available air monitoring data: California Air Resource Board (CARB), Pesticide Action Network – North America (PANNA), and other data sources.
    b) Development of a volatilization screening tool to estimate flux based on physicochemical properties of a pesticide.
    c) Use of more refined soil models to estimate flux.
    d) Use of air models to estimate concentrations around a treated field.

EPA's goal is to have a set of procedures that include transparent methodologies and data inputs that will guide the assessment of bystander exposure resulting from field volatilization of conventional pesticides in a straight-forward and user-friendly fashion. The Agency sought

comment from the Panel on the adequacy of the toxicological and exposure assessment methodologies; the applicability, analysis, and use of available air monitoring data; the strengths and limitations of the models being considered by the Agency for predicting flux of conventional pesticides; and the overall presentation of the issues related to field volatilization of conventional pesticides with respect to scientific integrity and public transparency.

PUBLIC COMMENTERS

Oral statements were presented by:

1. Susan Kegley, Ph.D., Principal and CEO, Pesticide Research Institute on behalf of Pesticide Action Network
2. Larry Jacobs on behalf of Jacobs Farm/Del Cabo Inc.
3. Jennifer Sass, Ph.D. on behalf of the Natural Resources Defense Council and others

Written statements were provided by:

1. Kenneth Racke, Ph.D. on behalf of Dow AgroSciences, LLC
2. Carol Dansereau on behalf of Farm Worker Pesticide Project
3. Jennifer Sass, Ph.D., National Resources Defense Council (NRDC) on behalf of Natural Resources Defense Council and others
4. Susan Kegley, Ph.D. Principal and CEO, Pesticide Research Institute on behalf of Pesticide Action Network - North America
5. Larry Jacobs on behalf of Jacobs Farm/Del Cabo, Inc.
6. Anne Katten on behalf of California Rural Legal Assistance Foundation
7. Dona Hippert and Lisa Arkin on behalf of Oregon Toxics Alliance
8. Jorga Stewart, private citizen
9. Carolyn Ashlock and Warren Trotter, private citizens
10. Lynn Bower, private citizen
11. Jan Wroncy, Gaia Visions, private citizen
12. Tom Kerns, private citizen
13. Maxine Centala, private citizen
14. Jean Public, private citizen

SUMMARY OF PANEL DISCUSSION AND RECOMMENDATIONS

TOPIC A: Exposure Assessment Issue

Traditionally, the Agency's assessment of bystander inhalation exposure to volatile pesticides has relied extensively on the use of air monitoring data. However, for the fumigants, an exposure assessment methodology was developed that combined the use of air models and air monitoring data. The Agency has taken the exposure assessment methodologies developed for the fumigants and further adapted them by utilizing soil models to predict field volatilization of conventional pesticides from plant and soil surfaces. Based on this premise, the Agency has identified several key factors for consideration by the Panel. They include the evaluation of the approaches and data sources used in the tiered exposure estimation methodology and use of soil models for predicting flux of conventional pesticides. Specifically, the Agency identified the following issues for the Panel to consider:

1. Tier I Approach for Identifying Volatile Chemicals of Concern for Risk Assessment, Air Concentration. The Tier I approach incorporates the use of vapor pressure alone to arrive at a saturated concentration in air. The estimated air concentration can be compared with available toxicity data to evaluate inhalation exposure concerns to human and other terrestrial organisms.

   Please comment on the Agency's approach for using the Tier I air concentration estimation method as a screening procedure. Please discuss the strengths and limitations of the screening approach. Please identify any alternative methods and/or physical-chemical properties, if any, which may be utilized as a screening procedure to identify chemicals with potential inhalation exposure concerns.

Panel Response

The Panel chose to combine its response to Question 1 with that of Question 2a (see below).

2. Tier II Approach for Identifying Volatile Chemicals of Concern for Risk Assessment, Volatility and Flux Models. Two options are being considered to refine the Tier I estimation method. Option A incorporates the use of physical-chemical properties including application rate, vapor pressure, solubility, and K_{oc} in an empirically-derived function to estimate flux rates. This option has less [sic] constraints and requires fewer input parameters to generate flux rates as compared to Option B described below.

   a. Given the state of the science, please comment on the applicability of using the Option A model to predict flux rates. Please discuss the strengths and limitations of this approach and how these impact the results. Please identify any alternative methods, if any, which may be utilized to identify chemicals with potential inhalation exposure concerns.

Panel Response to Questions 1 and 2a

The Panel identified problems regarding the use of a single vapor pressure value to arrive at a saturated vapor concentration for a Tier I approach for identifying volatile chemicals of concern. The major finding was that this approach has only a limited connection with how a pesticide is actually used in the real world, *e.g.,* as a formulation perhaps diluted in water with other additives like surfactants that may increase or decrease the effective vapor pressure relative to the active ingredient(s) value in its pure form. Therefore, the proposed Tier I approach may generate concentration predictions that are higher or lower than actually found in field measurements and will not necessarily provide predictions that are protective.

The Panel suggested that an alternative to the use of the proposed Tier I approach is to use the Woodrow et al. (1997) correlation approach to estimate a maximum 24-hour flux value. This approach is based on correlations between measured data and the physicochemical properties of the compounds, and it does not require an inordinate amount of data. This regression could be updated with all the latest studies and other relevant factors to increase its accuracy, a sensitivity analysis could be conducted on the model, then additional safety factors could be used to make it more protective as needed for a Tier I approach.

Tier II, Option B, is a refined process that utilizes fate and transport models to predict flux rates of applied pesticides that off-gas from treated fields. Current fate and transport models consider mechanisms related to volatilization, biodegradation, abiotic degradation, physicochemical properties, runoff, crop uptake, and leaching to account for the transformation and movement of the entire initially applied material. Volatilization mechanisms from bare soil and crop canopy surfaces are also important processes that the Agency believes ought to be considered to fully account for volatilization and diffusion from the vadose zone and canopy into the atmosphere. The Agency has utilized two models, the Pesticide Root Zone Model (PRZM) and the Pesticide Emission Assessment at Regional and Local Scales (PEARL) that incorporate these mechanisms and have the utility for the prediction of flux rates from treated fields. Option B requires extensive knowledge on environmental fate properties, as well as information related to application site, crop management and meteorology.

> b. *Please comment on the applicability of using fate and transport models to predict flux rates given the state of the science. Please discuss the strengths and limitations of both models and how these impact the results. Please identify any fate and transport model(s) which the Agency has not considered in this analysis which would be applicable for pesticide applications and crop management scenarios.*

Panel Response

The Panel agreed that the concept of coupling a fate and transport model, such as PRZM or PEARL, to predict fluxes, with a model, such as the Pesticide Exposure Risk Model for Fumigants Model (PERFUM), to estimate air concentrations at different distances from the field is a sound Tier II approach for identifying volatile chemicals of concern within the confines of evaluating pesticide volatilization in treated fields (excluding spray drift). The Panel noted that

while dispersion models, such as PERFUM, have been validated for fumigants, they have not been validated for semi-volatile pesticides. In addition, models such as PRZM or PEARL have also not been sufficiently validated for predicting semi-volatile pesticide volatilization from soil or crops. The Panel stated that rather than basing volatile flux prediction models on back-calculation methods, such models should be validated with direct field measurements of flux under different cropping patterns, application techniques, rates, and frequency, and in different geographic regions. The Panel proposed that the Pesticide Emissions Model (PEM) (Scholtz *et al.*, 2002a, b) be considered as a promising peer-reviewed, alternative model to predict flux rates of semi-volatile pesticides in treated fields.

TOPIC B: Toxicological Assessment Issues

As the Agency's understanding of the state-of the science in inhalation toxicology has evolved so has the Agency's approach to conducting inhalation hazard and risk assessments. This evolution has seen the Agency move from converting oral doses to inhalation concentrations to using the RfC methodology and/or physiologically-based pharmacokinetic (PBPK) models. As OPP continues to work on refining the risk assessment paradigm, the Agency is seeking the SAP's input on a number of key factors. They include the use of oral toxicity studies when inhalation studies are not available and the use of aerosol inhalation toxicity studies to represent toxicity to vapors of the same chemical. Specifically, the Agency identified the following issues for the Panel to consider:

B.1) The analysis conducted by the Agency indicates that, in general, oral toxicity studies may not accurately represent the full spectrum of toxic effects that may occur as a result of inhalation exposure. The analysis also indicates that - unless the same endpoints are identified through both routes of exposure - oral toxicity studies frequently underestimate toxicity by the inhalation route. The Agency has not been able to discern any patterns in this under/over estimation. *Please comment on any potential patterns that the Agency has not identified.*

Panel Response

The Panel concluded that route-to-route extrapolations using oral toxicity data to assess toxicity via the inhalation route is only scientifically justified, if a validated PBPK model is available or if the pesticide falls into Category 3 chemicals according to the Agency's Inhalation Reference Concentration (RfC) classification scheme (*i.e.*, those gases/vapors that cause systemic effects and not point-of-entry effects) (EPA, 1994) and the following criteria are also met.

a. The toxicological effect of concern is systemic for both entry routes and this effect is independent of route of exposure.

b. The first pass effects in the liver for oral exposure and in the respiratory tract for inhalation exposure are minimal to nonexistent or, if there is some first pass effect, the metabolism following exposure is the same for both exposure routes.

c. The chemical will not be chemically modified by the gastrointestinal bacteria or enzymes or by the acidic environment in the stomach differentially than it would be in the more

neutral to slightly basic environment of respiratory tract fluids. Pesticide stability in gastric fluid should be of primary concern in considering the use of oral exposure data. If the compound is not stable in gastric fluid, the toxicological data on oral dosing would be of little to no value in estimating any inhalation toxicity.

d. The absorption efficiency for oral and inhalation exposure must either be identical or known, so that accurate values may be incorporated into any model. The absorption efficiency for oral and inhalation exposure must either be identical or known, so that accurate dose values may be incorporated into any model; the use of a default value of 1 for absorption is not justifiable in the absence of any data. Furthermore, the absorption cannot be differentially influenced by a toxic response unless this response is the same via both routes and/or is influenced by relatively the same extent via both routes.

Even when the above criteria may be met, the Panel recommended an additional Uncertainty Factor (UF) of 10 for the final extrapolation for inhalation toxicity from oral toxicity data. The Panel stated that the use of uncertainty factors is not a replacement for more accurate models and data from more inhalation toxicity studies. The Panel strongly recommended that the Agency obtain additional inhalation toxicity data if such data are not available and if the vapor form of the pesticide does not meet the criteria for route-to-route extrapolation as described above.

The Panel suggested that some of the problems associated with route-to-route extrapolation from oral toxicity studies to inhalation toxicity studies might be evaluated by simple *in vitro* studies, such as solubility and stability in simulated gut or lung fluid, or by consideration of the chemical structure of the pesticide and using structural activity relationships.

The Panel made the following additional comments with regard to route-to-route extrapolation:

a. The Panel discussed several problems with the way in which EPA used Haber's Law (or Rule), in making adjustments for the duration of exposure when calculating HECs from repeated exposure inhalation toxicity studies.

b. The Panel noted two problems with the IEC equation. First, species differences in surface area/body weight are not accounted for, so that the equation needs to be modified. In cases for pesticides in Category 3 where route-to-route extrapolation may be justified, it would be better to adjust the animal oral dose to a human equivalent oral before further adjusting this value to an IEC for humans.

c. The Panel recommended that the Agency consider Benchmark Dose (BMD) analysis when concentration-response data are available and are amenable to modeling.

B.2) For a significant number of conventional pesticides, inhalation toxicity studies are not available. *Please comment on the scientific strengths and weaknesses of available approaches that may be used in the interim to assess inhalation hazard in the absence of inhalation toxicity studies.*

Panel Response

Overall, the Panel strongly recommended that the Agency conduct additional inhalation toxicity studies to adequately assess inhalation hazard. One Panel member provided a summary of alternative approaches reported in the literature that have been used to assess inhalation hazard in the absence of inhalation toxicity studies, *e.g.*, "threshold of regulation" or "threshold of no toxicological concern" (see Appendix C for a discussion of these procedures). The Panel stated that these approaches should not be used in the interim to assess inhalation hazard for pesticides in the absence of inhalation toxicity studies for the following reasons. These screening approaches do not involve a chemical-specific hazard identification or dose-response assessment and were originally developed to assess oral hazard in the absence of oral toxicity studies, and not inhalation hazard. While in a few cases the methodology has been used to evaluate inhalation hazard, the exposure durations in these cases (*i.e.*, chronic, lifetime exposure and one-hour intermittent exposure) do not correspond to the exposure durations the Agency is evaluating.

B.3) For inhalation toxicity studies the test material is typically aerosolized. After volatilization, however, the Agency anticipates exposures to vapors rather than the aerosolized particles. *Please comment on the predictive capabilities of aerosol studies to identify potential toxic effects and/or quantify the dose-response resulting from exposure to vapors. Is the Panel aware of any studies that quantitatively compare inhalation toxicity after exposure to vapors and aerosols? In the absence of such data, can the Panel recommend an approach to account for the potential differences between vapors and aerosols?*

Panel Response

The presence of a semi-volatile organic compound in either the vapor or particle phase will have impacts on site of deposition, absorption, and potentially dose and toxicity. The biological impact of the relative portions in each phase, however, has been poorly studied. Therefore despite known mechanisms for potential differences in dose and absorption, the ability to predict toxicity of vapors from aerosol studies is fairly limited. The Panel knew of no studies that have investigated the health impact of exposure to a single semi-volatile chemical under different phases. Some studies with mixtures have utilized techniques to either remove the particulate phase by filtration or the vapor phase by using a denuder to study the role of those materials independently (McDonald *et al.*, 2007). In many cases, the removal of the particulate phase of the mixture has not resulted in biological effects that differ from those obtained with the total mixture, especially for systemic effects. These mixture studies, however, may not be appropriate to answer questions related to a single component study. The Panel stated that there remain fundamental questions (beyond just the deposition site) related to the relative toxicity of vapors and particles. These must be considered as adding uncertainty when attempting to predict biological effects from exposure to the vapor versus particulate form of any given pesticide.

The Panel noted that any aerosol study of a semi-volatile compound will include that compound in both the vapor and particle phase. As a result, previous inhalation toxicity studies for semi-volatile pesticide registration contained the vapor phase of that compound in the aerosol.

Unfortunately, the studies conducted for pesticide registration did not measure the vapor portion of the mixture. The exposure monitoring in the inhalation toxicology studies was conducted with measurement of either weight gain or chemical content on filters that were used to trap the aerosol. That approach only measured the particle portion of the aerosol and the vapor that may have adsorbed on to the filter during sample collection. The vapor portion, which would have been present in these atmospheres, was not measured.

ADDITIONAL DISCUSSIONS ON INHALATION TOXICITY

The Panel discussed various topics as an outgrowth of the charge questions.

1. *Appropriate averaging times and sampling devices for field measurements of volatilized pesticide.* The Panel recommended that the Agency collect exposure data with shorter collection times than 24-hours and to use these data in health effects evaluations.

2. *Protocols for new inhalation toxicity studies (possible experimental designs).* The Panel recommended that inhalation toxicity studies should be conducted with durations of exposure of up to 90 days.

3. *Uncertainty factors for quality of the database and what constitutes a minimum database.* The Panel recommended that EPA establish criteria for short-term studies (*e.g.*, 1, 7, 14, 28 days) and that they use an additional UF of 10, if only a minimum database is available for their assessment.

4. *Moving toward a cumulative or total risk assessment.* The Panel recommended that total exposure be assessed to more fully encompass all types of inhalation exposures for the risk assessment process. The hazard quotient (HQ) or similar approach should be considered to assess risk from each of the types of exposure that contribute to the total potential exposure following application of pesticides.

TOPIC C: Risk Assessment Issues

The Agency discussed its methodology for combining the exposure estimation methodologies and inhalation toxicological approaches to estimate postapplication bystander inhalation risks resulting from field volatilization of conventional pesticides. In estimating postapplication bystander inhalation risks, there are a few principles that should be followed: (1) It is important to properly match the duration of the exposure with a proper toxicity study of comparable duration. (2) Both dissipation of air concentrations around a treated field as well as when retreatment of the field may occur need to be considered. (3) Clearly define the uncertainties and limitations of this type of assessment. The Agency has identified the following issues for the Panel to consider with respect to estimating postapplication bystander inhalation risks:

Please comment on the strengths and limitations of the Agency's use of the empirical and modeled air concentrations in the provided risk assessment case study. Does the Panel agree that the postapplication bystander inhalation risk estimate case study appropriately matches

the duration of the exposure with the proper toxicological study of the same duration? Please comment on the scientific strengths and weaknesses of conclusions and characterization regarding the estimated risks presented in the case study.

Panel Response

C.1) Please comment on the strengths and limitations of the Agency's use of the empirical and modeled air concentrations in the provided risk assessment case study.

The strengths and weaknesses of the use of the Woodrow empirical model to assess risk are much the same as the Panel discussed related to Topic A, Question 2a (see above). The main strength of this model is its basis in multiple studies over a wide range of vapor pressures; a secondary strength is that its results are in the range of the results of the air concentrations estimated by more sophisticated computer-modeled air concentrations. The limitations of the empirical model are that it is based on a limited range of crops, weather, and locations, and does not take into account the potential effect of an activity coefficient on vapor pressure, and it is applicable only to the first day post-application.

The strengths of the computer-based modeled air concentrations are that they can account for dynamic changes in post-application conditions and residue history. Their weaknesses are the limited knowledge that users have of the internal components of these models, the concern that the components do not model evaporation from foliage as well as they model evaporation from soil, and that the particular analyses presented to the Panel inappropriately decoupled the variance of the flux from the variance of dispersion.

C.2) Does the Panel agree that the postapplication bystander inhalation risk estimate case study appropriately matches the duration of the exposure with the proper toxicological study of the same duration?

The Panel agreed that the case study appropriately matched the duration of the exposure with the proper toxicological end point, although there were some questions regarding the specificity of the target population within this particular case study. Again, a great deal of the Panel's discussion on Topic B apply here, particularly the limitations of the route-to-route extrapolation process and the need to apply toxicological data collected from one exposure to another. Thus, the success of this case study to achieve its goal is tempered by the limited confidence that the Panel has in extrapolating toxicological data considering both route and duration of the exposure.

However, in a broader sense, the Panel agreed that the example did not adequately consider the ability to model differences in duration of the exposure. In earlier sections, the Agency presented three types of exposure scenarios for risk assessment, short- (up to 30 days), intermediate- (up to 90 days) and long-term (greater than 1 year). The example only presented modeled air concentrations for short-term exposures. The models did not estimate intermediate and long-term exposures. Two of the five pesticides listed in Table 3 (of the Agency's background document) used for modeling short-term exposures have reported soil

half-lives of greater than 150 days, illustrating that intermediate exposures from single applications may occur. The Panel recommended the Agency consider adding longer term exposures of more than 30 days in the models to address intermediate and long-term chronic exposures and to match toxicological studies.

C.3) Please comment on the scientific strengths and weaknesses of conclusions and characterization regarding the estimated risks presented in the case study.

The Panel broadly agreed that the case study included all or most of the important elements to conduct a proper risk assessment. The strength of the inhalation toxicity and exposure data bases for the chemical chosen led the Panel to conclude that the inhalation hazard and exposures assessments, and Margin of Exposure (MOE) analysis were realistic based on field monitoring data. However, the Panel had a range of recommendations for how this model and the risk assessment process could be improved, and reservations if such an analysis were applied to many other chemicals. For instance, the toxic endpoints are unlikely to be as strong for other chemicals, the details and general applicability of some steps in the process were not well-defined, PRZM or PEARL were not optimized for their application to the evaporation of semi-volatile pesticides, and the impact of the propagation of uncertainty and safety factors within the process on the final result is uncertain. An alternative risk assessment approach based on the Volatilization Hazard Ratio (VHR) was presented and the case study chemical was evaluated using this approach.

DETAILED RESPONSES TO CHARGE QUESTIONS

TOPIC A: Exposure Assessment Issue

Traditionally, the Agency's assessment of bystander inhalation exposure to volatile pesticides has relied extensively on the use of air monitoring data. However, for the fumigants, an exposure assessment methodology was developed that combined the use of air models and air monitoring data. The Agency has taken the exposure assessment methodologies developed for the fumigants and further adapted them by utilizing soil models to predict field volatilization of conventional pesticides from plant and soil surfaces. Based on this premise, the Agency has identified several key factors for consideration by the Panel. They include the evaluation of the approaches and data sources used in the tiered exposure estimation methodology and use of soil models for predicting flux of conventional pesticides. Specifically, the Agency identified the following issues for the Panel to consider:

A.1) Tier I Approach for Identifying Volatile Chemicals of Concern for Risk Assessment, Air Concentration. The Tier I approach incorporates the use of vapor pressure alone to arrive at a saturated concentration in air. The estimated air concentration can be compared with available toxicity data to evaluate inhalation exposure concerns to human and other terrestrial organisms.

Please comment on the Agency's approach for using the Tier I air concentration estimation method as a screening procedure. Please discuss the strengths and limitations of the screening approach. Please identify any alternative methods and/or physical-chemical properties, if any, which may be utilized as a screening procedure to identify chemicals with potential inhalation exposure concerns.

A.2) Tier II Approach for Identifying Volatile Chemicals of Concern for Risk Assessment, Volatility and Flux Models. Two options are being considered to refine the Tier I estimation method. Option A incorporates the use of physical-chemical properties including application rate, vapor pressure, solubility, and K_{oc} in an empirically-derived function to estimate flux rates. This option has less [sic] constraints and requires fewer input parameters to generate flux rates as compared to Option B described below.

a. Given the state of the science, please comment on the applicability of using the Option A model to predict flux rates. Please discuss the strengths and limitations of this approach and how these impact the results. Please identify any alternative methods, if any, which may be utilized to identify chemicals with potential inhalation exposure concerns.

Tier II, Option B is a refined process which utilizes fate and transport models to predict flux rates of applied pesticides which off-gas from treated fields. Optimum fate and transport models consider mechanisms related to volatilization, biodegradation, abiotic degradation, physicochemical properties, runoff, crop uptake, and leaching to account for the transformation and movement of the entire initially applied material. Volatilization

mechanisms from bare soil and crop canopy surfaces are also important processes, which the Agency believes ought to be considered to fully account for volatilization and diffusion from the vadose zone and canopy into the atmosphere. The Agency has utilized two models, the Pesticide Root Zone Model (PRZM) and the Pesticide Emission Assessment at Regional and Local Scales (PEARL) which incorporate these mechanisms and have the utility for the prediction of flux rates from treated fields. Option B requires extensive knowledge on environmental fate properties, as well as information related to application site, crop management and meteorology.

b. *Please comment on the applicability of using fate and transport models to predict flux rates given the state of the science. Please discuss the strengths and limitations of both models and how these impact the results. Please identify any fate and transport model(s) which the Agency has not considered in this analysis which would be applicable for pesticide applications and crop management scenarios.*

Panel Response to Questions 1 and 2a

During the progression of the panel discussion, Questions 1 and 2a became linked. Therefore, the Panel combined its response to Question 1 with that of Question 2a.

Vapor pressure clearly is an important physical property in evaluating the behavior of pesticides, especially with respect to volatilization from plant surfaces. However, the Panel concluded that the use of the Tier I approach for identifying volatile chemicals of concern was overly simplistic and of no real value as a screening tool. The Panel identified four major issues with the Tier I screening approach (discussed below). The Panel proposed the Woodrow *et al.* (1997) correlation approach as an alternative to the proposed Tier I approach (referred to as Tier II Option A within the EPA background document, p. 24) to estimate a maximum 24-hour flux value that then could be used to calculate a maximum air concentration over 24-hours.

Major Issues with the Tier I Approach

This report has organized the Panel's responses into the following four groups of topics: overestimation of air concentrations, temperature considerations, low vapor pressure (VP) pesticides, and pesticide formulation considerations.

1. *Overestimation of air concentrations.* The saturated vapor concentration calculated from vapor pressure appears to generally over-predict the maximum concentration measured in field settings by such a wide margin as to make the predictions useless. In the examples within the EPA background document paper, *i.e.,* Table 4, p. 36, the proposed Tier I approach overestimates the observed concentrations by about two orders of magnitude when the pesticide is on foliage and five orders of magnitude when on soil.

   Vapor pressure is the sole variable both in the Tier I model and in the Tier IIA model for volatilization following foliar applications.[1] Vapor pressure also plays a central role in

[1] In contrast, both of the Tier IIB methods (PRZM and PEARL) use Henry's law constant instead of vapor pressure (that constant is related to VP, as will be discussed *circa* Eqn. A4 below). .

the Tier IIA model for volatilization from soil as evidenced from the 0.88 R^2 correlation between log flux and just the log VP across the 10^6 range of VP values used to derive that model (Woodrow *et al.*, 1997). However, as a simple predictor, the Tier I model over-estimated the maximum measured field concentrations of the pesticides presented in Table 4 of the Agency background document (p. 36). The over-estimates for the four chemicals applied to foliage ranged from factors of 12- to 174-fold (with a geometric mean of 48-fold); however, the over-estimate for the one soil application (Chemical D) was 27,472 times greater than its maximum monitored concentrations. Thus, at first glance. Thus, at first glance, the Panel concluded that a simple comparison of vapor pressure to toxicity was too conservative to be useful.

In contrast to the above generalization, several recent studies conducted in the United Kingdom showed errors in the opposite direction. One Panelist reported on studies (sponsored by the UK Department for Environment Food and Rural Affairs, Defra) in which the air concentrations of two of the five pesticides studied were higher following application than the concentration predicted by the Tier I model. In particular, the air concentrations of epoxiconazole and prothioconazole were higher than predicted by the Tier I model. A summary of these findings is presented below while a more detailed report on these data is provided in Appendix A. In the first of these studies (Bulter-Ellis and Miller, 2008; Figure A-2 of Appendix A) in which epoxiconazole was applied to 4.8 hectares of a cereal crop, air concentration at a height of 0.7 meters and 2 meters downwind from the field's edge of a 4.8 hectare 2-3 hours after application was one and a half times the predicted saturated vapor concentration. And in a similar study at another location (Appendix A, Figure A-3), measured air concentrations from 0-90 minutes after application were nearly two times the predicted saturated vapor concentrations. In a second U.K. trial (Defra Project PS2023) conducted in October 2009 at a laboratory independent of the first facility (Table A-4 of Appendix A), the air concentrations of prothioconazole peaked about 8 hours after application at over twice its saturated vapor concentration. These observations cast doubt on the reliability of the proposed Tier I approach in the other direction.

2. *Temperature considerations.* The Tier I model as proposed would use a vapor pressure value based on only a single temperature (typically 20-25°C) and therefore does not provide a realistic prediction of the concentration on hotter than normal days. Temperatures observed in agricultural regions differ significantly from region to region, season to season, or even from day to night, and this variation is enough to change the vapor pressure by a factor of two in either the positive or negative direction.

   Vapor pressure should be described as a function of temperature. Surface temperature should be used in the models to estimate volatilization from the earth's surface. One panelist commented that possibly some of the pesticide behavior observed in the UK data presented in Appendix A could be due to the difference between surface temperature and air temperature. A pesticide will volatilize in proportion to its vapor pressure at the surface (soil or leaf) temperature that can exceed the air temperature on a sunny day by about 10°C. The temperature of the pesticide residues in the UK studies may not have been accounted for in their reported data. As a rule of thumb, an increase in a liquid's temperature of 12°C (21°F) will increase its vapor pressure by a factor of 2.

3. *Low vapor pressure pesticides.* Vapor pressure data for pesticides with very low vapor pressures can be very difficult to obtain and may be subject to substantial errors and uncertainty. These low vapor pressure substances are the most likely to be 'screened out' by a simple pure vapor pressure approach.

   It is difficult to accurately measure vapor pressures as low as those for the pesticides in the UK studies (*circa* 10^{-7} to 10^{-8} mmHg). For example, for all six of the fourteen pesticides considered by Woodrow *et al.* (2001) that had reported vapor pressure values of less than 10^{-2} mmHg (and these only ranged from 10^{-3} to 10^{-4} mmHg), the reported VP values underestimated the vapor pressure values predicted by their physical characteristics by an average of 1.6-fold.. Thus, the values for the vapor pressure of epoxiconazole and prothioconazole reported in the UK studies may have been in error.

4. *Pesticide formulation considerations.* A pesticide's vapor pressure as a pure chemical [active ingredient(s)] has only a limited connection to its vapor pressure as it is actually used in the real world. For example, pesticides are typically applied as a formulation, perhaps diluted in water with other additives like surfactants, and these factors may elevate the vapor pressure of the active ingredient relative to the pure compound value. Therefore, the proposed Tier I approach may generate concentration predictions that are higher or lower than either the vapor pressure that exists in field conditions or that is actually found in field measurements, and thus will not necessarily provide predictions that are protective. In the case of the UK data, the chemical's vapor pressure in an aqueous mixture on foliage may have been increased beyond that reported for a pure or neat substance. The potential magnitude of that effect is described below. Whether or not an activity coefficient or one of the other explanations described above applies to the recent findings in the UK is unknown at this time; however, unless an explanation can be found, those findings cast doubt on the whole premise of these models.

Predicting a Pesticide's Vapor Pressure in a Mixture

The best model for predicting a chemical's vapor pressure in a mixture is the use of an empirical adjustment to Raoult's law for an ideal mixture (Eqn. A1).[2] Such an adjustment is called an "activity coefficient" and is typically given the symbol γ (a lowercase Greek gamma), and X_i is the molar concentration of the chemical within the mixture. Equation A2 both defines γ_i mathematically and expresses the empirical concept behind it.

$$P_{vapor,\,i} = \gamma_i \times X_i \times P_{vapor} = \gamma_i \times \text{Raoult's predicted } P_{vapor,\,i} \qquad \text{Eqn. A1}$$

$$\gamma_i = \frac{\text{Measured or actual } P_{vapor,\,i}}{\text{Raoult's } P_{vapor,\,i} = X_i \times P_{vapor}} \qquad \text{Eqn. A2}$$

A chemical's activity coefficient is not actually a constant but is broadly a function of both the solvent in which it is mixed and its concentration in that mixture. Figure B-1 in Appendix B

[2] More detailed information on the behavior of vapor pressure in mixtures is provided in Appendix B.

depicts the pattern of γ as a function of *X* for a few common organic solvents in water. As a component in a mixture gets more and more dilute, its γ value eventually reaches a constant γ∞ (the ∞ sign denotes infinite or a very high dilution). The magnitude of some activity coefficients in water is certainly sufficient to cause major deviations from an ideal mixture. For instance, a γ^{∞} of 50 is sufficient to increase a chemical's vapor pressure by a factor of more than 2-fold when it is present in water at molar fraction of 10% to 30%, and a γ^{∞} of 400 or more can increase a chemical's vapor pressure by more than an order of magnitude. Unfortunately in the case of semi-volatile pesticides applied to crops, the pesticide concentration in water is an independent and dynamic variable (a pesticide applied in water may start out dilute, rapidly become concentrated as the water in the droplet evaporates, and potentially become dilute again due to dew, *etc.*). In addition to water, the pesticide is likely to be absorbed into the organic components of a leaf's cuticle, in which a different value of γ would apply.

The Panel acknowledged that the value for a given chemical's γ∞ in water can be predicted from its Henry's constant via Eqn. A3 (although to be used quantitatively in that equation, the value of H_i must be in the same units as P_{vapor}).

$$\gamma_i^{\infty} = \frac{H_i}{P_{vapor}} \qquad \text{Eqn. A3}$$

The Panel commented that most of the Tier II, Option B models appear to use a Henry's law constant, although it is not clear just how it is used. Henry's Law allows one to predict the vapor pressure of a component in a water solution based on a fixed empirical coefficient often called a "Henry's Law constant" and denoted by the symbol "H_i" herein. A common expression of Henry's Law might look like Eqn. A4. While such constants are widespread and often available for pesticides, a known limitation of Henry's law is that it only applies to very dilute mixtures. Use of Henry's law for more concentrated mixtures will introduce another set of errors that are only touched on in Appendix B.

$$P_{vapor,i} = H_i \times X_i \qquad \text{Eqn. A4}$$

In principal, an activity coefficient could be applied to Raoult's law to correct any vapor pressure for its non-idea behavior in a mixture. Computer codes have been developed to predict activity coefficients for various mixtures and can be applied to semi-volatile pesticides (Muro-Suñé *et al.*, 2005). However, such an approach may have limited utility as part of a screening tool for these pesticides because the magnitude of the correction depends on the composition of the mixture. The Panel thought that it might be feasible to find the highest product of $\gamma_i \times X_i$, but for a more complete dynamic prediction necessary for a Tier IIB model, every formulation would need to be evaluated separately due to the varying components in applied mixtures. Additionally, any effects due to changing mixture composition and temperature post-application also would need evaluation. This would require a very complex set of computations that go beyond the goals of a Tier 1 approach.

The Woodrow et al. (1997) Approach as an Alternative to the Tier I Approach

The Panel proposed the Woodrow *et al.* (1997) approach as an alternative to the proposed Tier I approach (referred to as Tier II Option A in the EPA background document, p. 24) to estimate a maximum 24-hour flux value that then could be used to calculate a maximum air concentration. The Woodrow approach is based on empirical data in addition to selected physicochemical properties of the compound when applied to soil. It allows for an evaluation of pesticides applied directly to water or plants or incorporated into soil, and yet it does not require an inordinate amount of data. This simplified model could be updated with all the latest studies and other relevant factors to increase its accuracy. A sensitivity analysis could be conducted to refine this model. Additionally, confidence intervals around the model predictions could be calculated and safety factors could be added to the results of the model predictions to make it more protective as needed for a Tier I.

As described in the EPA background document, Woodrow *et al.* (1997) established a correlation (Eqn. A5). between *ln Flux* ($\mu g/m^2$ *hr)* and *ln* $[(VP \times CF \times AR)/(K_{oc} \times S_w)]$ for soil applied pesticides, where *VP* is vapor pressure (Pa), *CF* is a conversion factor 133.32 Pa/torr, *AR* is the application rate (in units of kg/ha), K_{oc} is the soil organic carbon-water partition coefficient (mL/g), and S_w is the pesticide's aqueous solubility (mg/L.

$$X = 19.35 + 1.0533 \ln\left[\frac{VP \times CF \times AR}{K_{OC} \times S_W}\right] \qquad \text{Eqn. A5}$$

Similar correlations just involving vapor pressure were also developed for pesticides applied to plants or inert surfaces, and pesticides applied to water. While this approach also relies on the vapor pressure of the compound, the correlation is grounded in measured flux data that could be used to calculate a more realistic air concentration value.

The Panel recommended that EPA update the correlation equations published by Woodrow *et al.* (1997) with data published since this work was carried out to strengthen the reliability of predictions, *e.g.*, Leistra *et al.* (2006). In addition, the Panel recommended that additional studies be commissioned by EPA to expand the number of data points in the regression for flux from foliage and flux from soil. Consideration should also be given in future studies to plot designs that can improve the quality of the measured flux data, *e.g.*, see Majewski *et al.* (1990, 1991) for a description of comparing multiple methods for measuring pesticide soil volatilization rates.

Some Panelists were concerned that the Woodrow approach would not adequately assess the potential impact of some spray from foliar applications bypassing the foliage and falling to the soil surface. Several panelists commented that foliage residues would, in most cases, contribute the largest fraction of the total flux to the air, at least for the first 24-hours after application. However, over longer spans of time, flux from the soil could contribute more significantly to the overall flux. However, this issue is a minor concern for the continued use of this model because only the maximum flux rate is used in the correlation and the model only to estimate the maximum flux rate on the first day and not to provide an estimate of subsequent temporal trends

or concentrations beyond the first day. As discussed in Woodrow *et al.* (1997), the Panel suggested that leaf surface area be added as a modifying factor to make the model more robust when applied to different crops. Soil moisture should also be added to refine the soil portion. Pesticides volatilize more rapidly from moist soil than from dry soil, other factors being constant. Other additions aimed toward refining the flux model for soil applications would include the depth of incorporation of the chemical after application, soil temperature, and wind speed at the soil surface.

The Panel noted that there is a lot of uncertainty in the physicochemical data and measuring volatilization fluxes (Majewski, 1996). A major factor in the Woodrow model is the *Koc* term, which is not currently required at pesticide registration. However, a highly correlated measure, *Kow*, *i.e.,* octanol-water partition coefficient (EPA, 1996) is required and if the *Koc* is estimated from the *Kow*, another source of uncertainty will be added to modeled air concentrations. The Panel recommended that EPA should define whether the *Koc* will be estimated from the *Kow*, and by what method, or if literature values will be used.

The Panel suggested that it was possible to assign confidence intervals to the regression to incorporate the uncertainty associated with the measurements used to generate the flux values. These intervals would provide a high and low range of likely values and therefore address the Agency's concerns that the model would under-predict the 24-hour exposure for some compounds. For example, Johnson *et al.* (1995) describe a method for evaluating the quality of physicochemical property data available in the literature or in registrant submitted data. In addition, the Panel recommended that a sensitivity analysis of the terms within the model would be useful, perhaps breaking the regression equation into pieces rather than having everything lumped together. A further suggestion from the Panel for using the Woodrow *et al.* (1997) approach would be to set a receptor location next to the field (*i.e.*, 2-5 meters) and an air concentration of concern estimated from the toxicity data. From this information a maximum flux value could be calculated that would be needed to generate this concentration of concern. Flux values independently generated from the Woodrow model, including confidence intervals, could then be compared with the maximum flux value determined from the air concentration of concern for screening purposes (see also the Vapor Hazard Ratio (VHR) discussion in Topic C). One caveat with respect to the Woodrow model is that some chemicals are applied in such a way that the first 24-hours after application may not include the period of maximum flux. For example, soil incorporated pesticides need time to diffuse to the soil surface or pesticides broadcast as granules into rice fields must dissolve before maximum flux from the water surface can occur. In these exceptional cases, the second or subsequent 24-hour period may contain the period(s) of maximum flux rather than the first 24-hour period.

Panel Response to Question 2b

Major Recommendations/Findings

The Panel agreed that the concept of coupling a fate and transport model such as PRZM or PEARL to predict fluxes, with a model such as the Pesticide Exposure Risk for Fumigants (PERFUM) to estimate air concentrations at different distances from the field is sound as a Tier II, Option B modeling approach for identifying volatile chemicals of concern within the confines of evaluating pesticide volatilization in treated fields (excluding spray drift).. However, the Panel

also suggested that new insights might be gained by conducting the proposed Tier II process in reverse order: use a dispersion model (*e.g.*, Screen3, ICS3, or PERFUM) to establish a maximum acceptable flux that will not produce a concentration of concern at a given receptor location and then use either a Tier I or Tier II screening procedure to establish a maximum acceptable flux estimate for the specific compound. The Panel suggested that the use of the Pesticide Emissions Model (PEM) (Scholtz *et al.*, 2002 a, b) be considered as an alternative to the use of PRZM, as this model was created to describe processes controlling volatilization.

The Panel recommended that novel technologies (sensors or rapid samplers) be explored to achieve higher temporal resolution in the concentration datasets for flux measurements. Innovative methods to characterize gas and particle-phase concentrations during flux measurements and downwind air sampling should be encouraged. For example, the Agency might consider applying other methods for flux estimates, such as EPA method OTM-10 that applies remote sensing techniques. The Panel noted that whatever method is considered, each will have different uncertainties and sensitivities to measured parameters.

The Panel offered several recommendations and suggestions regarding validation and further development of flux models.

1. The Panel stated that rather than basing volatile flux prediction models on back-calculation methods, such models should be validated with direct field measurements of flux under different cropping patterns, application techniques, rates, and frequency, and in different geographic regions. Only once the flux model is validated is the use of multi-year/multi-climate zone meteorological datasets appropriate.

   The Panel concluded that the flux model evaluation presented in the background document is not sufficient to validate the proposed flux models, *i.e.*, PRZM or PEARL. Both models appear to over-predict the "observed" field fluxes. The Panel stated that the "observed" fluxes were not accurate enough or represented an insufficiently extensive database to show that the models performed well.

   For example, in some cases the models, PRZM and PEARL, used within the Agency's examples (Table 5 of the Agency background document, p. 47) predicted roughly twice the measured air concentration, while in others it was as great as seven-times the measured concentration. One possible explanation for such deviations is that the measured data used for comparison were not based on actual source volatilization flux data, but on a back-calculation of source volatilization fluxes that required an air dispersion model to estimate concentrations away from the application site. The model predictions presented for PRZM and PEARL also included estimations of temperature fluctuations as well as other meteorological conditions explained by the EPA experts.

   The model should first be validated with field data that reflects actual experimental conditions before it is evaluated for efficacy and ruggedness (with averaged data). Only then can the predictive capability of the model be accurately assessed. The data used in the Agency's examples were projecting a protective model, but they were too imprecise for an evaluation of the model's validity. The Panel emphasized that the use of actual field flux data to evaluate Tier II models would be paramount in assessing the accuracy

and reliability of the model's predictive capabilities. To that end, the Panel recommended that additional field studies be conducted because those presented in the Agency's background document did not demonstrate that the model could accurately predict vapor phase concentrations.

The Panel made a number of further recommendations related to this general issue of validating evaporation and dispersion models.

a. The evaluation of the flux models should be handled separately from the risk assessment. Flux models using probability meteorology would involve a wide range of atmospheric concentrations; thus, adding unrealistic variability to the surface flux predictions. Therefore, it is important first to understand the model, assure that it predicts known conditions accurately, and then add a realistic level of variability as input to the model to ensure a conservative risk assessment.

b. The model results should be compared to a number of actual field flux measurements, preferably obtained with multiple field flux measurement methods, in order to be proven effective. For example, direct flux measurements, including eddy accumulation as well as indirect methods, such as flux-gradient relationships, should be used in addition to the Gaussian plume inversions used by the Agency. The Panel pointed out that many such studies exist, and some are provided in the list of suggested references.

c. The Panel questioned the use of the Gaussian plume inversion approach for estimating fluxes within an orchard. Trees in an orchard can create coherent eddies organized within the geometry of the orchard, and create a stable sub-canopy layer of air. These effects are not taken into account in the plume models employed to infer fluxes. This is also a problem for atmospheric concentrations estimated from orchard emissions using PERFUM, because the Panel understood that PERFUM is based on the Gaussian plume model and is not adapted to simulate dispersion and transport within plant canopies.

d. The Panel expressed concern that the uncertainty associated with physicochemical properties, such as the *Kow*, soil half-life, and photodegradation, are not incorporated into the current modeling efforts. The Panel recommended that the Agency perform sensitivity analyses to determine the factors (input data and model parameters) that are most important in predicting the flux, and then the impact of uncertainty in these inputs should be used to evaluate the range of fluxes predicted by the model. Flux estimates, even for a given day of meteorological conditions, can be computed as a probability distribution.

e. Flux model evaluations should include situations involving both soil and vegetation sources from soil and vegetation application of pesticides.

f. The Panel recommended that more frequent time point measurement of flux rates should be taken immediately after application, *i.e.*, hourly flux rates. Hourly flux

estimates should be considered for the following reasons: 1) volatilization rates tend to be higher in the hours just after application (*e.g.,* 2-fold as shown in Table C-1) than the daily average, 2) day/night changes in surface temperatures can lead to a substantial difference in surface temperature, thus in vapor pressures, and thus, volatilization rates, and 3) conditions of high atmospheric stability, typically at night, can lead to atmospheric concentrations that are much higher than the daytime average. The sensitivity of the more technologically-advanced mass spectrometers should be enough to measure the levels that would be found at very short sample durations, especially because those initial samples are taken at a time when the airborne concentrations are apt to be the highest.

g. In addition, the Panel indicated that downwind concentration data should also be collected during the field validations of the flux model in order to validate a coupled flux/dispersion model approach for semi-volatile pesticides. While dispersion models have been field-validated for fumigants, they have not been field-validated for semi-volatile pesticides or in other soil types or crop scenarios. Because of their high volatility, fumigant vapors tend to remain airborne for longer periods than semi-volatile pesticides, which tend to be removed from the air mass by condensation or adsorption on downwind air particles, soil, or foliage surfaces. By absorption onto particles and potentially re-volatizing, semi-volatile pesticides with higher *Kows* (*i.e.*, log *Kow* greater than four) have a different fate than fumigants, may have a longer persistence in the environment, and may be transported into homes where the pesticides may reside in house dust (Harnly et al, 2009).

2. Volatilization flux prediction models should include pesticide degradation products. For example, photodecomposition is important to include because it is generally a "loss" term. However, transformation reactions may also be a source of more toxic chemicals, *i.e.*, oxones (oxygen analogs of many organophosphorus pesticides) (Harnly *et al.*, 2005).

The Panel stated firmly that models used to predict the atmospheric concentration of a pesticide should include enough terms to make the predictions act as reasonable surrogates of the measured concentrations. This includes terms that both add to the vapor phase concentration and those that decrease vapor phase concentration (*i.e.,* reduce the parent compound). If the model does not include subtraction terms it will not accurately follow the progression of the flux and is likely to significantly over predict the vapor phase concentration. This more sophisticated Tier II model should be used to evaluate pesticides once they have been screened at Tier I, but any advanced model should include photodegradation as part of the modeling process. Many of the newer pesticides are photo-labile. For example, pyrethorioids often decay rapidly even once collected from field samples. Much of this degradation can be attributed to photodegradation, reinforcing the need for inclusion of photodegradation terms in any model used as a Tier II assessment tool. A photodegradation term will represent the extent to which the vapor phase concentration of the pesticide in the atmosphere is reduced by photoloysis (although not to zero). Existence of this term in the model will help to differentiate the more photo-labile compounds from others that may have a similar vapor pressure, but do not undergo photodegradation.

utilized in the flux modeling. The Panel stressed that the Agency should be more attentive to the physics of the models being tested.

7. The Panel suggested that PEM (Scholtz *et al.*, 2002a, b) would be a reasonable alternative to the use of PRZM or PEARL, as this model was designed to model the behavior of the pesticide into the ground rather than release to vapor, the release to vapor is almost the discard process, the "1 - N term." While a precise model should be able to utilize all calculated compartments, these models (PRZM and PEARL) focus on the soil compartments and therefore potentially over-emphasizes the fraction retained in the soil. Those assumptions will then cause an underestimation of the fractions released as vapor. The need for accurate volatilization flux data will be paramount in evaluating the validity of the proposed model.

8. The Panel also suggested that EPA hold scientific workshops or conferences to bring together the scientific community to aid in the assessment of proposed models and improvements (if necessary), and eventually, the validation of these flux models. Panel members volunteered to help in this endeavor.

9. And finally, the Panel suggested that the Agency consider developing a model of multiple application events in the same region/air shed. In that regard, the Panel made the following comments and recommendations described below.

   a. The Panel agreed that the Tier II models used by the Agency to estimate volatilization flux and air concentrations should focus first on single applications. However, some panelists indicated that the impact of applications on multiple surrounding fields and/or regional use should eventually be considered. In addition, the presentation from Jacobs Farm during the public comment section also raises concerns regarding re-volatilization of deposited pesticides (repeated cycles of volatilization, transportation, and re-deposition of applied pesticides), especially during fog events. Such re-volatilization processes are not considered within in the current modeling efforts.

   b. In addition, the Panel noted that to derive the greatest benefit from the downwind transport and dispersion models, they should be applicable to modeling air concentrations from multiple applications of the same pesticide in a locale, air shed, or air basin. An example of what could be done for fumigants and semi-volatile pesticides is available in a published study on methyl bromide volatilization from several treated fields in the Salinas Valley, CA (Honaganahalli and Seiber, 2000).

   c. The Panel suggested that the impact of crop management practices such as irrigation, tilling, mulching, and burning of fields may have a potential to increase volatilization. These practices and their effects on pesticide volatilization should also be considered.

Terminology Corrections in Agency's Background Document

The following terminology corrections in the Agency's background document were provided by one Panelist.

1. The following text from page 19 describing flux measurement methodology should be edited for accuracy.

"Flux studies also are typically designed to allow for the generalization of results using a computer simulation air model. More accompanying information is generally collected in these studies including meteorology at differing heights typically with thermo anemometers to provide high resolution information about environmental conditions which are important in understanding the movement of pesticides from the treated area and reducing the uncertainty associated with the flux calculations."

   a. Flux measurements are typically designed to measure the flux, not the concentration, of a trace gas. One can infer fluxes from an array of concentration measurements, such as has been described here, but this is not what most micrometeorologists would call a flux study. See Dabberdt *et al.* (1993) for a concise description of flux measurement methods.

   b. In comparison to sonic anemometers, thermo anemometers are not the most sophisticated tool to use to measure turbulent wind associated with turbulent transport. Thermo anemometers are not used to reduce uncertainty in the calculations. The Agency's wording reflects a weak understanding of the research underlying these transport mechanisms,

2. The following text from page 20 needs to be edited.

"There are a number of recognized common flux methods in the peer-reviewed literature. Some of the common methods are the Indirect or Back-calculation Method, the Aerodynamic Method (Majewski *et al.*, 1993), and the Integrated Horizontal Flux Method (Wilson and Shum, 1992)."

There are additional flux methods that should be considered by the Agency, for example, eddy covariance. The Agency should review Dabberdt (1993) and modify this section as they see fit after reviewing the available literature.

TOPIC B: Toxicological Assessment Issues

As the understanding of the state-of the science in inhalation toxicology has evolved, so has the Agency's approach to conducting inhalation hazard and risk assessments. This evolution has seen the Agency move from converting oral doses to inhalation concentrations to using the RfC methodology and/or physiologically-based pharmacokinetic (PBPK) models. As OPP continues to work on refining the risk assessment paradigm, the Agency is seeking the SAP's input on a number of key factors. They include the use of oral toxicity studies when inhalation studies are not available and the use of aerosol inhalation toxicity studies to represent toxicity to vapors of

the same chemical. Specifically, the Agency identified the following issues for the Panel to consider:

B.1) The analysis conducted by the Agency indicates that, in general, oral toxicity studies may not accurately represent the full spectrum of toxic effects that may occur as a result of inhalation exposure. The analysis also indicates that - unless the same endpoints are identified through both routes of exposure - oral toxicity studies frequently underestimate toxicity by the inhalation route. The Agency has not been able to discern any patterns in this under/over estimation. *Please comment on any potential patterns that the Agency has not identified.*

Panel Response

Route-to-Route Extrapolation Issues

The Panel noted that EPA placed significant emphasis in the background document on the ability to use toxicological data generated from oral exposures to establish Inhalation Equivalent Concentrations (IECs) even though they recognized that such a strategy is seldom valid. Nonetheless, the Panel evaluated this approach and recommended that if such extrapolation is considered that the guidelines as outlined in *Principles of Route-to-Route Extrapolation for Risk Assessment* should be followed (Gerrity and Henry, 1990) and only for those pesticides that meet the criteria noted below. One Panelist commented that this book was an outgrowth of a workshop sponsored by the EPA in March 1990. Many concepts and issues raised in this book were also later addressed by Rennen et al. (2004), with essentially the same conclusions being reached. Basically, the only chemicals that are candidates for route-to-route extrapolation are those having no portal of entry effects and those whose kinetic behavior is independent of exposure route. The summary report section of Gerrity and Henry (1990) provides a decision tree for assessing how the information available for a given chemical can be used to identify the path forward and ascertain if sufficient data are available to attempt any route-to-route extrapolation.

Review of the available literature evaluating route-to-route extrapolation indicates that, for the most part, there is no pattern of consistency in the results when oral toxicity data are used to model inhalation toxicity for most chemicals. Extrapolation results appear to be somewhat random, with oral to inhalation extrapolation resulting in either over-estimation or under-estimation of inhalation toxicity depending upon the specific model approach used. The only chemical class in which there appears to be some consistency is systemic toxicants that have long half-lives in the body.

The Panel concluded that route-to-route extrapolations using oral toxicity data to assess toxicity via the inhalation route is only scientifically justified, if a validated PBPK model is available or if the pesticide falls into Category 3 chemicals according to the Agency's Inhalation Reference Concentration (RfC) classification scheme (*i.e.*, those gases/vapors that cause systemic effects and not point-of-entry effects) (EPA, 1994) and the following criteria are also met.

a. The toxicological effect of concern is systemic for both entry routes and this effect is independent of route of exposure.

b. The first pass effects in the liver for oral exposure and in the respiratory tract for inhalation exposure are minimal to nonexistent or, if there is some first pass effect, the metabolism following exposure is the same for both exposure routes.

c. The chemical will not be chemically modified by the gastrointestinal bacteria or enzymes or by the acidic environment in the stomach differentially than it would be in the more neutral to slightly basic environment of respiratory tract fluids. Pesticide stability in gastric fluid should be of primary concern in considering the use of oral exposure data. If the compound is not stable in gastric fluid, the toxicological data on oral dosing would be of little to no value in estimating any inhalation toxicity.

d. The absorption efficiency for oral and inhalation exposure must either be identical or known, so that accurate values may be incorporated into any model. The absorption efficiency for oral and inhalation exposure must either be identical or known, so that accurate dose values may be incorporated into any model; the use of a default value of 1 for absorption is not justifiable in the absence of any data. Furthermore, the absorption cannot be differentially influenced by a toxic response unless this response is the same via both routes and/or is influenced by relatively the same extent via both routes.

Even when the above criteria are met, the Panel recommended an additional UF of 10 (as an example) for the final extrapolation for inhalation from oral data. The Panel stated that uncertainty factors are not a replacement for more accurate modeling and additional data from more inhalation toxicity studies. The Panel provided a detailed discussion as to why it is scientifically justifiable to request additional inhalation toxicity data. Discussions and data presented during the meeting clearly show that repeated exposures to pesticides are occurring. Based on this discussion, the Panel strongly recommended that the Agency obtain additional inhalation toxicity data if such data are not available and if the vapor form of the pesticide does not meet the criteria for route-to-route extrapolation as described above.

Use of *In Vitro* Studies

The Panel noted that some of the problems associated with route-to-route extrapolation from oral toxicity studies to inhalation toxicity studies might be evaluated by simple *in vitro* studies, such as solubility and stability in simulated gut or lung fluid (as discussed in more detail below), or by consideration of the chemical structure of the pesticide and using structural activity relationships. For example a pesticide may be very stable in a synthetic lung fluid, which has a much higher pH than a human gastric fluid. Pesticide instability in a gastric fluid would therefore suggest a much lower toxicity through ingestion than inhalation.

Solubility of the inhaled chemical within lung fluid is the first step in the toxicological process and can be modeled using a synthetic lung fluid (Dennis *et. al.*, 1982; Eidson and Griffith, 1984). This parameter may be assessed as part of a screening process. Similarly, synthetic gastric fluids may be used as a screen for considering not just bio-solubility, but also pesticide stability through the ingestion pathway. Both of these solubility tests can also be used to screen

particulates as well. Finally, if a route-to-route exposure model for inhalation of vapors is to be evaluated, then a more appropriate dosing model might be injection rather than ingestion.

The Panel provided a hierarchy of *in vitro* models that could be used for assessing bioavailability in humans of any pesticide. The overall delivered dose can be approximated by animal exposure studies. Cell culture experiments can potentially be used to predict target organ processes. Dependent on dose, they can be used to evaluate cell death, mutation or, at lowest dosing levels, metabolism, but not solubility or even stability because the fluids used to stabilize the cells do not always accurately represent the fluids interacting with the target organ. The next level in the hierarchy would be determining the bioaccessibility of the pesticide. In these studies, the soluble component must pass through a semi-permeable membrane that would approximate the actual target tissue of interest. The final level would be creating an estimate of the biosolubility of the tested compound. This can be done using simulated biological fluids. Use of simulated biological fluids has the potential to correct for interspecies differences because the biological fluid can be created using a formulation (or recipe) based on ratios of constituents that most closely approximate that occurring in humans or animals used in exposure testing above. For the bioaccessibility and biosolubility *in vitro* models, the physiological endpoint being approximated is dose delivered to the next compartment, generally the blood.

Equation for IEC

The EPA background document presents an equation for calculating an IEC that attempts to take into account various factors, such as minute ventilation, animal body weight, exposure duration, absorption efficiency, etc.; however, the Panel noted two problems with this equation. First, species differences in surface area/body weight are not accounted for, so that the equation needs to be modified. In cases where route-to-route extrapolation may be justified, *i.e.*, pesticides that fit into EPA's RfC Category 3, it would be better to adjust the animal oral dose to a human equivalent oral dose (*i.e.,* adjustment based on body weight raised to the ¾ power (EPA, 2005) before further adjusting this value to an IEC for humans. The Panel stressed that the use of an IEC equation should be restricted to cases where the vapor falls into EPA's RfC Category 3 and also meets the four extrapolation criteria listed earlier.

NOAELs vs. BMDs

The EPA background document mainly focuses on the use of No Observed Adverse Effect Levels (NOAELs) and Lowest Observed Adverse Effect Levels (LOAELs) as bases for calculating IECs or Human Equivalent Concentrations (HECs). The Panel noted that the value of the NOAEL or LOAEL is sensitive to the nature of the experimental design used to conduct the study and, therefore, recommended that the Agency consider Benchmark Dose (BMD) analysis when concentration-response data are available and are amenable to modeling. The Panel added that because BMDs can be established for various percentages of effects, the Agency would have various regulatory management options to consider that reflect magnitude of risk. The Agency could then select the option(s) that most closely align with their specific risk management goals.

ATTACHMENT D, Pt. 2

Use of Haber's Law in Duration of Exposure Adjustments

The Panel stated there were several problems with the way in which EPA used Haber's Law (or Rule), in making adjustments for the duration of exposure when calculating HEC's from repeated exposure inhalation toxicity studies. As stated on p. 59-60 of the Agency background document, "Thus, application of this procedure provides an automatic margin of protectiveness for chemicals, for which C_{max} alone may be appropriate, and it reflects the maximum dose for agents for which total or cumulative dose is the appropriate measure."

First, Haber's Law has been shown not to be applicable to a great number of toxicological responses. Miller and colleagues (2000) showed that this Law is merely a special case of the generalized power law family. Basically, one arrives at Haber's Law when $\alpha = 1$ and $\beta = 1$ in the generalized power law equation, given as $C^{\alpha} x T^{\beta} = k$, where C is exposure concentration, T is the duration of the exposure and k is a fixed level of effect.

Second, whether the use of Haber's Law provides an automatic margin of protectiveness is entirely dependent upon the values of α and β in the power law family of curves. Miller *et al.* (2000) illustrated these differences (see Figure 9 in this paper). Figure 1 below is a reproduction of Figure 9 in Miller *et al.* (2000). For example, when $\alpha > 1$ and $\beta < 1$, the use of Haber's Law to extrapolate from high to low level exposures actually results in an under prediction of risk (see Case C, Figure 1). In contrast, the Panel indicated that the Agency is assuming that $\alpha > 1$ and $\beta > 1$, *i.e.*, Case D, Figure 1. During her presentation at the meeting, Ms. Annie Jarabek, EPA, Office of Research and Development (ORD), National Center for Environmental Assessment (NCEA), illustrated the issue of conservatism dependence as a function of where one is on the rectangular hyperbola relating concentration and time to a fixed level of biological response.

One Panel member noted that NCEA is in the process of developing software that can fit a variety of generalized power law family C x T models. These models would likely be of great interest to OPP, particularly those that relate to acute exposure modeling and those that account for $C^{N} x T$, where N captures the ratio of α and β (see EPA, 2008; page 10, document on the ten Berge models).

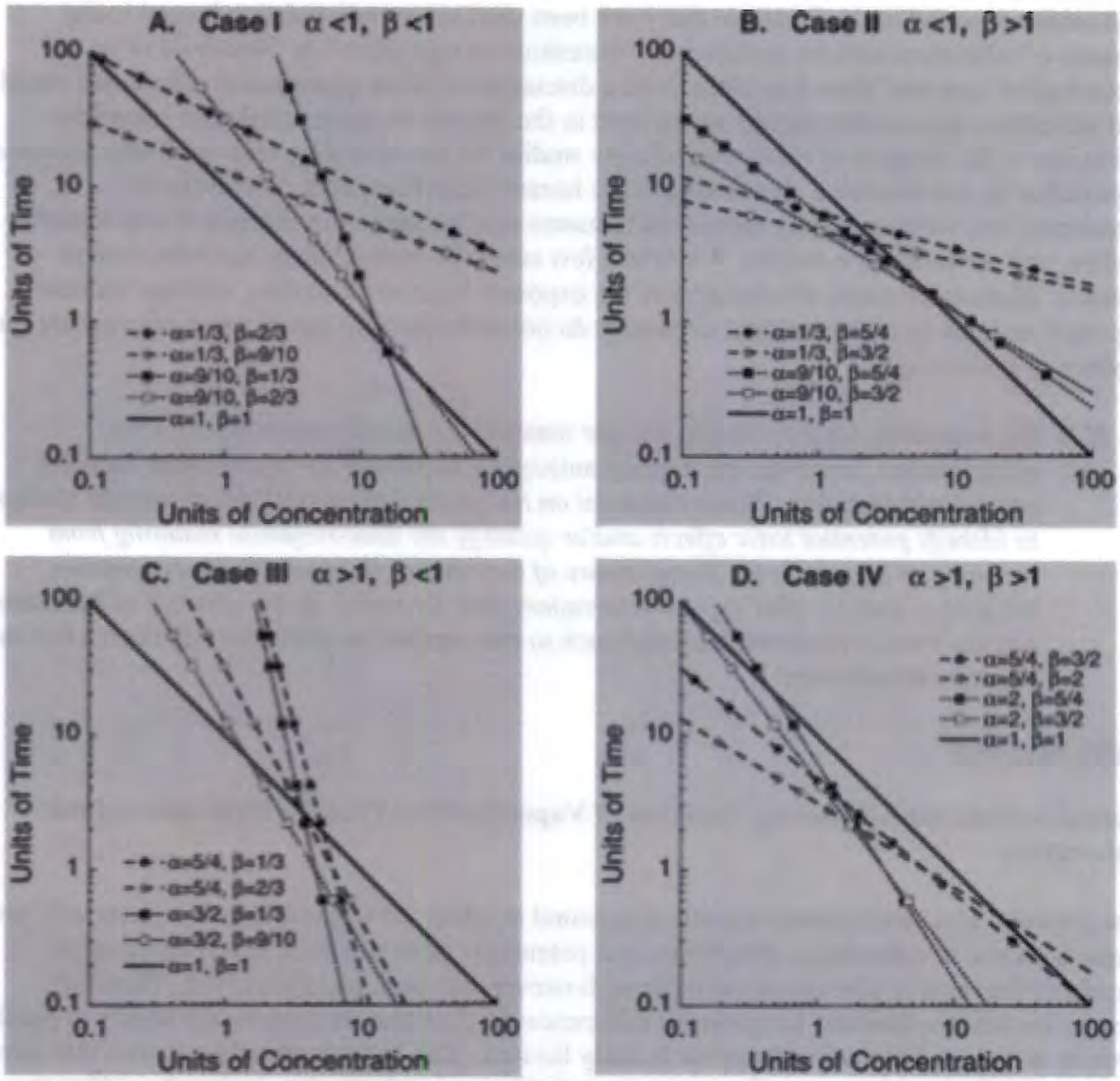

Figure 1. Log time-log concentration plots for the general power law family, $C^{\alpha} x\, t^{\beta} = k$. The panels depict the four combination of α and β. Included for reference is the line of identity (Haber's rule) corresponding to $\alpha - \beta = 1$. Here k is fixed at 10 for all plots. Reproduced from Figure 9 in Miller *et al.* (2000), with permission.

B.2) For a significant number of conventional pesticides, inhalation toxicity studies are not available. *Please comment on the scientific strengths and weaknesses of available approaches that may be used in the interim to assess inhalation hazard in the absence of inhalation toxicity studies.*

Panel Response

Overall, the Panel strongly recommended that the Agency conduct inhalation toxicity studies to adequately assess inhalation hazard. One Panel member provided a summary of alternative

approaches reported in the literature that have been used to assess inhalation hazard in the absence of inhalation toxicity studies, *e.g.*, "threshold of regulation" or "threshold of no toxicological concern" (see Appendix C for a discussion of these procedures). The Panel stated that alternative approaches should not be used in the interim to assess inhalation hazard for pesticides in the absence of inhalation toxicity studies for the following reasons. These screening approaches do not involve a chemical-specific hazard identification or dose-response assessment and were originally developed to assess oral hazard in the absence of oral toxicity studies, and not inhalation hazard. While in a few cases the methodology has been used to evaluate inhalation hazard, the duration of the exposure in these cases (*i.e.*, chronic, lifetime exposure and one-hour intermittent exposure) do not correspond to the duration of exposure the Agency is evaluating.

B.3) For inhalation toxicity studies the test material is typically aerosolized. After volatilization, however, the Agency anticipates exposures to vapors rather than the aerosolized particles. *Please comment on the predictive capabilities of aerosol studies to identify potential toxic effects and/or quantify the dose-response resulting from exposure to vapors. Is the Panel aware of any studies that quantitatively compare inhalation toxicity after exposure to vapors and aerosols? In the absence of such data, can the Panel recommend an approach to account for the potential differences between vapors and aerosols?*

Panel Response

Considerations for Addressing Question of Vapor/Particle Toxicity Differences in the Laboratory

The presence of a semi-volatile organic compound in either the vapor or particle phase will have impacts on site of deposition, absorption, and potentially dose and toxicity. The biological impact of the relative portions in each phase, however, has been poorly studied. Therefore despite known mechanisms for potential differences in dose and absorption, the ability to predict toxicity of vapors from aerosol studies is fairly limited. The Panel knew of no studies that have investigated the health impact of exposure to a single semi-volatile chemical under different phases. Some studies with mixtures have utilized techniques to either remove the particulate phase by filtration or the vapor phase by using a denuder to study the role of those materials independently (McDonald *et al.*, 2007). In many cases, the removal of the particulate phase of the mixture has not resulted in biological effects that differ from those obtained with the total mixture, especially for systemic effects. These mixture studies, however, may not be appropriate to answer questions related to a single component study. The Panel stated that there remain fundamental questions (beyond just the deposition site) related to the relative toxicity of vapors and particles. These must be considered as adding uncertainty when attempting to predict biological effects from exposure to the vapor versus particulate form of any given pesticide.

In the case of semi-volatile organic compounds (SVOC), chemicals can exist in the gas and particle phase simultaneously. The relative proportion in either phase will depend on concentration, temperature, humidity, and other particulate matter in the air. Volatile flux from crops will result in concentrations that are substantially below the saturation vapor pressure of

that imparts conservative calculations (*i.e.*, for the range cited here, one would use 50%) and then apportion the uptake across the three major respiratory tract regions. For example, if most of the vapor is taken up in the ET region and some is delivered to the TB region but very little reaches the alveolar region, then the total uptake might be apportioned as 40% ET and 10% TB.

4. Use the appropriate ventilation data for the animals in the pesticide aerosol study and compute what vapor phase exposure would be needed to yield the same value of the dose metric as was computed in Step 2. One may well find that the region for uptake is different than the region where the pesticide aerosol is primarily deposited. However, since the cells lining the non-olfactory epithelia in the ET and TB regions are reasonably similar (Miller *et al.*, 2010), a mass per unit surface area dose metric is likely to still be an acceptable metric.

5. Next, make similar calculations for humans as was done in Step 5 to see what levels humans would have to be exposed to for what periods of time in order to achieve the same numerical value of the dose metric calculated for the animals.

6. Now move forward with RfC or HEC calculations as inputs to the assessment of risk.

The Panel noted that this procedure is a possible way to obtain information on the vapor form when only aerosol exposure studies are conducted. The modeling approach is intended to help provide some interpretation of the findings from the aerosol study in assessing potential effects from exposure to the vapor. The Panel briefly discussed this procedure and thought it was reasonable; however, they recommended that EPA assess it further to determine its ultimate utility.

ADDITIONAL DISCUSSIONS ON INHALATION TOXICITY

The Panel discussed various topics that were an outgrowth of the charge questions. The order of the topics is not in order of priority.

Appropriate Averaging Times and Sampling Devices for Field Measurements

Currently, measurements of pesticide levels collected from fields where crops have been treated with pesticides are based upon 24-hour samples. During the public comment period, Dr. Susan Kegley of the Pesticide Research Institute presented data showing significant variability in air concentrations of volatile pesticides over periods of time shorter than 24-hours. Many individuals and families live either alongside of crop fields or even within orchards. Thus, there are significant opportunities for both children and adults to be repeatedly exposed to volatilized pesticides. Moreover, given the large variability in exposure levels in the first hours following pesticide application and then the subsequent temporal variability, the Panel recommended that the Agency collect exposure data with shorter collection times than 24-hours and to use these data in health effects evaluations. For research purposes, samples collected over 2- to 4-hour periods will likely be needed to assess the variability of the pesticide concentration in the air soon after pesticide application and over the next few days.

Appropriate sampling devices for the above studies would be able to separate the vapor form from the particulate form of the pesticide, or at a minimum ensure that both the vapor and particulate portion of the atmosphere are included. This can be accomplished with denuders and filters, or a combination of filters succeeded by vapor sorbents. Samplers with automatic switching manifolds to permit high frequency sample switching are commercially available. Given the activities in which workers must engage, there is a strong likelihood that individuals surrounding the fields and the workers themselves are exposed repeatedly to both respirable pesticide particles and vapors. The Panel recommended that the Agency collect exposure data for both, so that the relative contribution of each to overall exposure may be assessed.

Protocols for Inhalation Toxicity Studies

The Panel recommended that inhalation toxicity studies should be conducted with an exposure duration of up to 90 days. For example, an inhalation toxicity study conducted at 1, 7, 14, 28, and 90 days of exposure would generate an adequate database to address both acute and subchronic exposures. The length of daily exposure may be guided by what is seen in monitoring studies when collection times shorter than 24-hours are used. These timeframes are reflective of the duration of human exposure to respirable particles and vapors via inhalation. The Panel recommended that the length of exposure per day should be guided by new field studies that are conducted to assess the variability in air concentration of the pesticide particles and vapors.

The earlier time points (*i.e.,* 1, 7, 14, and 28 days) would tend to capture the initial application period for the pesticide, any potential reapplications due to weather conditions, as well as additional weekly applications of a pesticide. The Panel indicated that the 90-day time point would provide a link for using a subchronic study to assess potential effects due to chronic exposure effects, with perhaps an UF added to adjust for not having chronic exposure data. The Panel pointed out that results of the subchronic studies may guide the potential requirement of chronic studies that intend to investigate long term effects such as cancer..

In addition, the studies should consider the likely types of biological effects that might be expected for the given type of pesticide. The Panel pointed out that the Agency should consider that new toxicity studies take advantage of the scientific advances in understanding inhalation toxicity and exposure assessment that have evolved over the past 30 years.

Overall, the appropriate approach for the inhalation study will be one that, to the extent possible, mimics human exposure conditions. The Panel noted that new vapor inhalation studies for semi-volatile compounds may be limited, perhaps, to cases where there is a predicted level of flux above a certain threshold. This may be determined, as discussed above, through flux modeling derived through fate/transport studies.

Uncertainty Factors for Quality of the Database and What Constitutes a Minimum Database

Other EPA offices (*e.g.,* Office of Water, Office of Research and Development, Office of Air and Radiation) and regulatory programs have established criteria for what constitutes a minimum

database for chronic exposure that must be available so that a health assessment, an inhalation RfC for chronic exposure, an oral RfD, etc. can be determined (*e.g.*, see EPA, 1994). The Panel recommends that EPA establish such criteria for short-term studies (*e.g.*, 1, 7, 14, 28 days) and that they use an additional UF of 10, if only a minimum database is available for their assessment.

Moving Toward a Cumulative or Total Risk Assessment

The Agency's background document notes that potential pesticide exposures can occur from three types of scenarios: during application directly, due to application drift and due to volatilization following application. Furthermore, exposure may be to respirable particles, or vapors, or both. While the background document is only concerned with volatilization of pesticides following application, the Panel noted that a broader view is going to be needed for the future. Therefore, the Panel recommended that total exposure be assessed to more fully encompass all types of inhalation exposures for the risk assessment process. For example, the Panel acknowledged the difficult situation that was occurring to crops on the Jacobs Farm (oral comments provided by Mr. Larry Jacobs, Pescadero, CA and owner of Jacobs Farm). They concluded that the situation on Jacobs Farm demonstrates that there is a problem with volatilization and possibly re-entrainment of particulate pesticides. If this situation is due to such transport processes, then individuals should also experience pesticide exposures from volatilized pesticides and, possibly, respirable particles.

The Panel suggested that the Agency consider the hazard quotient (HQ) or similar approach to assess risk from each of the types of exposure that contribute to the total potential exposure following application of pesticides because, at a minimum, workers are exposed to respirable particles and vapors both during direct application and drift. Moreover, those living close to fields where pesticides are applied are likely to receive exposure from more than the volatilization route. Hazard quotients from different pathways (or from different pesticides) could be added together to calculate an overall hazard index [HI]. If the resulting HQ or HI is $\leq$ 1, then adverse health effects would not be expected. If the HQ or HI is slightly > 1, then it would not necessarily mean that health effects would occur but that further evaluation was warranted. If the HQ or HI is significantly > 1, then it would indicate health effects would be likely to occur. The hazard quotient would seem to offer a better alternative than a MOE approach to aggregate risk. Hazard indices for pesticides with the same mode of action could be used for assessing cumulative risk from inhalation exposure to multiple pesticides.

TOPIC C: Risk Assessment Issues

The Agency discussed its methodology for combining the exposure estimation methodologies and inhalation toxicological approaches to estimate postapplication bystander inhalation risks resulting from field volatilization of conventional pesticides. In estimating postapplication bystander inhalation risks, there are a few principles that should be followed: (1) properly match the duration of the exposure with a proper toxicity study of comparable duration; (2) both dissipation of air concentrations around a treated field as well as when retreatment of the field may occur, need to be considered; and (3) clearly define the uncertainties and limitations of this

type of assessment. The Agency has identified the following issues for the Panel to consider with respect to estimating postapplication bystander inhalation risks:

> *Please comment on the strengths and limitations of the Agency's use of the empirical and modeled air concentrations in the provided risk assessment case study. Does the Panel agree that the postapplication bystander inhalation risk estimate case study appropriately matches the duration of the exposure with the proper toxicological study of the same duration? Please comment on the scientific strengths and weaknesses of conclusions and characterization regarding the estimated risks presented in the case study.*

Panel Response

> *C.1) Please comment on the strengths and limitations of the Agency's use of the empirical and modeled air concentrations in the provided risk assessment case study.*

The strengths and weaknesses of the Woodrow empirical model are much the same as the Panel discussed above in Topic A, Question 2a. The main strength of this model is its basis in multiple studies over a wide range of vapor pressures; a secondary strength is that its results are in the range of the results of the air concentrations estimated by more sophisticated modeled air concentrations. The limitations of the model are that it is based on a limited range of crops, weather, and locations, does not take into account the potential effect of an activity coefficient on vapor pressure, and is applicable only to the first day post-application.

The strengths of the computer-based modeled air concentrations are that they can account for dynamic changes in post-application conditions and residue history. Their weakness is the limited knowledge that users have of the internal components of these models, the concern that the components do not model evaporation from foliage as well as they model evaporation from soil, and that the particular analyses presented to the Panel inappropriately decoupled the variance of the flux from the variance of dispersion.

Discussion of the Tier IIA Model (Woodrow *et al.* Model)

Several panelists again suggested that the accuracy of the Tier IIA, Woodrow *et al.* (2007) model's estimation of airborne pesticides concentrations could be more precise by adding more variables (see earlier discussion under Topic A, Charge Question 2a. The Woodrow model's basic structure of a log-log regression lends itself to multiple regression analysis. Two variables of initial interest would be the influence of the pesticide's application rate and foliage density on flux. Some panelists cautioned that worst-case scenarios could not be easily identified for this model because of the narrow set of conditions that were available and used when the model was originally developed by Woodrow *et al.* (1997).

On the other hand, one Panel member suggested that the utility of the Woodrow model could be expanded by using its predicted 24-hour flux rate in conjunction with a mass balance to derive a per hour flux rate. One such expansion could be derived by assuming that the flux rate decreases exponentially with time, as depicted in Equation C1, where τ is the exponential rate constant.

most vapors. Therefore, in most cases, re-equilibration will be defined not by concentration, but by other factors such as the presence of other particulate matter, and conditions of temperature and humidity. These other factors can be measured, and incorporated into models using gas/particle partitioning models (see Odum *et al*, 1996; Pankow *et al*., 1994). An additional exposure estimation concern includes the flux over time of the pesticides.

Depending on the vapor pressure of the compound studied, it is indeed plausible that significant amounts of vapors existed in the laboratory studies that were conducted to determine pesticide toxicity. The gas/vapor partitioning models cited above can be used to predict the fraction of compound in the gas and particle phase at a given environmental condition and concentration. For a simple case where only one compound is present, this model can utilize first principles. However, it should be noted that for a semi-volatile material where the aerosol was sampled with a filter, one could obtain inaccurate dose estimates due to potential volatilization of vapors from the filter and/or adsorption of vapors onto the filter.

Once actual exposure is more accurately defined, the appropriate exposure scenario can be developed. One issue is the fact that the majority of laboratory inhalation studies were done with aerosolized formulations or powders at high concentrations. These conditions do not represent the true physical form of the pesticide as it exists after volatilization at relatively low concentrations. The toxicologically relevant impact of this is that vapors may be 50-100 % absorbed in the respiratory tract; depending on the reactivity of the material, the vapors may or may not penetrate deep into the respiratory tract prior to removal. Studies with highly reactive and highly soluble vapors in rodents have shown that the biological response is limited to effects in the nasal passages and upper tracheobronchial (TB) airways. Particles may have more or less deposition (and, therefore, dose) than vapors, with the deposition amount and location in the respiratory tract dependent upon the size of the aerosol. Because of the aerosol generation approaches used in the past, the particles in the laboratory studies will likely be 2-3 micrometers and be polydisperse. In this case, the majority of the material will deposit deep into the respiratory tract, and at a much lower fraction than the vapors. Polydispersivity, however, will result in some overlap of deposition sites with those for vapors whose uptake sites can be predicted with reasonable certainty.

An approach to link aerosol data with vapor data would need to make the assumption that the form of the material will not impact absorption once it is removed from the air stream. Once more, the site of deposition would need to be considered, perhaps using currently available modeling approaches for particles and evolving modeling approaches for vapors. In this case, dose relationships between the studies can be established, and responses can be related in a more appropriate way. Once the site of deposition is better understood, another important consideration is the nature and type of biological effects observed. These may be able to be related to the site of deposition for local effects, and to bioavailablity/absorption for systemic effects. For compounds that show high absorption, the vapor and particle components of the aerosol will likely show reasonably similar systemic biological responses following deposition.

Potential Modeling Approach for Using Aerosol Studies to Assess Vapors

Based on the discussion during the meeting, the Panel understood that EPA has 25-40 year old aerosol pesticide studies that might provide some insight into the potential toxicity of the volatilized form. These studies essentially used the "neat" form of the pesticide, *i.e.,* a very high technical grade purity chemical. The Panel suggested a potential approach to examine these studies for useful toxicological information regarding the volatilized form of the pesticide:

1. The Multiple-Path Particle Dosimetry Model (MPPD) should be run for various mass median aerodynamic diameter (MMADs) and geometric standard deviation (GSD) combinations within the interval of MMAD greater than 2-3 μm and a GSD of 2-2.4, to establish variability of the predicted deposition. This will enable the Agency to assess the variability/sensitivity of the results obtained in subsequent steps. For example, if one uses the rat as the experimental animal, the MPPD model can be used with this input data to determine the amount of the pesticide aerosol deposited in the major regions of the respiratory tract (Anjilvel and Asgharian, 1995; National Institute for Public Health and the Environment (RIVM), 2002). The MPPD model is publically available for free and can be downloaded from http://www.ara.com/products/mppd.htm.

   The Panel added that EPA's Regional Deposited Dose Ratio (RDDR) model could be used for other animal species (EPA, 1994). In older aerosol studies, the MMAD and the GSD of the aerosol were not reported; however, the methods used, during the aerosol generation did not create particle distributions with a MMAD greater than 2-3 μm and a GSD of 2-2.4. Lack of reporting these values for these studies, therefore, was not a concern.

2. Because the MPPD model can provide estimates of various dose metrics (*e.g.,* mass per unit surface area, number of particles per alveolus, etc.), the next step is to determine which dose metric best fits or is likely to best correlate with the biological response of interest. For the extrathoracic (ET) region, the total mass deposited per unit surface area .is probably the only dose metric that can currently be calculated. To do this, one needs to use an estimate of ET surface area (see Ménache *et al.,* 1997) and the mass deposited in the ET obtained from running the MPPD model. MPPD provides the TB surface area for the size of the animal studied, but the alveolar surface area currently displayed by MPPD reflects only the surface area of the alveolar ducts. Miller *et al.* (2010) provide an algorithm to estimate the total alveolar surface area.

3. Use air: blood partition coefficients, solubility, Henry's law values, etc. to determine the likely regions of the respiratory tract in which the vapor form of the pesticide will deposit. For example, Overton and Jarabek (1990) show how this information can be used to identify likely respiratory tract regions (*e.g.*, ET and TB, alveolar) where specific gases and vapors will be deposited up following exposure.

   By examining various gas uptake studies published in the literature, the Panel thought that EPA would find that total respiratory tract uptake ranges between 50 to 100 % in laboratory animals. For whatever range is found, use the lower value of the interval, as

Photodegradation rates of pesticide vapors have been studied for several pesticides, (see review by Atkinson *et al.*, 1999). In some cases, photodegradation rates of pesticide vapors are quite significant, such as for those organophosphorus thions (*i.e.*, sulphur analogs) that are converted to oxons. However, the oxons are more toxic than the thions, and thus could contribute significantly to the hazard associated with the vapor during downwind drift. Therefore the chemical product of the photodegradation reaction must also be considered.

3. Volatilization dispersion prediction models should include scenarios with temperature inversions. Temperature inversions are common in some parts of the US whereby an air mass may be trapped and normal dissipation and dilution due to air mixing and ventilation are impeded. Inversions could create the potential for exposures to airborne pesticide concentrations that are higher and for a longer duration of exposure than expected.

   The Panel stressed that monitoring and modeling should be flexible enough to take into account unusual topographical, meteorological, and other environmental features that might affect exposures resulting from pesticide residues in the air. For example, temperature inversions are common in some parts of the United States. Temperature inversions in agricultural valleys or other topography can trap the air mass, and impede normal vapor dissipation due to dilution during air mixing and ventilation. This could create the potential for exposures to airborne pesticides that are higher than expected, and for longer than normal duration. In the San Joaquin Valley of California, an area of very high pesticide use including in the winter, wintertime inversions can lead to ground fog that: (a) traps a cool air mass, (b) modifies dissipation by photolysis and/or wind dilution, and (c) creates a partition phase (pesticide suspended in fog water droplets) that can concentrate some pesticides, aiding their deposition to non-target crop foliage, and potentially changing the airborne composition from one dominated by the vapor form to one that is aerosol-dominated. How processes such as this affect exposures of people to airborne pesticides need to be taken into account in designing monitoring and modeling programs.

4. The Panel recommended expanded pesticide use reporting and a national air monitoring network for pesticides to more accurately assess community exposure.

   The Panel recommended that more expansive pesticide usage data, as well as more regional air pesticide monitoring data, be generated to more accurately predict exposure by the public. For example, Panel members noted that the pesticide use reporting system in California has been extremely useful for research scientists and policy makers to increase the understanding of the environmental fate of pesticides. This type of system could also be used in other regions, especially where volatile or semi-volatile pesticide use is high. A standardized air monitoring network in agricultural regions of the US would also be extremely useful. These data could be used to further validate pesticide emission and atmospheric transport models and to estimate pesticide exposures to communities.

5. The Panel suggested a more careful evaluation of the literature on ambient air monitoring in agricultural communities, *e.g.*, Lee *et al.* (2002), Kollman (2002), as well as field volatilization flux measurements, and modeling, *e.g.*, Raupach *et al.* (1996).

   The Panel recommended that there be a broader review of the volatilization flux measurement literature. Studies exist that can be used to evaluate these models. A rich literature exists concerning flux measurement methodologies, uncertainties, and measurement requirements. For example, scientists such as Ralph Nash, Dwight Glotfelty, Alan Taylor, and William Spencer have carried out studies that discuss in detail the factors that influence the volatilization flux from soil and other surfaces (see "Recommended References"). Other studies report the volatilization flux of pesticides from flooded rice fields as contributors to measured airborne residues of pesticides in the Sacramento Valley, CA (Seiber *et al.*, 1989). Also recommended are papers by Scott Yates (USDA-ARS, Riverside) that include the use of field flux chambers for measuring volatilization in the field. Many of the field volatilization flux papers published before 1995 have been compiled in Majewski and Capel (1995). Rich literature also exists concerning transport within forest canopies. A selection of literature that the Agency might find useful is provided in the "Recommended References" list in the "Reference" section of this report.

   In the introduction of the background document, EPA presented citations to available air monitoring data collected both in population centers of agricultural communities or "ambient" levels and adjacent to fields or "application site" monitoring. Additional summary articles are available (Lee *et al.*, 2002). The California Department of Pesticide Regulations (CDPR) has also summarized near-field and ambient data available as of 2000 (Kollman, 2002; Tables 1-2). The Panel recommended that EPA present summary tables of all of these measured air concentrations for two reasons. First, these levels would be useful in describing the breadth and magnitude of potential public health concern for pesticide volatilization from agricultural fields. Secondly, these levels may be used, at every step in the process, to evaluate air modeling efforts.

6. The Panel recommended that the Agency gain a better understanding of the physics of the models they are proposing for this application. It was not clear whether the Agency was using a variable laminar boundary layer depth that is a function of evapotranspiration, as originally proposed by Jury *et al.*, (1983), or a constant laminar boundary layer depth. The Jury *et al.* (1983) proposal is not physically sound for the purpose used by the Agency. A constant laminar boundary depth layer is more physically realistic (*e.g.*, Panofsky and Dutton, 1984). PRZM appears to use a constant laminar boundary layer depth. The Panel was not yet able to ascertain the treatment of this issue in PEARL. The Agency's background document did not clarify this issue.

   The models proposed (PRZM and PEARL) appear to assume a zero atmospheric mixing ratio and to neglect aerodynamic resistance when estimating fluxes, which should tend towards an over-prediction of the flux. This should be kept in mind when evaluating the models. PRZM documentation described assumptions concerning the aerodynamic resistance, but the use of this resistance in the model was not clear. PEARL documentation was difficult to obtain. Thus it was difficult to evaluate the assumptions

$$\text{flux rate at any time} = \text{initial flux rate H } e^{-t/\vartheta} \qquad \text{Eqn. C1}$$

For example, if the application rate were two pounds per acre (using an "eyeball" average of the application rates in Table 1 of Woodrow *et al.* (1997) and taking into account that 1 kg/ha = 1.03 lb/acre, then the initial residue would be equivalent to an average deposition of 0.22 g/m² across a flat field. For a pesticide with a vapor pressure of 1 x 10^{-4} mm Hg, the Woodrow model (translated in Equation C2 out of its log-log format) would predict an initial 24-hour flux rate of 0.0032 /m²/hr.[3]

$$\text{Flux}_{day\ 1} \text{ in Woodrow model} = 8.574 \times 10^{6} \times P_{vapor}^{\ 0.85543} \qquad \text{Eqn. C2}$$

One can use Equation C3 to calculate this flux is equivalent to a rate coefficient [ϑ] of 2.3 days or an exponential half-life of 1.6 days (where HL = ϑ H ln2).

$$\vartheta = \frac{t\ (or\ 1\ day\ in\ the\ Woodrow\ case)}{\ln\left[\frac{initial\ residue}{initial\ residue - (24 \times flux_{24-hr})}\right]} = \frac{1}{\ln\left[\frac{0.22}{0.22 - (24 \times 0.0032)}\right]} = 2.33 \text{ days} \qquad \text{Eqn. C3}$$

The average exposure within any chosen interval of duration from application to time *t* can be calculated by using this half-life within Equation C4.

$$\text{average over span "}t\text{"} = \text{initial flux H} \frac{\int_0^t \exp(-t/\tau)\, dt}{t} = \text{initial flux H} \frac{\tau \times (1 - \exp(-t/\tau))}{t} \qquad \text{Eqn. C4}$$

The first part of Table C1 presents a range of examples of these averages as a fraction of the initial value. The second part of Table C1 presents the ratios of the average within the designated interval to the average within the first 24-hours. The Panel commented that in the context of other short-term toxicological effects, it is notable that the maximum ratio between the first hour and the full 24-hours never exceeds 2-fold, at least within the range of half-lives examined. (Note: For this example, these values are smaller than the values given at the meeting.) Table C2 presents the instantaneous flux rates at the same points in time just for information. Again, the Panel pointed out that these extrapolations should be viewed only as a potential extension of the Woodrow model (a Tier IIA model) and should not be viewed as a replacement for estimations of flux rates from the more comprehensive models in Tier IIB. For instance, exponential dissipation based only on evaporation would over-estimate vapor exposures if the pesticide also dissipated into the soil and decayed by other mechanisms.

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[3] Equation C2is a simplification of Equation 4 in the Agency's background document for flux from foliage based on the following generic logic:
$\ln(flux) = A + B \ln(C \times P_{vapor}) = A + B \ln(C) + \ln(P_{vapor}^{\ B})$
$flux = e^{A + B \ln(C) + \ln(Pvapor)} = e^{A + B \ln(C)} \times e^{B \ln(Pvapor)} = e^{A + \ln(C)} \times P_{vapor}^{\ B}$

Table C1. Results of using an exponential decay model to extrapolate from the average flux values in the first 24-hours to the average over various other time intervals of hours (or days), "t" in Equation 8C.

Average within the interval as a fraction of the instantaneous initial value.

			interval of the average in hours					(7-days)	(21-days)
		τ in hours	1	2	4	8	24	168	504
HL =	0.5	17.312	0.972	0.944	0.893	0.801	0.541	0.103	0.034
in days	1	34.625	0.986	0.972	0.944	0.893	0.721	0.204	0.069
	2	69.249	0.993	0.986	0.972	0.944	0.845	0.376	0.137
	3	103.874	0.995	0.990	0.981	0.962	0.893	0.496	0.204
	4	138.499	0.996	0.993	0.986	0.972	0.918	0.579	0.268
	5	173.123	0.997	0.994	0.989	0.977	0.934	0.640	0.325
	10	346.247	0.999	0.997	0.994	0.989	0.966	0.792	0.527
	20	692.494	0.999	0.999	0.997	0.994	0.983	0.888	0.710
	30	1038.740	1.000	0.999	0.998	0.996	0.989	0.923	0.792

Ratio of the average value within the interval to the 24-hour average.

			interval of the average in hours					(7-days)	(21-days)
		τ in hours	1	2	4	8	24	168	504
HL =	0.5	17.312	1.796	1.746	1.650	1.480	1.000	0.190	0.063
in days	1	34.625	1.366	1.347	1.309	1.238	1.000	0.283	0.095
	2	69.249	1.175	1.166	1.150	1.117	1.000	0.445	0.162
	3	103.874	1.115	1.109	1.099	1.078	1.000	0.555	0.229
	4	138.499	1.085	1.081	1.074	1.058	1.000	0.631	0.291
	5	173.123	1.068	1.065	1.059	1.047	1.000	0.685	0.348
	10	346.247	1.034	1.032	1.029	1.023	1.000	0.820	0.545
	20	692.494	1.017	1.016	1.014	1.012	1.000	0.903	0.723
	30	1038.740	1.011	1.011	1.010	1.008	1.000	0.934	0.801

Table C2. Results of using an exponential decay model to extrapolate from the average flux values in the first 24-hours to the instantaneous flux at various other time intervals of hours (or days).

Fractional value at the end of each time period

			interval in hours					(7-days)	(21-days)
		τ in hours	1	2	4	8	24	168	504
HL =	0.5	17.312	0.944	0.891	0.794	0.630	0.250	6.1E-05	2.27E-13
in days	1	34.625	0.972	0.944	0.891	0.794	0.500	0.008	4.77E-07
	2	69.249	0.986	0.972	0.944	0.891	0.707	0.088	0.001
	3	103.874	0.990	0.981	0.962	0.926	0.794	0.198	0.008
	4	138.499	0.993	0.986	0.972	0.944	0.841	0.297	0.026
	5	173.123	0.994	0.989	0.977	0.955	0.871	0.379	0.054
	10	346.247	0.997	0.994	0.989	0.977	0.933	0.616	0.233
	20	692.494	0.999	0.997	0.994	0.989	0.966	0.785	0.483
	30	1038.740	0.999	0.998	0.996	0.992	0.977	0.851	0.616

Ratio of the average value within the interval to the average 24-hour value.

			interval in hours					(7-days)	(21-days)
		τ in hours	1	2	4	8	24	168	504
HL =	0.5	17.31	3.78	3.56	3.17	2.52	1.00	0.000	9.09E-13
in days	1	34.62	1.94	1.89	1.78	1.59	1.00	0.016	9.54E-07
	2	69.25	1.39	1.37	1.33	1.26	1.00	0.13	0.00
	3	103.87	1.25	1.24	1.21	1.17	1.00	0.25	0.01
	4	138.50	1.18	1.17	1.16	1.12	1.00	0.35	0.03
	5	173.12	1.14	1.14	1.12	1.10	1.00	0.44	0.06
	10	346.25	1.07	1.07	1.06	1.05	1.00	0.66	0.25
	20	692.49	1.03	1.03	1.03	1.02	1.00	0.81	0.50
	30	1038.74	1.02	1.02	1.02	1.02	1.00	0.87	0.63

Discussion of Tier IIB Flux Models

A summary of a number of suggestions made by different Panel members regarding the use of flux models is provided below.

1. Several Panelists suggested that the Agency use the volatilization flux models to explore an array of crop and use patterns and to explore multi-field applications and multiple applications of a given pesticide within a field. One variation on this theme was the idea of using the models to develop "sentinel" worst-case scenarios. Because worst cases can vary, an array of exposure scenarios should be explored stratified by chemical, crop, and region. An assumption of a unit flux should be used with an appropriate air dispersion model, or models, and representative historical meteorological data to identify worst-case scenarios and predicted vapor concentrations in air. Predicted concentrations for substances with fluxes not equivalent to unity can be estimated by multiplying the unit flux predictions by a given substance's flux. In the absence of information on particular pesticide fluxes, a literature review of losses over say 24-hours (*e.g.,* Smit *et al.*, 1998) may permit the identification of a reasonable worst case assumption of the proportion volatized that can be used to estimate the flux over the same time period.

2. The Panel believed that the way the models were used in the Agency's case studies was inappropriate. For example, PERFUM is a model that was developed and field validated to estimate fumigant volatilization and downwind movement and concentrations under typical fumigation field conditions, *i.e.*, flat, fallow fields, usually covered with plastic tarps. This model was not, as far as the panel members were aware of, validated for any other class of pesticide or other field conditions. Not only was this model used to evaluate semi-volatile pesticides with very different physicochemical properties than fumigants, it was also applied to field environments that were radically different (orchard and cabbage fields) from the typical fumigation field (fallow, flat). The only way this model, or any other for that matter, can be reliably used to predict source volatilization flux and downwind air concentrations is to field validate them under the typical field conditions the various pesticides in question will be used.

3. The confidence in using the PERFUM model for semi-volatile pesticides can be increased considerably by conducting several field volatilization/model evaluation studies on select semi-volatile pesticides used on a range of crop types (*e.g.*, cover crop, short row crop, tall row crop, orchard) in representative geographic and climactic areas (or major agricultural areas) where the selected semi-volatile pesticides will normally be used. The field and modeling data from these studies can then be further evaluated by applying/using the results from one study to each of those areas where the other studies were conducted, as was done in the presented case study. Then those results can be compared to the actual field and modeled results. This will show almost immediately if this kind of exercise has any validity. If the results show promise, then the models can be further refined/fine-tuned, and subsequent predictions of the model on other semi-volatile pesticides in other field situations can be better defended because the model has been rigorously tested and compared against a variety of actual field situations.

4. Another suggestion was to prioritize risks by starting with an acceptable concentration. This suggestion was independent of prioritizing based on use of the Vapor Hazard Ratio (VHR) discussed below.

5. Several panelists reiterated an earlier point made in response to Topic A to not decouple the flux and dispersion models. [4] Decoupling probably increased the variability in the outcome of the examples presented in the Agency background document because of the covariance between the effects of many of the same variables that affect both outcomes.

C.2) Does the Panel agree that the postapplication bystander inhalation risk estimate case study appropriately matches the duration of the exposure with the proper toxicological study of the same duration?

The Panel agreed that the case study appropriately matched the duration of the exposure with the proper toxicological end point, although there were some questions regarding the specificity of the target population within this particular case study. Again, a great deal of the Panel's discussion on Topic B apply here, particularly the limitations of the route-to-route extrapolation process and the need to apply toxicological data collected from one duration of exposure to another. Thus, the success of this case study to achieve its goal is tempered by the limited confidence that the Panel has in extrapolating toxicological data considering both route and duration of the exposure.

However, in a broader sense, the Panel agreed that the example did not adequately consider the ability to model differences in duration of the exposure. In earlier sections, the Agency presented three types of exposure scenarios for risk assessment, short- (up to 30 days), intermediate- (up to 90 days) and long-term (greater than 1 year). The example only presented modeled air concentrations for short-term exposures. The models did not estimate intermediate- and long-term exposures. Two of the five pesticides listed in Table 3 (of the Agency's background document) used for modeling short-term exposures have reported soil half-lives of greater than 150 days, illustrating that intermediate exposures from single applications may occur. The Panel recommended the Agency consider adding longer-term exposures of more than 30 days in the models to address intermediate and long-term chronic exposures and to match toxicological studies. People who live in agricultural communities for their entire lives may be exposed to volatilized pesticides that may pose chronic, life-long health risks.

The Panel had a number of points regarding the uncertainty of matching the duration of the exposure with the proper toxicological study of the same duration. A brief summary of these remarks is presented below. Most of these points were made previously during the Panel's response to charge questions in Topic B.

1. Several Panelists discussed uncertainty in the specifics of the exposed population used

[4] By using the term, "decoupling," the Panel was referring to running a Monte Carlo simulation on a range of weather conditions to determine an array of flux values via PRZM or PEARL. Subsequently, a statistical percentile from that array is used as an input to PERFUM which is then used to run a separate, but statistically independent Monte Carlo simulation on the same range of weather conditions.

within the Agency's case study and the implications to the appropriateness of the UFs that were applied to address concerns about children's protection. The Panel indicated that the answer to the charge question depends both upon the UFs that were applied and the quality of the toxicological data such as the inclusion of developmental toxicity studies or studies conducted during critical life stages (i.e., children, teenage years) in the database. A new suggestion that may have broad applicability to the Agency was to view developmental/reproductive oral studies as informative in terms of effects, but not to be used quantitatively in terms of inhalation toxicity. In oral studies, if developmental, reproductive, or childhood effects occur at significantly higher concentrations than the critical effect, this would provide qualitative information on whether these effects may occur at low concentrations after inhalation exposure.

Toxicology for Risk Assessment (TERA) organized three peer consultations, two in 2005 and one in 2007, on issues related to risk assessment for children. One of the 2005 consultations, together with the follow-up in 2007, addressed issues related to toxicokinetic differences between adults and children, and the second 2005 consultation addressed issues related to the adequacy of the database uncertainty factor: These documents are available at http://www.tera.org/peer/AdultChildTK/ACTKWelcome.htm.

2. The Panel reiterated that all of the previous discussion in Topic B regarding the applicability of Haber's law applies to this case study. In the end, without other inhalation study data, the Panel thought it was difficult to tell for most pesticides whether a toxicity study conducted at one duration of exposure extrapolated to another is appropriate as previously discussed.

3. Similarly, the Panel reiterated its discussion in Topic B regarding the limitations of route-to-route extrapolation.

4. One Panelist strongly suggested that the term "RfC" not be used to describe "reference concentration" if an oral toxicity study is used in a route-to-route extrapolation of inhalation toxicity. The term reference concentration is typically used for values where UFs have been applied to the appropriate point-of-departure; therefore, it would be inappropriate to use this term to describe a value that has not been adjusted for uncertainties in the data.

5. Another Panelist commented that the Agency should consider the advantages of setting a fixed value for the Margin of Exposure (MOE) before completing the exposure assessment versus evaluating the quality of the MOE that results from such an assessment.

6. In the context of future assessments of inhalation hazards, the Panel was interested in how the MOE approach could be used to combine the three routes of exposure via volatilization, spray drift, and respirable particles to assess cumulative or aggregate inhalation risk. Alternatively, the Panel suggested that it might be easier to calculate a Concentration of Concern (CoC) (which incorporates UFs) first and then calculate the hazard quotient (HQ), which is the ratio of the breathing level IE to the CoC. Hazard

quotients from different pathways (or from different pesticides) could be added together to calculate an overall hazard index (HI). For additional information, HQs from different pathways (or from different pesticides) could then be added to calculate a HI. If the resulting HQ or HI is ≤ 1, then adverse health effects would not be expected. If the HQ or HI is slightly > 1, then it would not necessarily mean that health effects would occur but that further evaluation was necessary. And if the HQ or HI is substantially > 1, then it would indicate health effects would be likely to occur. The hazard quotient would seem to offer a better alternative than a Margin of Exposure (MOE) approach to aggregate risk.

C.3) Please comment on the scientific strengths and weaknesses of conclusions and characterization regarding the estimated risks presented in the case study.

The Panel broadly agreed that the case study included all or most of the important elements to conduct a proper risk assessment. The strength of the inhalation toxicity and exposure data bases for the chemical chosen led the Panel to conclude that the inhalation hazard and exposures assessments, and MOE analysis were realistic based on field monitoring data. However, the Panel had a range of recommendations for how this model and the risk assessment process could be improved, and reservations, if such an analysis were applied to many other chemicals. For instance, the toxic endpoints are unlikely to be as strong for other chemicals, the details and general applicability of some steps in the process were not well-defined, PRZM or PEARL were not optimized for their application to the evaporation of semi-volatile pesticides, and the impact of the propagation of uncertainty and safety factors within the process on the final result is uncertain. An alternative risk assessment approach based on the VHR is presented below.

Strengths

The ability to predict the concentration of vapors emanating from pesticides sprayed onto fields and crops and to assess the risks to bystanders is challenging. The multi-component and multi-step models that predict exposure are not simple, and the toxicity data pertinent to these exposure patterns are rarely available. Given these challenges, the Panel was in broad agreement that the process being pursued by the Agency includes all the ingredients needed to characterize the risks of such vapor exposures. The Panel noted that the Agency did a credible job in the case study as presented. The Tier IIB flux models have the ability to integrate the variability in real world settings and to generate a distribution of exposures that can be useful in risk assessment. The magnitude of the variations in the results that were presented was internally consistent and seemed realistic. Also, the magnitude and distribution of the resulting MOEs were believable.

Weaknesses

In the larger sense, the Panel concluded that the case study is about as good of a result as this approach can achieve at the present time. The pesticide selected for this case study may be one of the few that have sufficient data to link exposures over different intervals with relevant toxicological data points. The Panel was not confident that this process could be extended to many other compounds because of the scarcity of data. The Panel concluded that filling the gap in vapor inhalation toxicity data and performing more field studies to validate the vapor flux and

dispersion models are at least equally important. However, filling the gap in data of vapor inhalation toxicity data would be more complex, take longer, and be more costly than conducting more field assessments (or utilizing more existing field studies) to increase confidence in the exposure values.

Several Panelists agreed that exposures as short as (or shorter than) 1 hour should be of interest to the Agency. As discussed in Topic A, such initial peak exposures should be derived either from a dynamic model or (preferably) from actual measurements over shorter intervals than has been the historic practice, and not from average conditions over longer intervals. However, it may turn out that exposures in the first hour are only about a factor of two greater than the averages in the first 24-hours (see Table C1, exposures predicted from an exponential decay model using Equation C4 above).

The Panel stated that the Agency should be more diligent in describing the uncertainty that surrounds each variable within the PRZM and PEARL models. The Panel provided several ways to better understand the uncertainties in each variable and suggested opportunities to increase the confidence in the model outcomes. For instance, the Panel identified multiple sources of uncertainty in both the predicted vapor exposures and in the exposure limits in the PRZM and PEARL models. Several panelists suggested that sensitivity analyses should be conducted on the variables within the models to distinguish the relative contributions of each variable to the model outcomes. Others commented on the perhaps more urgent need to have additional field studies conducted in different cropping scenarios to increase the confidence in the exposure value predicted by these models.

One Panelist stated that the toxicity estimates have uncertainties. There are several ways to express this uncertainty:

- Include a discussion of the UFs used to calculate the CoC, including the database UF. The larger the value of the UFs, the larger the uncertainty. If the Agency uses a MOE approach, the value of the level of concern (i.e., the value of the MOEs) would express the uncertainty.
- If the dose-response data are amenable to benchmark dose modeling, the ratio between the benchmark concentration (BMC), the central estimate, and the 95% upper confidence limit of the BMC (BMCL) is a quantitative measure of uncertainty in the dose-response data. The larger the ratio, the larger the uncertainty.
- A probabilistic method using the distribution of the UFs to calculate a CoC could be used to derive a range of CoCs and quantify uncertainty.

In discussing potential pesticide studies that could be used as a model for exposure through inhalation affecting multiple systems, one panelist suggested paraquat. There have been multiple studies on the inhalation toxicity of paraquat (Dinis-Oliveira *et al.*, 2008; Haley, 1979) om addition to its neurotoxic effects (Haley, 1979; Liou *et al.*, 1997; Dinis-Oliveira, 2006) and its use in animal models to study Parkinson's disease (Betarbet, 2000; Gorrell, 1998; Thirachelvam, 2000). The principal limitation in paraquat's use as a model chemical for inhalation risk assessment is that it is a charged compound. However, paraquat has a reported VP of $1x10^{-7}$ mmHg, low but similar to the VP for a couple pesticides in Table 3 and 4 in the Agency's background document. Moreover, there is potential inhalation exposure to paraquat (a defoliant),

e.g., smoking leaves of treated plants. Paraquat residues may be re-volatilized when smoking a cigarette, *i.e.*, the VP substantially increases when heated to a cigarette's burn temperature, for example. The smoke carries the paraquat into the deep long where it can be extracted by lung fluid. Re-volatilized residues would be estimated using flux models, assuming that they are sophisticated enough to account for mixtures (paraquat, water, and anything else that may be co-applied to the crop). Thus, paraquat can provide a working toxicological pesticide model for multisystem negative health outcomes associated with an inhalation pathway.

Alternative Risk Assessment Approach

The broad charge to the Panel for Topic C combined aspects of the exposure prediction in Topic A and the toxicological assessments in Topic B. The Panel discussed an alternative risk assessment approach that is applicable to the Occupational Safety and Health Administration (OSHA) and allows both hazard and exposure assessments to be combined in a simple, yet potentially useful way. Using this approach, the vapor exposure hazard is first separated into its chemical-specific components via the VHR and its environmental components via the "Environmental Dilution Ratio" [EDR]. A determination of acceptable exposures involves the simultaneous use of both ratios as EDR/VHR. In principle, both VHR and EDR involve only physical and toxicological properties; in that sense, they are strictly science. The final question of whether one decides that an acceptable value of the EDR/VHR ratio representing a given use condition for a particular pesticide should be a value of only 1 or an MOE of 100 or another level of concern is a policy decision.

A brief summary of the alternative VHR/EDR approach applicable to OSHA is provided below, and a more detailed explanation is provided in Appendix D. For OSHA, the acceptability of an exposure is defined by a simple ratio of the chemical concentration to which employees are exposed [abbreviated herein by C] divided by that chemical's exposure limit [EL] where health effects would not be expected to occur. The EL is often an OSHA permissible exposure limit [PEL] or Threshold Limit Value [TLV®]. Such EL values do not have explicit uncertainty factors built into them.. To be acceptable in those other settings, the EL for a given chemical must simply be greater than its measured or predicted C, or the ratio in Equation C5 must be greater than one.

$$\frac{EL}{C} \text{ must be} > 1 \qquad \text{Eqn. C5}$$

Predicting an acceptable setting using Equation C6 is complicated because both EL and C are affected by so many variables. An alternative way to define this same level of acceptability is by using the ratio of two other ratios as shown in Equation C7. Both the EDR and the VHR are related to C and EL, respectively.

$$\frac{EL}{C} = \frac{EDR}{VHR} \text{ must be} > 1 \qquad \text{Eqn. C6}$$

The VHR is mathematically defined by Equation C7 as the amount of dilution a given chemical needs between the chemical's saturated vapor concentration right at the source and an acceptable vapor concentration or EL value defined by the chemical's toxicity. A list of VHR values for pesticides was extracted from a broader list of chemicals compiled by Popendorf (2006) is found in Appendix D. This list was rank ordered from the pesticide with the highest VHR to the lowest VHR. These VHR values are not directly applicable to those the Agency might use. In the case of the Agency and this charge, a pesticide's toxicity would be defined quantitatively based on the duration of the exposure, *i.e.*, acute, sub-chronic, and chronic. Thus, each pesticide is likely to have a different VHR for each exposure scenario (but potentially a similar rank-order in each such scenario's list). Such a rank ordered list of VHR values could easily identify those pesticides with the greatest (and those with a negligible) vapor hazard. In this sense, the VHR provides a more useful list than a list based on the current Tier I criteria of just a chemical's vapor pressure. VHR values could also be modified slightly by including some of the soil interaction terms from the Tier IIA Woodrow model into the numerator for those pesticides that might have a higher flux from soil than from foliage.

$$VHR = \frac{P_{vapor} \ \ in\ units\ \ of\ \ ppm\ \ or\ \ mg/m^3}{exposure\ \ limit\ \ in\ the\ \ same\ \ units} \qquad \text{Eqn. C7}$$

The EDR is mathematically defined by Equation C8 as the amount of dilution that a given environmental setting can or does create between the chemical's saturated vapor concentration right at the source and the vapor concentration at any defined location, *e.g.*, 10 meters downwind of a sprayed field. A separate list of EDR values could be generated for various application and location settings using a combination of flux and dispersion models as currently proposed by the Agency. This procedure is analogous to what several panelists referred to as "back-calculating exposure."

$$EDR = \frac{P_{vapor} \ \ in\ units\ \ as\ \ in\ the\ VHR}{P_{partial} \ \ in\ any\ \ defined\ \ location} \qquad \text{Eqn. C8}$$

The criterion implied by Equations C5 and C6 do not incorporate any Margin of Exposure (MOE).To relate the EDR and VHR approach to the MOE approach for an "acceptable exposure" to a pesticide, the ratio of both EL/C and EDR/VHR would also need to exceed the MOE for that chemical, as depicted in Equation C9. An MOE with a value of 100, for example, would provide an added level of assurance that exposures (or doses) to pesticide workers or to the public will not exceed limits set based on toxicological studies in a laboratory.

$$\frac{EL}{C} = \frac{EDR}{VHR} \ must\ \ be > MOE \qquad \text{Eqn. C9}$$

Looking at uses of volatile (or semi-volatile) pesticides in terms of EDR and VHR values provides a method to evaluate the environmental conditions separately from the various pesticides. For instance, the Panel thought that the Agency could explore the use conditions that result in low EDR values and thereby target high risk crops (*e.g.*, as a function of foliage density or height), weather (*e.g.*, as a function of atmospheric stability or temperature), or regions (*e.g.*,

as a function of soil conditions). Similarly, the Panel noted that the Agency could identify those pesticides that have a high VHR based on an array of temporal exposure scenarios with their appropriate toxicological end points.

The following discussion illustrates how both the EDR and VHR have direct applications to the case study presented to the Panel in Section 5 of EPA's background document. The EDR in the case study is the ratio of saturated air concentration (in Table 4, these values are 660 $\mu g/m^3$ for chemical C_1 and 16 $\mu g/m^3$ for chemical C_2, or 209 $\mu g/m^3$ when combined[5]) divided by the predicted air concentration (Table 9 contains both a Tier IIA prediction of 5.3 $\mu g/m^3$ and the corresponding "Max 24-hour Air Concentrations" Tier IIB predictions of 2 or 4 $\mu g/m^3$ by PRZM and PEARL, respectively). These values yield an EDR of 209/5.3 = 39 for Tier IIA or 105 and 52 for PRZM and PEARL, respectively. These predicted EDR values are all reassuringly similar to the mean and range of the minimum observed EDR values corresponding to the "Percent Departures from Study" in Table 4.[6] An extension of this approach would be to create a table of VHR values structured like Table D-1 of Appendix D in which each exposure limit applicable to general industry is replaced by a HEC or RfC concentration applicable to pesticides. Assuming for the moment that the values in Table D-1 were applicable, then those pesticides with a VHR greater than EDR/MOE = 39/100 = 0.39, if used under the same conditions (*i.e.*, with the same EDR value), would result in an MOE of less than 100, an exposure level that is generally considered to be of concern.

EDITORIAL COMMENTS ON THE BACKGROUND DOCUMENT

Several panelists made the following editorial comments to improve the clarity and adequacy of the overall presentation of the issues in the Agency's background document.

Errors were noted in the Agency's Equations 2, 3, and 4, and Tables 4 and 5

The units in Equations 2, 3, and 5 should be given as $\mu g/m^2/hr$ (as in Woodrow *et al.*, 2001), not $g/m^2/hr$ as shown in the Agency's background document. This error was apparently not internal to the $\mu g/m^2/s$ flux predictions presented in Table 5; however, this discrepancy was disconcerting to the external reader trying to reconcile high gram values predicted by Tier IIA model as shown with models that predict evaporation of liquid solvents.

In Table 4, the ratio for Chemical B of the Tier I Model concentration of 340,000 ng/m^3 to the maximum monitored concentration of 27,700 ng/m^3 is 12.5 not 125 as implied by 12,493%. Another discrepancy that may or may not be due to an error was uncovered when trying to validate the Tier IIA flux rates for chemical D in Table 5. The only way to get the value of 0.91 $\mu g/m^2/s$ is to assume a depth into the soil at which the chemical was applied but which was

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[5] The saturation vapor concentration of chemical C is based on its description of a 30:70 mixture (assumed to be a molar ratio) of isomer C_1 and isomer C_2 (p. 35 and 54) and Raoult's law such that the mixture's concentration is (660 x 0.3) + (16 x 0.7) = 209 $\mu g/m^3$. . Raoult's law is likely to apply to a neat mixture of these two chemicals because they are very similar to each other; however, both may behave non-ideally if they are diluted in water.

[6] The "Percent Departures from Study" values in Table 4 equal 100 x ([the Tier I Model Concentration] / [the Maximum Monitored Concentration] - 1) = 100 x (EDR - 1). . Similarly, the EDR = (% Departure/100) + 1. . The fact that the latter values in the table are "maximum" concentrations, mean that the resulting ratios and EDR values are the minimum values that have been observed.

not provided to the reader.

Table 8 and Table 10 Values

One Panelist made a minor editorial comment regarding certain toxicological calculations used in the case study. Some values in Table 10 do not correspond to corresponding values in Table 8. The same Panelist attempted to reproduce the HEC values in Table 8, but there was not enough information provided in the case study to do so. For clarity, the EPA should include the detailed equations and intermediate calculations used to calculate the Short Term (1-30 days), Acute HEC, and Short-term HEC from the 7-day and 21-day inhalation study.

Definition of Terms

One Panel member suggested that EPA define the meaning of different terms used in the background document, *e.g.*, concentration of concern (CoC); level of concern (LOC); acute, short-term, intermediate durations of the exposure. One concern was use of the abbreviation "IE" to describe the inhalation exposure to which an individual is exposed because of the potential confusion with the abbreviation of IEC. Perhaps, "ground-level concentration (GLC)" or "ground-level exposure concentration (GLEC)" would be a better term.

Latest Developments in Toxicity Assessments

The Agency should include references to the most up-to-date scientific methods in toxicity assessments for chemicals with adequate toxicity data and indicate they plan to use these techniques. There is no mention of the latest developments in toxicity assessments such as the use of benchmark dose modeling to calculate an appropriate point of departure, the use of categorical regression, the use of data to justify UFs that are different from the default UF of 10, the use of data to calculate chemical-specific adjustment factors, or the use of the Multiple Pass Particle Dosimetry Model (MPPD). The background document does not discuss the potential for certain segments of the population to have differential susceptibility, e.g., children compared with adults.

Physicochemical Properties

Information on physicochemical properties of the pesticides should be provided in the background document. Vapor pressure as well as solubility data would be helpful.

Haber's Rule

The Agency should provide a more thorough discussion of Haber's rule as modified by ten Berge et al. (1986) in the background document, *e.g.*, Jarabek (1995).

LOC for Pesticide C

In the case study, the LOC for Pesticide C was a factor of 30. A discussion on why a database UF was not considered would be helpful for transparency.

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ATTACHMENT D, Pt. 3

Appendix A. Comparison of Measured Air Concentrations in Field Studies in the United Kingdom with Predicted Air Concentrations using EPA's Tier I Approach

The EPA background document "Scientific Issues Associated with Field Volatilization of Conventional Pesticides" included cases studies where field data showed lower measured concentrations than the proposed Tier I procedure estimated, see Table A-1 below. For these examples, the use of the proposed Tier I saturated air concentrations overestimate the observed concentrations by several orders of magnitude.

Table A-1. Tier I Saturated air concentrations (Cs) presented in EPA's document, "Scientific Issues Associated with Field Volatilization of Conventional Pesticides" (information taken from Tables 3 & 4)

Chemical	A	B	C1	C2	D
VP (torr)	9.70E-06	1.80E-05	3.00E-06	7.20E-07	2.40E-02
MW (g/mole)	263.21	350.59	406.92	406.92	189.32
Cs ($\mu g/m^3$)	137	340	66	16	244521
Maximum measured concentration ($\mu g/m^3$)	4.38	27.7	3.8	0.2	8.9
ratio of Cs to "maximum measured C"	31	12	174	80	27,474

The Chemicals Regulation Directorate in the UK commissioned several recent studies to monitor residues in air following applications of pesticides with a range of vapor pressures, the lowest of which are less than the case studies examples referred to in the EPA background document. These pesticides are shown in Table A-2. The Tier I saturated air concentration of trifluralin is estimated at about 1900 $\mu g/m^3$. In field experiments, however, the highest concentration observed was about 90 $\mu g/m^3$, see Figure A-1. This level of overestimation is similar to that observed in the case studies provided in the EPA background document.

Table A-2. Examples of Tier I Saturated air concentrations (Cs) for pesticides included in recent field studies performed in the UK for the Chemicals Regulation Directorate

Chemical	Trifluralin	Fenpropidin	Epoxiconazole	Tebuconazole	Prothioconazole
VP (Pa)	1.40E-02	1.70E-02	1.00E-05	1.30E-06	4.00E-07
VP (Torr)	1.05E-04	1.28E-04	7.50E-08	9.75E-09	3.00E-09
MW (g/mole)	335.5	273.5	329.76	307.81	312.2
Cs ($\mu g/m^3$)	1896	1877	1.33	0.162	0.0504

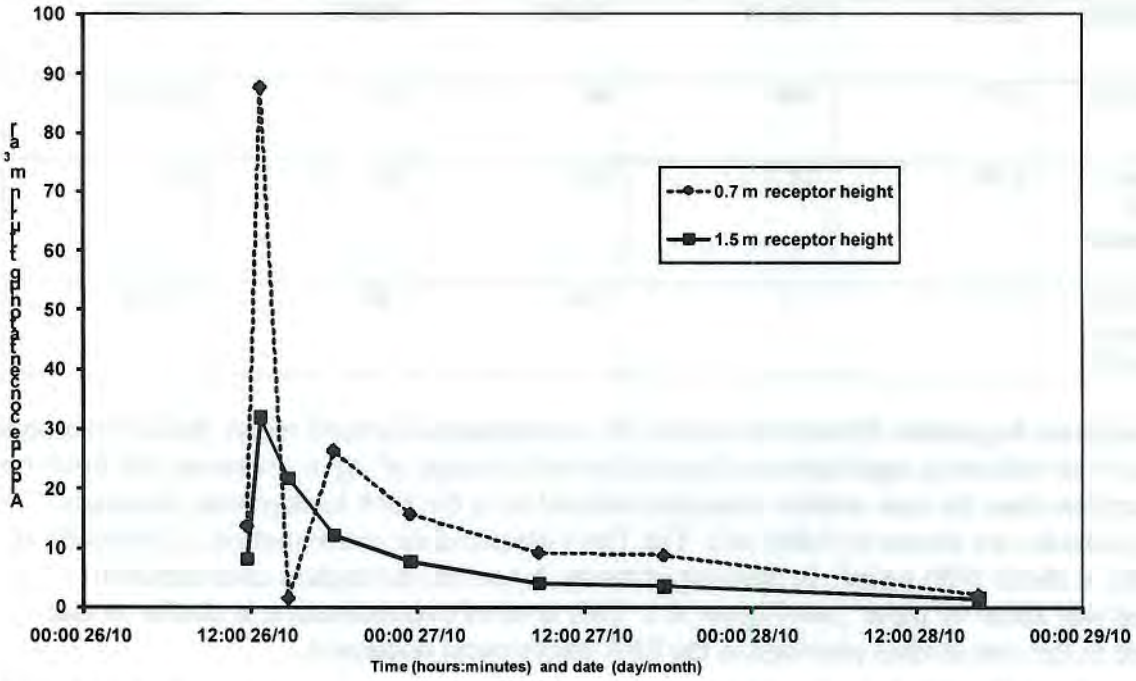

Figure A-1. Highest measured air concentrations during monitoring of trifluralin residues in air at receptor heights of 0.7 meters and 1.5 meters, both at 2 meters from the edge of 1 ha plot after application to bare soil without incorporation. [Data from 2007 Defra Project Report PS2008: Measurements of bystander contamination during and post the application of pesticides relevant to arable crops in typical UK conditions Part 2: studies with a volatile formulation, available online at: http://randd.defra.gov.uk/Document.aspx?Document=PS2008_5212_FRP.docm.

The Tier I saturated air concentration of fenpropidin is also about 1900 $\mu g/m^3$ while that of epoxiconazole is about 1 $\mu g/m^3$. These two pesticides were applied together as a tank mix of formulated products in a series of experimental trials. The results from the first trial are shown in Figure A-2 and Figure A-3. The observed concentrations of fenpropridin were usually higher than those of epoxiconazole, and at most were about two orders of magnitude below the Tier I estimate. However, on the first day, after the application was completed and on the next day concentrations of epoxiconazole were equivalent to the Tier I estimate were observed. In a second trial done in a later year, field concentrations of epoxiconazole were above the Tier I concentration (see Figure A-4).

The air sampling method for these pesticides involved the use of Occupational Safety and Health Administration (OSHA) versatile sampling (OVS) tubes. The first stage of these tubes is a filter that collects particles, and between a third and a half of the total pesticide in many air samples was found on the filter, but this mass is not included in the vapor phase concentration value. An absorbent to collect vapors comprises the second stage.

Members of the Panel questioned the possibility of a sampling artifact in the UK studies whereby particulate-phase pesticide residues were captured thereby increasing the measured air concentration value. The sampling artifact could have occurred if a portion of the particulate-phase pesticide was stripped from the filter during sampling and retained by the sorbent bed in the second stage of the sampler. If this mechanism were applicable, then much of the sample may not have been vapors in the field, and the vapor concentration may not have exceeded saturation. However, researchers in the UK had already anticipated this potential [artifact] and had undertaken a trial involving particulate sampling of epoxiconazole and fenpropidin using a personal cascade impactor collecting particles 0.3-50 µm alongside the OVS tubes. The results of this parallel sampling indicated no evidence that a significant proportion of the material collected in the OVS tubes was associated with contaminated particles. Further tests in a wind tunnel and observations of the dust collected confirmed that had significant quantities of contaminated particles been present in the air they would have been detected on the impactor plates (Defra Project PS2016).

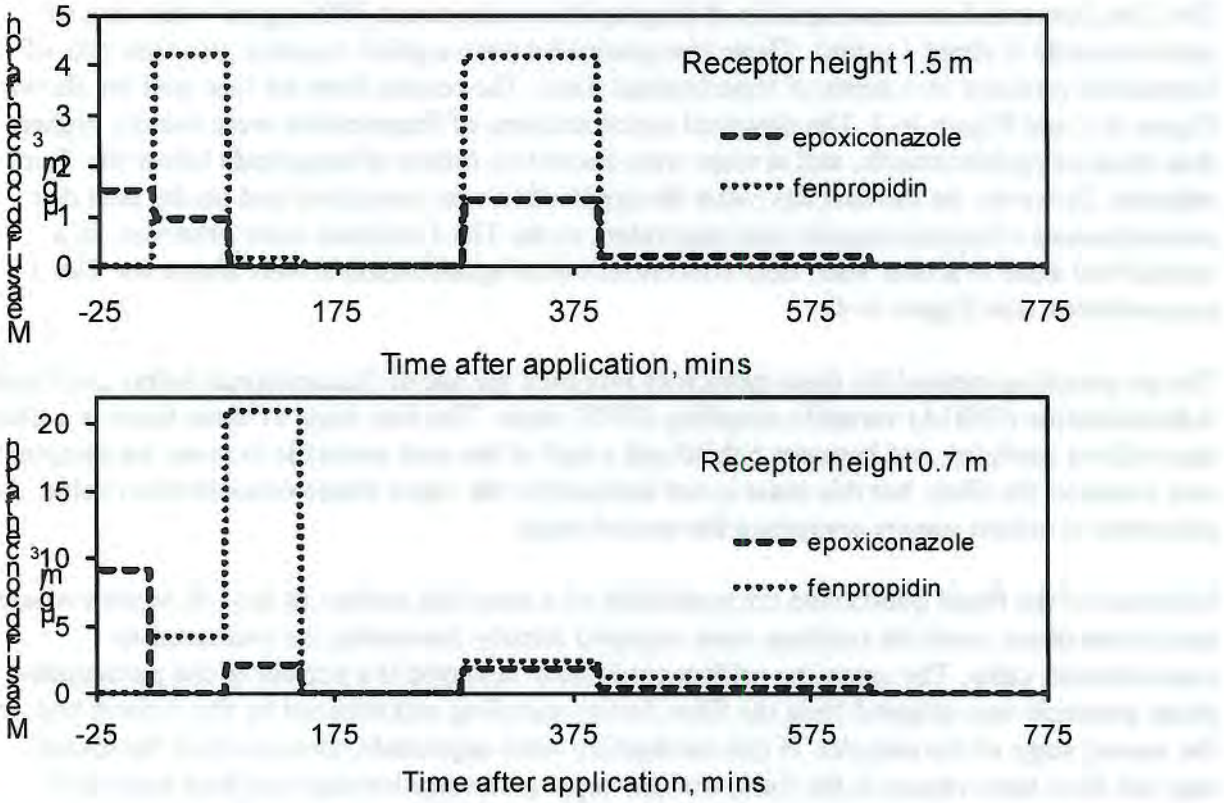

Figure A-2. Measured air concentrations of epoxiconazole and fenpropidin residues at two receptor heights, at 2 meters downwind from the edge of a 4.8 ha treated area following application to an established cereal crop in 2006. [Data from Defra Project PS2005, reported in Bulter-Ellis MC and Miller PCH (2008), Progress in the development of a bystander and resident exposure assessment model, Aspects of Applied Biology 84, 2008, International Advances in Pesticide Application.]

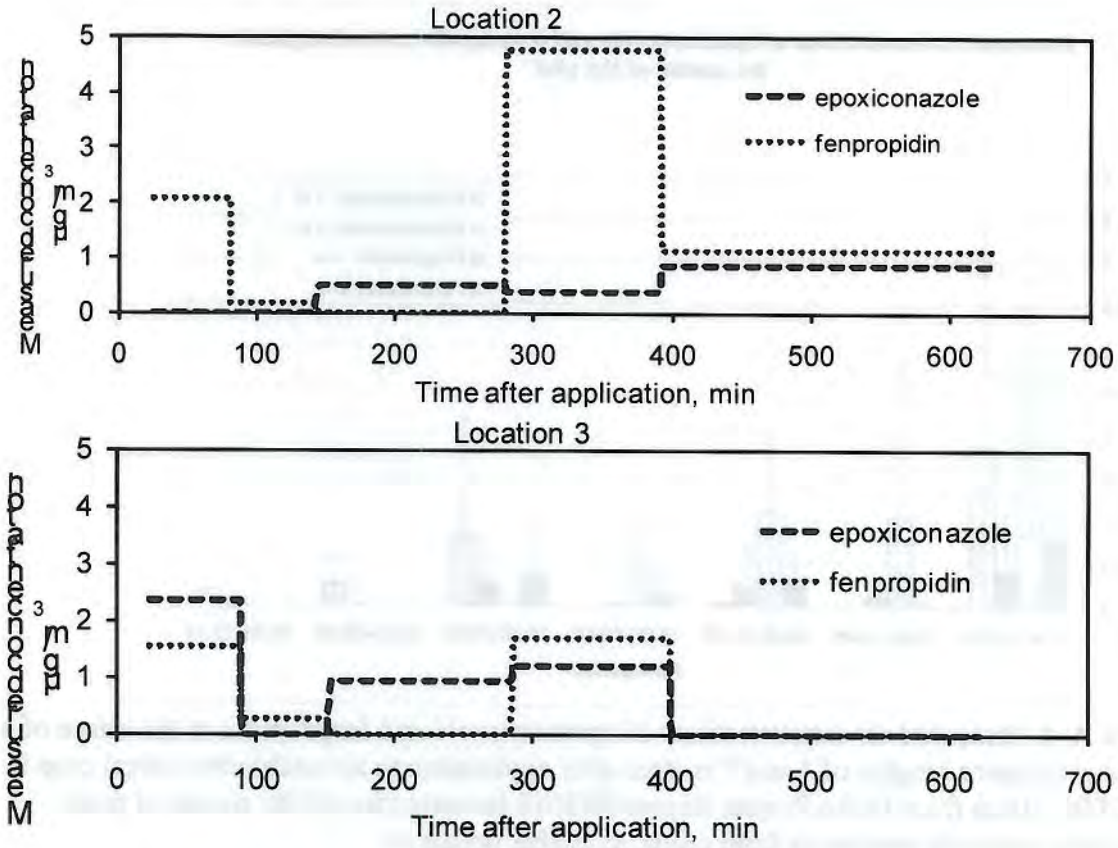

Figure A-3. Further results from monitoring of epoxiconazole and fenpropidin residues in air at 0.7 metre receptor height, at two additional locations both 2 meters downwind from the same application as referred to in Figure A-2 [Data from Defra Project PS2005, reported in Bulter-Ellis MC and Miller PCH, 2008, Progress in the development of a bystander and resident exposure assessment model, Aspects of Applied Biology 84, 2008, International Advances in Pesticide Application.]

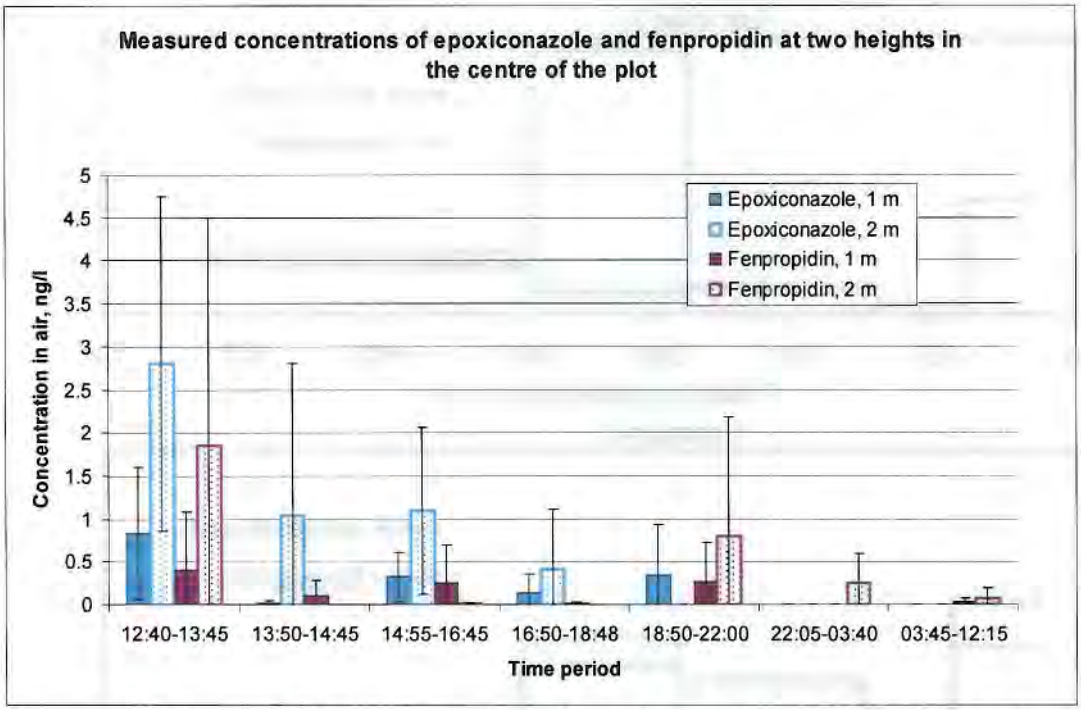

Figure A-4. Measured air concentrations of epoxiconazole and fenpropidin at the centre of a 3.7 ha plot at receptor heights of 1 and 2 metres after application to an established cereal crop in June 2008. [Data from Defra Project Report PS2016 Investigation of the source of post-application pesticide emissions from crops, available online at: http://randd.defra.gov.uk/Document.aspx?Document=PS2016_8607_FRP.doc.

The air sampling method for epoxiconazole, fenpropidin, thiabendazole and prothioconazole all involved the use of Occupational Safety and Health Administration (OSHA) versatile sampling (OVS) tubes. The first stage of these tubes is a filter that collects particles, and between a third and a half of the total pesticide in many air samples was found on the filter. An absorbent to collect vapors comprises the second stage. Whether a significant portion of the measured vapor could have been stripped from the particulate form on the filter and retained by the sorbent bed in the second stage was of some concern because if this mechanism were applicable, then much of the sample may not have been vapors in the field, and the vapor concentration may not have exceeded saturation. However, to investigate this possibility a trial with epoxiconazole, and fenpropadin, involving particulate sampling was done using a personal cascade impactor collecting particles 0.3-50 µm alongside the OVS tubes. The results of this parallel sampling indicated no evidence that a significant proportion of the material collected in the OVS tubes was associated with contaminated particles. Further tests in a wind tunnel and observations of the dust collected confirmed that had significant quantities of contaminated particles been present in the air they would have been detected on the impactor plates (Defra Project PS2016).

A third U.K. trial (Defra Project PS2023) was conducted in October 2009 at a laboratory independent of the first facility. In that trial a similar tank mix of epoxiconazole and fenpropidin were applied to grass. Preliminary results show that maximum concentrations of fenpropidin were similar to those in the previous trials, while maximum concentrations of epoxiconazole were slightly lowers those seen before.

Table A-3. Measured air concentrations of epoxiconazole and fenpropidin (mean of 3 samples) in the centre of a 12 meter untreated square in a large field following experimental application to a grass crop in 2009. [Preliminary data from Defra Project PS2023.]

	Epoxiconazole			**Fenpropidin**		
	OVS Sorbent ng m-3	OVS Filter ng m-3	Total ng m-3	OVS Sorbent ng m-3	OVS Filter ng m-3	Total ng m-3
30 min	66	410	476	6430	16047	22477
1 hour	<LOQ	<LOQ	<LOQ	7265	10414	17679
2 hours	<LOQ	<LOQ	<LOQ	4213	3538	7751
4 hours	<LOQ	<LOQ	<LOQ	2115	1963	4078
6 hours	<LOQ	<LOQ	<LOQ	87	604	691
8 hours	<LOQ	<LOQ	<LOQ	121	462	583
12 hours	<LOQ	<LOQ	<LOQ	85	509	594
24 hours	<LOQ	<LOQ	<LOQ	23	167	190
36 hours	<LOQ	1	1	10	136	146
48 hours	<LOQ	14	14	5	22	27
3 days	<LOQ	8	8	1	16	17

Finally, preliminary results are also available from monitoring a commercial application of products containing prothioconazole and tebuconazole to wheat in the summer of 2009, the Tier I estimates for these compounds are 0.05 and 0.2 $\mu g/m^3$, respectively. These data, Table A-4, show that the maximum air concentrations of tebuconazole were about $1/10^{th}$ of the Tier I estimate. However, the maximum observations for prothioconazole were above the Tier I estimate for that compound. These observations cast doubt on the reliability of the proposed Tier I approach particularly for compounds with low vapor pressures.

Table A-4. Measured air concentrations of prothioconazole and tebuconazole (mean of 3 samples) in the centre of a 12 meters untreated square in the centre of a large field following a commercial application to a cereal crop in 2009. [Preliminary data from Defra Project PS2023.]

	Prothioconazole			**Tebuconazole**		
	OVS Sorbent ng m-3	OVS Filter ng m-3	Total ng m-3	OVS Sorbent ng m-3	OVS Filter ng m-3	Total ng m-3
15 min	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
1 hour	7.78	<LOQ	7.78	<LOQ	<LOQ	<LOQ
2 hours	38.33	21.11	59.44	<LOQ	16.39	16.39
4 hours	44.17	35.14	79.31	<LOQ	16.53	16.53
6 hours	38.06	22.36	60.42	<LOQ	4.44	4.44
8 hours	59.17	57.92	117.09	<LOQ	5.42	5.42
12 hours	48.33	32.01	80.34	<LOQ	5.35	5.35
24 hours	24.68	4.17	28.85	0.93	4.54	5.47
36 hours	43.33	29.1	72.43	1.78	2.45	4.23
48 hours	23.08	1.76	24.84	3.26	15.05	18.31
3 days	3.78	1.26	5.04	<LOQ	0.79	0.79
4 days	2.73	1.33	4.06	<LOQ	0.91	0.91
5 days	1.44	0.66	2.1	<LOQ	0.37	0.37
6 days	0.88	0.39	1.27	<LOQ	0.49	0.49
7 days	0.84	<LOQ	0.84	0.22	<LOQ	0.22
8 days	0.24	<LOQ	0.24	<LOQ	<LOQ	<LOQ
9 days	0.22	<LOQ	0.22	<LOQ	<LOQ	<LOQ
10 days	0.2	<LOQ	0.2	<LOQ	<LOQ	<LOQ
11 days	0.19	<LOQ	0.19	<LOQ	<LOQ	<LOQ
12 days	<LOQ	<LOQ	<LOQ	0.08	<LOQ	0.08

Appendix B. Three Approaches for Predicting Vapor Pressure of a Chemical within a Mixture

Note: One panelist provided this discussion adapted from Chapter 6 of *Industrial Hygiene Control of Airborne Chemical Hazards* by W. Popendorf (CRC Press, 2006).

The vapor pressure of a chemical within a mixture can be predicted by three basic approaches:

- Raoult's Law for ideal mixtures,
- an empirical adjustment to Raoult's Law for non-ideal mixtures, and
- Henry's Law for dilute mixtures in water.

Each approach has its advantages. Raoult's Law is simple, but many mixtures are not ideal. Unfortunately, failure to anticipate that a mixture is not "ideal" usually results in underestimating the health hazard or/and overexposing people to mixtures evolving organic vapors. Empirical coefficients are illustrative of non-ideal effects but are not simple to predict. And Henry's Law coefficients exist for many chemicals in water, but their accuracy is limited to very dilute mixtures.

Raoult's Law for Ideal Liquid Mixtures

Raoult's law is based on the premise that molecules within a liquid mixture all act independently of each other, leading to the logic that the number or density of molecules of each component in the mixture that would be present at the liquid's surface at any given moment would be reduced in proportion to that component's molar fraction within the liquid. Raoult's Law can be written as Eqn. B-1.

$$P_{vapor,\,i} = X_i \times P_{vapor} \qquad \text{Eqn. B-1}$$

The notation $P_{vapor,i}$ is used to designate the i^{th} chemical's vapor pressure when it is present as a component of a liquid mixture. The symbol X_i, indicates each component's molar concentration in the liquid state, *viz.*, X_i is the moles of component "i" per mole of liquid mixture.

Chemical mixtures that obey Raoult's Law are called an ideal mixture, and mixtures that deviate from Raoult's Law are called non-ideal mixtures. Most chemicals behave like a component in an "ideal mixture" when it is present at high liquid concentrations (more than 50% pure) or/and when its molecules are structurally similar to the molecules of the other components within a mixture. Unfortunately, the molecular interactions within mixtures of organic solvents and water are sufficiently strong that such mixtures often deviate from Raoult's Law.

The Need for an Empirical Adjustment to Raoult's Law

The more different that the molecules in a liquid mixture are from each other, the more likely they are to interact and their vapor pressures are to deviate from Raoult's Law. The opportunity for different molecules to interact is small as long as there are still many identical molecules around (*i.e.*, when the mixture is relatively concentrated), but deviations will get larger when most of the other molecules are "different" (*i.e.*, when a component in a mixture is dilute).

An "activity coefficient" is used in chemical engineering to adjust Raoult's simple, logical law for ideal mixtures to match the behavior of non-ideal liquid mixtures. This activity coefficient is given the symbol "γ" (a lowercase Greek gamma). This γ is sometimes also referred to as a "fugacity coefficient." Equation B-2 both defines γ, mathematically, and expresses the empirical concept behind it.

$$\gamma_i = \frac{measured\ or\ actual\ P_{vapor,i} = Y_i \times P}{Raoult's\ P_{vapor,i} = X_i \times P_{vapor}} \qquad \text{Eqn. B-2}$$

In a mixture, each component "i" will have its own γ_i. Each component's activity coefficient could be used in Eqn. B-3 to predict the absolute vapor pressure in units such as mmHg. Equation 3 will revert to Raoult's law (Eqn. 1) when γ =1.

$$P_{vapor,i} = \gamma_i \times X_i \times P_{vapor} = \gamma_i \times \text{Raoult's predicted } P_{vapor,i} \qquad \text{Eqn. B-3}$$

Figure B-1 can provide some graphical insight into how values of six organic solutes in water vary both among the six chemicals and also within each mixture as a function of "X." This figure shows how values increase as each solution gets more dilute and as the molecules become more dissimiliar from water. The γ^{∞} for very symmetric organic molecules like benzene occurs off-scale. The smallest γ^{∞} in this figure is for methanol because it is the chemical most similar to water. A γ value greater than one means that the component's vapor pressure is greater than predicted by Raoult's Law. Most organic chemicals when diluted in water will present a surprisingly high vapor hazard if their activity coefficient is not anticipated.

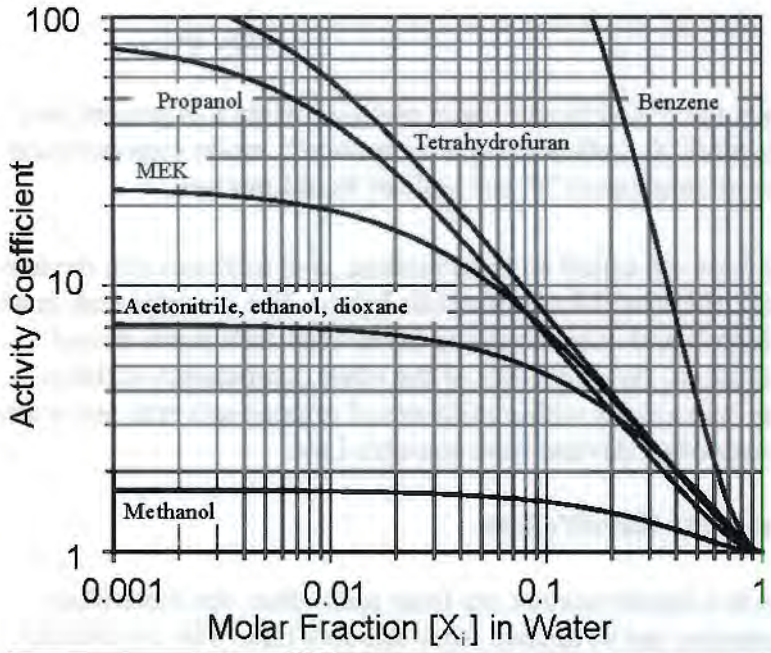

Figure B-1. Activity coefficient [γ_i] values for selected organic solutes in water. Adapted from Popendorf (2006).

Methods to Predict an Empirical Adjustment to Raoult's Law

The following four approaches to predict the activity coefficient γ_i were adapted primarily from Chapter 11 of the *Handbook of Chemical Property Estimation Methods* (Lyman et al., 1982), a jewel of a resource if one is challenged to estimate an otherwise hard-to-get or non-existent property value. Other sources of property estimation methods are also available (Reid *et al.*, 1987; Poling *et al.*, 2001). The middle two methods below rely on experimental data for the particular mixture, while the first and last methods can estimate γ without prior mixture-specific measurements.

A method first developed by Pierotti *et al.* (1959) and refined by Grain (1982) can be used to predict γ^∞ for binary mixtures. The method shown below in Eqn. 4 was described by Grain as "easy" in that its seven coefficients are based on the *overall* molecular structure of the solute and solvent (rather than on each component's individual moieties as in the Universal Functional Activity Coefficient (UNIFAC). Lyman's book contains four pages of tables and internal correction factors for the seven variables included in Eqn. 4.

$$\log \gamma_1^\infty = A_{1,2} + B_2 \times \left[\frac{N_1}{N_2}\right] + \left[\frac{C_1}{N_1}\right] + D \times (N_1 - N_2)^2 + \left[\frac{F_2}{N_2}\right] \qquad \text{Eqn. B-4}$$

The van Laar equation (*ca.* 1910) provides a generic relationship to estimate γ_i at any concentration based only on γ_1^∞ and γ_2^∞ derived either from measurements or other predictive methods (Hirata *et al.*, 1975; Poling *et al.*, 2001; Grain, 1982).

$$\ln \gamma_1 = \ln \gamma_1^\infty \times \left[1 + \frac{X_1 \ln \gamma_1^\infty}{X_2 \ln \gamma_2^\infty}\right]^{-2} \qquad \text{Eqn. B-5}$$

The most robust method to predict activity coefficients is via the group contribution method of UNIFAC (Poling *et al.*, 2001; Bishop *et al.*, 1982). While some software is available to implement the method, the lack of a database that defines the structures of even common chemicals or/and a user-friendly interface has kept these programs from being widely used.

Data in the Table B-1 excerpted from Grain (1982) allows one to compare activity coefficients at infinite dilution [γ^∞] for five common organic liquids in water The first column of γ^∞ values were experimentally measured; the second column of γ^∞ values were calculated by UNIFAC. The range of the percent errors by which the calculated γ^∞ differs from the measured γ^∞ that are presented in the third column of data suggests that UNIFAC does not always predict activity coefficients accurately. However, even the highest of these errors (131% for toluene in water) is much better than the 339,000% error that would result if no γ^∞ value were used at all (equivalent to assuming Raoult's Law or $\gamma^\infty = 1$ versus the γ^∞ of 3390 measured experimentally).

Table B-1. Comparison of activity coefficients at infinite dilution [γ^{∞}] for five common organic liquids in water (excerpted from Grain, 1982)

	Experimental γ_i^{∞} in water	Calculated γ_i^{∞} in water	% error	Experimental γ_{water}^{∞} in organic	$\frac{\gamma^{\infty}\ organic\ solvent}{\gamma^{\infty}\ water}$
Hexane	489,000	402,000	-18%	1880	260
Toluene	3390	7820	131%	3320	1.0
Benzene	1730	1670	-3.5%	226	7.7
Aniline	34.2	50.6	48%	4.98	6.9
Acetone	6.80	5.69	-16%	5.64	1.2
	average absolute error = 43%				

Henry's Law

Henry's Law assumes that the vapor pressure of component "i" [$P_{vapor,i}$] is proportionate to X_i via a fixed empirical coefficient called either a "Henry's Law constant" or "Henry's Law coefficient" denoted by the symbol "H_i" herein. A common expression of Henry's Law might look like Eqn. B-6.

$$P_{vapor,i} = H_i \times X_i \qquad \text{Eqn. B-6}$$

The major advantage of Henry's Law is the widespread availability of its constants (Lyman *et al.*, 1982; Yaws, 1992; MacKay *et al.*, 1992; Howard and Meylan, 1997; Sander, 2004). Unfortunately, the diverse origin of these coefficients has created an additional inconvenience because they have been developed with a considerable amount of variability in the units of H_i. Fortunately, on-line converters are available, *e.g.*, at the following sites:

www.mpch-mainz.mpg.de/~sander/res/henry-conv.html

www.epa.gov/athens/learn2model/part-two/onsite/henryslaw.htm

The limitation that Henry's Law only applies to dilute mixtures follows from the fact that H_i is a constant, independent of X_i. Based on the prior discussion of how γ_i decreases as the mixture gets less dilute, the relationship between H_i and γ_i can be found by equating a very dilute component's vapor pressure predicted by the empirical adjustment to Raoult's Law (Eqn. B-3) to that predicted by Henry's Law (Eqn. B-6).[7]

$$P_{vapor,i} = H_i\ X_i = \gamma_i\ X_i\ P_{vapor} \qquad \text{(repeating Eqns. B-3 and B-6)}$$

[7] While the relationship in Eqn. B-6 is conceptually true, technically γ^{∞} can only be unitless as shown, if both H_i and P_{vapor} have the same units. . Unfortunately, their units will likely not be the same, nor will their units be those commonly used within industrial hygiene.

$$H_i = \gamma^{\infty}_i P_{vapor} \qquad \text{Eqn. B-7a}$$

$$\gamma^{\infty}_i = \frac{H_i}{P_{vapor}} \qquad \text{Eqn. B-7b}$$

Henry's Law coefficient [H_i] will be constant as long as the component's concentration is sufficiently dilute that γ_i stays equal to the constant γ^{∞}. This range is best seen in Figure B-1. The curves of most activity coefficients flatten out to a constant γ^{∞} by the time the liquid molar concentration decreases to about 10^{-3}. The γ^{∞} of a few chemicals with small γ^{∞} values is already constant by the time the X_i reaches 10^{-2}, while those with large γ^{∞} values aren't constant until X_i is less than 10^{-4}. But in all cases, continued use of a Henry's constant for more concentrated mixtures will eventually over-estimate the real component's vapor pressure and its resulting airborne exposures.

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Appendix C. Alternative Approaches to Assess Inhalation Hazard in the Absence of Inhalation Toxicity Studies

One Panelist provided a discussion of alternative analytical approaches to assess inhalation hazard in the absence of inhalation toxicity studies. These approaches are screening approaches and do not involve a chemical-specific hazard identification and dose-response analyses. These screening approaches are Threshold of Regulation, Threshold on No Toxicological Concern, and the NOAEL-to-LC_{50} approach. These approaches were originally developed to evaluate oral exposure to trace levels of chemicals and to determine if toxicity testing should be required for those chemicals. There are only a few studies that evaluate inhalation hazard in the absence of inhalation toxicity studies and the exposure durations in these studies [chronic, lifetime exposure (Drew and Frangos, 2007) and one-hour intermittent exposure (Grant *et al.*, 1997)] do not correspond to the exposure durations the Agency is evaluating. Therefore, the majority of the Panel does not endorse the Drew and Frangos (2007) or Grant *et al.* (2007) approaches be used for pesticides in the interim in lieu of new inhalation toxicity studies. The following material is provided for informational and completeness purposes.

A Tiered Approach to Evaluate Acute Inhalation Toxicity (Texas Commission on Environmental Quality)

During the air permit review process, the Toxicology Division (TD) of the Texas Commission on Environmental Quality (TCEQ) frequently evaluates chemicals with limited toxicity data (LTD). When the minimum acute database requirement (as discussed in Section 3.4 of the Effects Screening Level (ESL) Guidelines (TCEQ, 2006)) is not met, an acute inhalation reference value (ReV) protective of a 1-hr intermittent exposure, is not developed. Instead, a tiered approach is used to either set a default screening value (i.e. Threshold of Regulation) or derive a generic health-based screening value depending on the availability of toxicity information and time and resource constraints (Figure C-1).

- Tier I – Threshold of Regulation (default ESL = 1 µg/m3)
- Tier II – Threshold of Concern and Use of LC_{50} Data (generic ESL)
- Tier III – Relative Toxicity/Potency Approach (generic ESL)

For the TCEQ, a generic short-term ESL is typically based on a one-hour averaging time. The following sections discuss the procedures used to set health-protective concentrations for LTD chemicals based on a tiered approach and the strengths and weaknesses of these approaches. The Tier III - Relative Toxicity/Potency Approach will not be discussed.

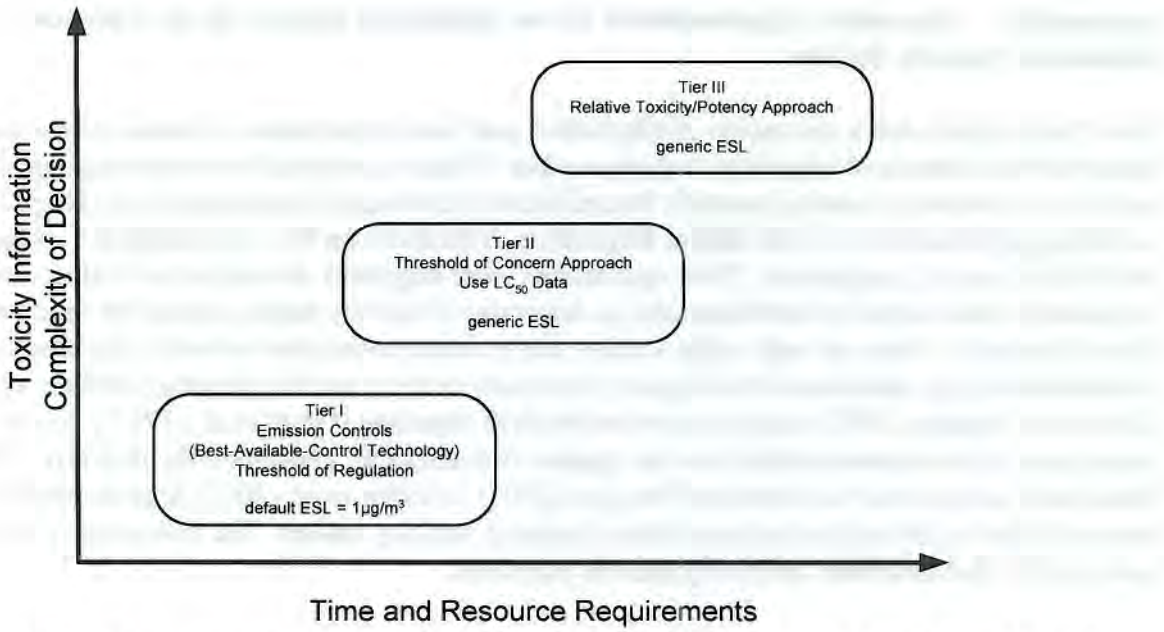

Figure C-1. A three-tiered approach to setting a default or a generic health-based ESL.

A Threshold of Regulation Approach

A Threshold of Regulation (TR) approach seeks to answer the question "if the concentration of a chemical in air is small, how small does an exposure have to be before it can defensibly be regarded as presenting trivial risk". Other terms used to describe this approach are "threshold of concern" or "threshold of no toxicological concern." Drew and Frangos (2007) provide a concise review of the statistical, analytical procedures used to establish the Threshold of Regulation approach adopted by US FDA and other organizations for oral exposure and its use in regulatory toxicology. Briefly, the fifth percentile of the cumulative percentage distribution of NOAELs in a large oral exposure database was determined and divided by an uncertainty factor of 100 (to account for animal-to-human uncertainty and human variability) to derive an estimate of an acceptable oral intake. Other investigators have used this approach for other products such as food flavorings, personal and household care products and pharmaceutical compounds (see Drew and Frangos (2007) and Grant *et al.* 2007 for references). Drew and Frangos (2007) developed a concentration of no toxicology concern (CoNTC) for evaluation of trace organic chemicals in air for evaluation of chronic exposure. This conservative generic CoNTC was derived by performing a route-to-route extrapolation from the virtually safe oral dose of 1.5 µg/person/day divided by a factor of two. Drew and Frangos (2007) proposed a generic CoNTC of 0.03 µg/m^3. It does not apply to metals, particulates or chemicals that produce sensory irritation.

Historically, the TCEQ has used a default of 1 µg/m^3 as a TR value for evaluation of a 1-hr intermittent exposure compared to the value of 0.03 µg/m^3 developed by Drew and Frangos (2007) for evaluation of chronic exposure. A value of 1 µg/m^3 is conservative. Based on an acute inhalation database for 97 chemicals, the fifth percentile of NOAELs would be 560 µg/m^3 (see Table 3, Grant *et al.*, 2007), and with a UF of 100 applied, it would be 5.6 µg/m^3 as a TR value for evaluation of a 1-hr intermittent exposure. This TR concentration is an estimate of threshold

air concentrations below which no appreciable risk to the general population would be expected to occur after a one-hour intermittent exposure.

Strengths:

1. A TR approach is conservative since inhalation toxicity of the most toxic chemicals is used to predict toxicity for all chemicals.
2. When a TR approach is used, inhalation toxicity information for a pesticide is not required – only the monitored/modeled vapor concentration
3. The TR value is based on statistical analyses of inhalation toxicity data, and the variability in the TR value can be determined
4. There is precedent in using a TR approach in a regulatory setting for oral data (*e.g.*, FDA 1995).
5. The monitored/modeled vapor concentration of semi-volatile pesticides in air are present in trace concentrations, and may be below the TR value.
6. Unnecessary animal studies are not performed because it identifies those chemicals that need additional testing.

Weaknesses:

1. The TR approach is only as good as the inhalation database used to derive the values. To develop a TR approach to evaluate different durations of exposure, one would need an adequate inhalation database for each exposure period. At the present time, an adequate database of inhalation toxicity data for vapors of pesticides for the durations of exposure the Agency uses in its assessment are not available.
2. The TR approach is conservative, and only chemicals that are present in air in trace concentrations would be screened out.
3. Since it is more difficult to conduct inhalation toxicity studies, the numbers of acceptable inhalation toxicity studies for vapor-phase semi-volatile pesticides would be small compared to oral toxicity studies.
4. The TR approach is a screening approach and does not involve a chemical-specific hazard identification and dose response analyses. Therefore, it does not produce a reference value that can be used in a margin-of-exposure approach or cannot be used to calculate a hazard quotient. Therefore, this screening approach does not lend itself to a cumulative assessment of different pathways of exposure or pesticides.

Classification System to Categorize Chemicals into Different Toxicity Potency Classes

If a classification system can be derived to categorize chemicals into different inhalation toxicity potency classes, then a Tier II threshold of no toxicological concern can be derived for each toxicity potency class. Drew and Frangos (2007) provide a concise review of determining a threshold of toxicological concern for different potency classes using the Cramer classification system (Cramer, 1978) for oral toxicity studies.

For oral exposure, the Cramer classification system successfully assigns chemicals into separate toxicity potency classes. For inhalation toxicity, both Grant *et al.* (2007) and Ford *et al.* (2006)

determined that the Cramer classification system (*i.e.*, a structurally –based system based on oral exposure) does not adequately place chemicals in separate toxicity potency classes predictive of inhalation exposures, so a structural alert system is not available for inhalation toxicity. The main difficulty is the inability to accurately predict whether chemicals would cause respiratory tract point-of-entry effects: "Currently, there are no internationally recognized screening methods in animals to predict the ability of chemicals to cause local effects of the respiratory tract" (Rennen *et al.*, 2004).

Grant *et al.* (2007) used LC_{50} data to classify chemicals into different inhalation toxicity potency classes. Ninety-seven chemicals were classified based on the Globally Harmonized System of Classification and Labeling of Chemicals proposed by the United Nations into different acute inhalation toxicity categories (from most toxic to least toxic): Category 1, Category 2, Category 3, Category 4, and Category 5. The tenth percentile of the cumulative percentage distribution of NOAELs in each category was determined and divided by an uncertainty factor of 100 to derive the following health-protective threshold of concern (TOC) concentrations for inhalation exposure: 4 $\mu g/m^3$ for chemicals classified in Category 1, 20 $\mu g/m^3$ for Category 2, 125 $\mu g/m^3$ for both Categories 3 and 4, and 1000 $\mu g/m^3$ for Category 5. These TOC concentrations are estimates of threshold air concentrations below which no appreciable risk to the general population would be expected to occur after a one-hour intermittent exposure.

Strengths:

1. By having a classification system to divide chemicals into different inhalation toxicity classes, one single threshold of regulation based on the most toxic chemical in the entire dataset is not used for a relatively nontoxic chemical.
2. This approach is conservative since the most toxic chemical within each class is used to represent the inhalation toxicity of all chemicals within one class.
3. The TOC values for each potency class are based on a statistical analysis of inhalation toxicity data, and the variability in the TOC value can be determined.
4. There is precedent in using a TOC approach in a regulatory setting, mainly for oral data
5. Unnecessary animal studies are not performed because it identifies those chemicals that need additional testing.

Weaknesses:

1. This approach is only as good as the inhalation database used to derive the values. At the present time, an adequate database of inhalation toxicity data for vapors of pesticides for the durations of exposure the Agency uses in its assessment are not available.
2. A predictive structural classification system is not available for inhalation data, so LC_{50} data are required.
3. Since it is more difficult to conduct inhalation toxicity studies, the numbers of acceptable inhalation toxicity studies is small compared to oral toxicity studies, so the numbers of chemicals in each separate toxicity potency class is small. This increases the uncertainty in the inhalation TOC concentrations.

4. This approach of classifying chemicals into different potency categories requires more inhalation toxicity studies than a single TR approach discussed above or the LC_{50} to NOAEL Ratios approach discussed in the following section.
5. The TOC approach is a screening approach and does not involve a chemical-specific hazard identification and dose response analyses. Therefore, it does not produce a reference value that can be used in a margin-of-exposure approach or cannot be used to calculate a hazard quotient. Therefore, this screening approach does not lend itself to a cumulative assessment of different pathways of exposure or pesticides.

LC_{50} to NOAEL Ratios

Layton *et al.* (1987) used oral LD_{50} data for estimating acceptable daily intakes for the evaluation of exposures to contaminants at hazardous waste sites. Venman and Flaga (1985) also proposed the use of LD_{50} data to establish provisional acceptable daily intakes for the evaluation of waste water contaminants. Both investigators calculated the ratio of NOAELs from chronic animal studies to oral LD_{50} data for different chemicals and determined the fifth percentile of the cumulative distributions of the ratios. The LD_{50} value for contaminants with limited toxicity data was multiplied by the fifth percentile ratio to derive a surrogate NOAEL. The surrogate NOAEL was divided by an UF of 100 in order to establish a conservative threshold dose below which no appreciable risk to human health would be expected to occur. Grant *et al.* (2007) used the basic approach of Layton *et al.* (1987) and Venman and Flaga (1985) for evaluating chronic oral toxicity and applied it to acute inhalation toxicity. Therefore, ratios of NOAELs from acute inhalation studies to LC_{50} data were calculated.

For the NOAEL-to-LC_{50} ratio approach, 55 chemicals with an inhalation NOAEL for an exposure duration $\leq$ 24-hours were used to calculate NOAEL-to-LC_{50} ratios. The tenth percentile of the cumulative percentage distribution of the ratios was calculated and divided by an uncertainty factor of 100 to produce a composite factor equal to 8.3×10^{-5}. For a chemical with limited toxicity information, this composite factor is multiplied by a 4-hour LC_{50} value or other appropriate acute inhalation lethality data as defined in Grant *et al.* (2007) to produce an estimate of a conservative threshold air concentration below which no appreciable risk to the general population would be expected to occur after a one-hour intermittent exposure.

Strengths:

1. LC_{50} data are available for most chemicals.
2. LC_{50} data are predictive of acute inhalation toxicity, since LC_{50} data were able to categorize chemicals into statistically significant distributions and toxicity potency classes.
3. This approach is conservative since the 5^{th} or 10^{th} percentile NOAEL-to-LC_{50} ratio is applied to the chemical-specific LC_{50} data.
4. The NOAEL-to LC_{50} ratio approach is based on a statistical analysis of inhalation toxicity data, and the variability in the ratio can be determined.

Weaknesses:

1. This approach is only as good as the inhalation database used to derive the values. At the present time, an adequate database of inhalation toxicity data for vapors of pesticides for the durations of exposure the Agency uses in its assessment are not available.
2. There should be an acceptable number of LC_{50} data and matching inhalation NOAEL data for each duration of exposure being evaluated.
3. Since it is more difficult to conduct inhalation toxicity studies, the numbers of acceptable pesticide inhalation toxicity studies is small, so the number of NOAEL-to-LC_{50} ratios may be inadequate.
4. This approach is a screening approach and does not involve a chemical-specific hazard identification and dose response analyses. Therefore, it does not produce a reference value that can be used in a margin-of-exposure approach or cannot be used to calculate a hazard quotient. Therefore, this screening approach does not lend itself to a cumulative assessment of different pathways of exposure or pesticides.

Time Extrapolation Factors or Assessment Factors

Refer to Kalberlah *et al.* (2002), Kramer *et al.* (1995, 1996), and Malkiewicz *et al.* (2009) for a discussion of time extrapolation or assessment factors. These references are provided for informational and completeness purposes only.

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Appendix D. Derivation and Elaborations on the Concepts of "Vapor Hazard Ratio" [VHR] and the "Environmental Dilution Ratio" [EDR]

Note: The following discussion is based on Chapters 5 and 8 of *Industrial Hygiene Control of Airborne Chemical Hazards* by W. Popendorf (CRC Press, 2006) and W. Popendorf: Vapor Pressure and Solvent Vapor Hazards. Amer. Ind. Hyg. Assoc. J., 45(10): 719-726 (1984).

With the eventual goal in mind of providing a theoretical framework for separating factors intrinsic to the chemical from factors intrinsic to the environment, this discussion begins with an explanation of evaporation. Although vapor pressure is not the only factor that affects evaporation, it is the only factor that is intrinsic to the chemical. All of the other variables that affect the evaporation rate depend either upon the physical nature of the liquid source (like its size and shape) or upon the speed and turbulence of the passing air.

As the vapors are swept away from the liquid surface by the passing air, more molecules evaporate from the liquid to maintain that same very localized equilibrium. Thus conceptually, the rate of evaporation is determined by how fast vapor molecules can diffuse across a thin, low speed "*boundary layer*" of air that always exists very close to a liquid surface. The thickness of that boundary layer (typically only a few millimeters along the vertical axis in Figure D-1) is determined by the air velocity, its turbulence, and the geometry of the liquid source (or its container if applicable).

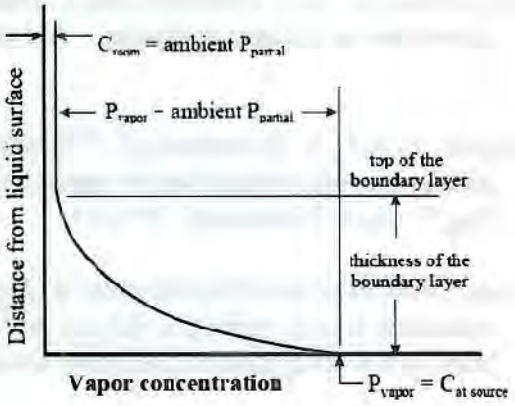

Figure D-1. Depiction of evaporation as diffusion due to a concentration difference across a boundary layer.

The vapor concentration (the horizontal axis in Figure D-1) at the liquid surface at the bottom of this boundary layer is always equal to the chemical's vapor pressure [P_{vapor}]. The concentration at the top of the boundary layer is the ambient concentration [ambient $P_{partial}$ or C_{room} if indoors].

Mechanisms of Vapor Generation

The liquid and its vapors are **always** considered to be in equilibrium with each other right at the liquid-air interface. The concentration of a vapor in equilibrium is (by definition) its vapor pressure [P_{vapor}]. P_{vapor} can be equated to C in mg/m³ and to ppm. The latter equation is written here as Equation D-1 because of the practical importance of ppm (*cf.*, C which also depends upon the chemical's molecular weight).

$$ppm\ at\ the\ source = \frac{P_{vapor} \times 10^6}{P_{atmosphere}} = \frac{P_{vapor}\ [mmHg] \times 10^6}{760\ [mmHg]} \qquad \text{Eqn. D-1}$$

These conditions set the parameters for what is modeled as molecular diffusion across that boundary layer. Thus, the evaporation rate is largely determined by a set of environmental conditions and one chemical-dependent condition: the chemical's vapor pressure. The chemical engineer's equation to predict evaporation rate is somewhat more complex than needed for field applications (Bird *et al.,* 1960; Sherwood *et al.*, 1975; Fiegley *et al.*, 1981; Bishop *et al.*, 1982) However, industrial hygiene research has used an equation similar to Equation D-2 in many studies (Fiegley *et al.*, 1981; Bishop *et al.*; Powell, 1984; Säämänen *et al.*, 1991; Braun and Caplan, 1992; Nielsen *et al.*, 1995; Nielsen and Olsen. 1995; Hummel *et al.,* 1996).

$$evaporation\ rate\ [G_{moles}] = (Geom.Coef.)\ (A)\ (V^{0.5})\ (P_{vapor} - ambient\ P_{partial}) \qquad \text{Eqn. D-2}$$

where …

G_{moles} = the evaporation rate in terms of moles of vapor generated per unit of time. Both $G_{mass} = MW \times G_{moles}$ and flux = G_{mass}/A could be created from this equation.

Geometric Coefficient (Geom. Coef.) = an empirical "evaporative mass transfer coefficient" that characterizes the effect of the source geometry (its size and shape) on the thickness of the "boundary layer."

Area (A) = the size or surface area of the volatile liquid (ft² or m²).

Velocity (V) = the velocity of the air passing over the evaporating source (*e.g.*, fpm or m/sec).

P_{vapor} = the vapor pressure of the evaporating chemical at its liquid temperature. The units of P_{vapor} are normally mmHg or Pascals which can be converted to mg/m^3 or ppm.

$P_{partial}$ = the partial pressure (or concentration) of the same vapor in the ambient air passing over the source. The chemical's ambient partial pressure is usually far enough below its vapor pressure, that ambient $P_{partial}$ can be disregarded and omitted, as shown in Equation D-3 and beyond.

$$evaporation\ rate\ [G_{mass}] = (MW)\ (Geom.Coef.)\ (A)\ (V^{0.5})\ (P_{vapor}) \qquad \text{Eqn. D-3}$$

One example of such an evaporation equation was developed for EPA by Caplan for organic solvents evaporating from a smooth surface like a spill of 0.5 to 3 feet in diameter (Caplan, 1989). These authors showed through experimental data that an equation equivalent to Equation

D-4 can predict the evaporation rate of a fairly wide range of organic chemicals from such a smooth surface to within a factor of about 2-fold.

$$G\ [mg/min] = (0.0706)\ (MW)\ (A)\ (V^{0.625})\ (P_{vapor}) \qquad \text{Eqn. D-4}$$

where …

G	=	the vapor generation rate, mg/min.
MW	=	the Molecular Weight, g/mole (added to convert moles to grams).
0.0706	=	the empirical "geom.coef."
A	=	the surface area of the liquid source, ft².
V	=	the air velocity over the liquid surface, ft/min.
P_{vapor}	=	the vapor pressure of the evaporating substance, mmHg.

In contrast to comments made within the Agency's background document, the value of a chemical's molecular diffusion coefficient has a minor impact on volatilization (due to the relatively narrow range of values of that coefficient among the chemicals of interest, such as a range of 1.11x among those pesticides in the white paper's Table 3) and even less of an impact on plume dispersion (in contrast to eddy diffusion in dispersion). Eddy diffusion is the dominant mechanism below and to the right of the line in Figure D-2 representing the 1.5×10^{-5} m^2/sec molecular viscosity of air (Smagorinsky, 1974; Smagorinsky, 1981; Atkinson, 1995). Molecular diffusion is too slow to be an important chemical transport mechanism *except* either over very small distances or over very long times.

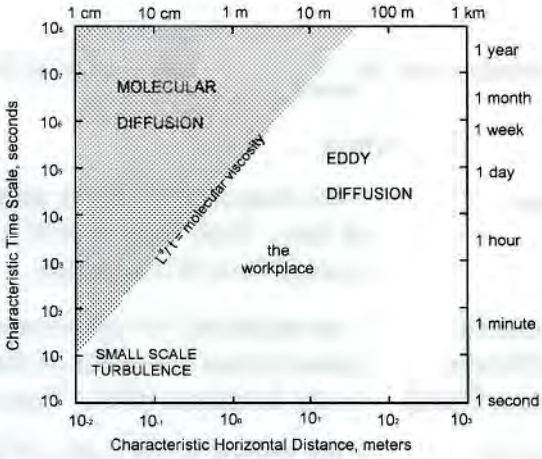

Figure D- 2. Adapted from Smagorinsky (1981).

Dilution of Vapors from Continuous Evaporation

The steady state concentration [C in mg/m³] equals the mass rate of evaporation (or G_{moles} HMW) divided into some apparent volumetric flow rate of fresh air (Q in m³/min) into which the contaminant appears that it is being diluted, as depicted by Equation D-5.

$$steady\ state\ C\ [mg/m^3] = \frac{G_{mass}\ [mg/m^3]}{Q_{apparent}\ [m^3/\min]} = \frac{G\ [moles/mim] \times MW}{Q_{apparent}\ [m^3/\min]} \qquad \text{Eqn. D-5}$$

It is important to understand that the subscript "apparent" is used because the plume of contaminant represented by "G" is not diluted into any easily definable volume flowing per minute (except in the case of thorough mixing inside a room served by general ventilation). It only appears to be diluted because the concentration [C] at any location is always less than C equivalent to P_{vapor} at the source. The amount of dilution will depend, for example, upon one's orientation and distance from the source.

At least in theory, Equations D-3 and D-5 could be combined into Equation D-6 (or Equation D-7) to predict the average concentration from continuous evaporation to which someone would be exposed. The concept is theoretical not only because "Geom.Coef." is difficult to predict for most sources (*e.g.*, a task relegated to PRZM or PEARL for pesticides) but also because $Q_{apparent}$ would depend upon the air flow pattern and the person's proximity to the source and the plume in a way that is currently difficult to predict (a task relegated to PERFUM). Nonetheless, this equation can be manipulated to good effect as explained below.

$$vapor\ C\ [mg/m^3] = \frac{(Geom.Coef.) \times (Area) \times V^{0.5}}{Q_{apparent}} \times MW \times P_{vapor} \qquad \text{Eqn. D-6}$$

$$vapor\ ppm = \frac{(Geom.Coef.) \times (Area) \times V^{0.5} \times 24.45}{Q_{apparent}} \times P_{vapor} \qquad \text{Eqn. D-7}$$

The arrangement of the variables within Equation D-7 suggests that the ppm vapor concentration at any location in the vicinity of an evaporating source can be viewed as having two groups of determinants: a group of environmental determinants (those that comprise the "ratio" on the right side of Equation D-7) and a chemical determinant (the chemical's vapor pressure on the far right side of Equation D-7).

- None of the **environmental determinants** of exposure within either Equation D-6 or D-7 have anything to do with the chemical *per se*. The numerator in the parentheses comprises only the physical characteristics of the source that affect its evaporation or flux rate. The denominator encompasses only the physical mechanisms that dilute the plume in the pathway from the source to the specific location at which the vapor concentration is being described. Together, this whole group of environmental variables determines by how much the vapors will get diluted between the P_{vapor} concentration existing at the source and the concentration [C] at the location of interest in an exposure scenario. Environmental factors only affect dilution, and (other than how temperature affects vapor pressure) they do not affect the initial vapor concentration at the source.

- The singular **chemical determinant** of exposure on the far right side of Equation D-7 has nothing to do with the environment in which the chemical is being used (again other than the source temperature affecting vapor pressure [P_{vapor}]. Popendorf (2006) showed that a temperature increase of approximately 12°C or 21°F is required to double the P_{vapor} of a wide range of organic solvents. This is less of an effect than many other factors affecting

vapor exposures to a pesticide such as its vapor pressure at normal temperature (spanning 4 to 6 orders of magnitude). Thus, the chemical component only affects the source of exposure, not the pathway or its dilution.

Most aerosols (except for fumes) have virtually no chemical-specific vapor pressure, but an equivalent G [mg/min] for aerosols can be defined for any particular aerosol source. Once an aerosol is generated, its dilution generally still depends only on $Q_{apparent}$ (as long as sedimentation is not important) (Bémer *et al.*, 2000). Thus, an aerosol concentration could be predicted using Equation D-6 in much the same way as for vapors if the aerosol generation rate G and environmental dilution due to $Q_{apparent}$ were known. However, $Q_{apparent}$ can rarely be predicted quantitatively in practice.

This partitioning allows one to examine the environmental determinants of exposure separate from the chemical determinants. Equation D-8 isolates the environmental determinants by dividing Equation D-7 by P_{vapor} and inverting. The resulting ratio is the amount by which the vapor pressure concentration at the source [P_{vapor}] is diluted to create the resulting concentration at any location. Because the ratio in Equation D-8 is the amount of dilution created by the environment, this ratio is called the "Environmental Dilution Ratio" or EDR. Again notice that all of the variables that comprise the EDR relate to the environment, not to the chemical being used in that environment. This ratio is unitless as long as the units of concentration in both the numerator and denominator are the same.

$$EDR = \frac{P_{vapor}\ converted\ to\ C\ or\ ppm}{C\ or\ ppm\ at\ a\ defined\ location} = \frac{Q_{apparent}}{(Geom.Coef.) \times (Area) \times V^{0.5}} \qquad \text{Eqn. D-8}$$

Separating the environmental from the chemical determinants of exposure allows chemical hazards to be viewed in a new way. In the traditional view, a chemical's concentration is either measured or predicted, then its acceptability is based on the ratio of the chemical's exposure limit [denoted herein as EL in units of either mg/m³ or ppm] to that chemical's measured or predicted concentration [denoted herein as either C or ppm and must be in the same units as the EL]. In the occupational health field, *e.g.*, within OSHA, this ratio must be greater than one to be acceptable, as expressed by Equation D-9a. To relate the occupational health field approach to EPA's MOE approach, the ratio of EL/C would need to exceed the pesticide's MOE, as defined in Equation D-9b.

Whether an acceptable ratio is 1 (as in Eqn. D-9a) or an MOE (as in Eqn. D-9b) is a policy decision outside the purview of this summary.

$$\frac{Exposure\ Limit\ [EL]}{measured\ or\ predicted\ C\ or\ ppm}\ must\ be > 1 \qquad \text{Eqn. D-9a}$$

$$\frac{Exposure\ \ Limit\ [EL]}{measured\ \ or\ \ predicted\ \ C\ \ or\ \ ppm}\ must\ \ be > MOE \qquad \text{Eqn. D-9b}$$

Knowing that P_{vapor} (or its equivalent in either ppm or mg/m³) is the maximum concentration at which a given chemical can exist as a vapor, one can anticipate that the maximum hazard that a chemical's vapor can **potentially** create is equivalent to the ratio of how much greater P_{vapor} is than its exposure limit. The "Vapor Hazard Ratio" or "VHR" as expressed mathematically in Equation D-10 is the maximum potential hazard that a given chemical's vapors can generate.

$$VHR = \frac{P_{vapor}\ \ in\ units\ \ of\ \ ppm\ \ or\ \ mg/m^3}{Exposure\ \ Limit\ [EL]\ in\ the\ same\ units} \qquad \text{Eqn. D-10}$$

The two properties that comprise the VHR (its vapor pressure and its exposure limit) are both intrinsic to a given chemical (and have nothing to do with the environment *per se*). Together, they specify the minimum amount by which a given chemical's vapor needs to be diluted by the environment to reduce its concentration from its vapor pressure at the source to its exposure limit in the breathing zone. If the vapors in some one's breathing zone are not diluted by as much as the chemical's VHR, then that person will be overexposed. In terms of EPA's MOE approach, the vapors must be diluted by the VHR times a desired MOE, *e.g.*, VHR x 100.

Nothing is wrong with evaluating acceptability as simply the ratio of measured results to an exposure limit, but the separate concepts of chemical dilution and environmental dilution can be re-combined advantageously. Think of looking at evaluation as a process of comparing the amount of dilution that a given chemical needs to the amount of dilution that a given environment actually creates. In terms of the MOE approach, this alternative view of evaluation is the same as asking if the Environmental Dilution Ratio in someone's breathing zone is sufficiently greater than the Vapor Hazard Ratio for the chemical being used, as expressed in Equation D-11.

$$\frac{C_{measured\ \ or\ \ predicted}\ /\ P_{vapor}}{EL\ /\ P_{vapor}} = \frac{EDR}{VHR}\ must\ \ be > MOE \qquad \text{Eqn. D-11}$$

Conceptually viewing acceptability in its separate chemical and environmental components can yield several benefits. For instance, one could anticipate that a given chemical should only be used in an environment that can dilute the vapors reaching a given population (the "EDR" for that setting) by as much as the toxicity of a given chemical requires its vapors to be diluted (the chemical's "VHR" times the applicable MOE). If an evaluation reveals that a defined population is being overexposed, then one can conclude that the environment cannot create sufficient dilution for the chemical being used. One could then compare the advantages of attempting to achieve an acceptable solution either by reducing the chemical's intrinsic VHR (*e.g.*, by substituting an intrinsically safer chemical) or by increasing the environment's ability to dilute a

chemical reaching someone's breathing zone (*e.g.*, by modifying or restricting the use conditions).

This concept could also be used in setting priorities for both pesticides with intrinsic vapor hazards and use-settings susceptible to creating vapor hazards. Table D-1 within this appendix lists the VHR for about 40 pesticides based on their industrial exposure limits (TLVs are intended for manufacturing settings, see Braun and Caplan, 1992). The VHRs in this table span over 11 orders of magnitude and 9 orders of magnitude excluding fumigants. For those chemicals with a VHR less than 1, its saturated vapors are less than its exposure limit, meaning they can only over-expose someone as an aerosol. A similar list could be generated to rank-order the pesticides of interest to the Agency, but using HEC or RfC concentrations in the denominator (instead of C). Such a list, the Panel noted, could interface with an array of Environmental Dilution Ratios (EDR values based on flux rate and dispersion scenarios modeled by PRZM, PERFUM, *etc.*) to potentially refine the Agency's priorities in terms of use-settings.

Table D-1. Pesticides that had an ACGIH TLV® in 2005 are listed in rank-order of their Vapor Hazard Ratio (VHR = P_{vapor} x 10^6 / TLV x 760). The compound's vapor pressure [P_{vapor}] is at 25°C unless otherwise indicated. Where the mass concentration is given in parentheses, the TLV® in ppm = C x 24.45/MW. Adapted from Popendorf (2006).

Chemical Name	TLV® [mg/m³]	CAS#	TLV [ppm]	P_{vapor} [mmHg]	VHR
Carbon disulfide		75-15-0	10	358.	47,105
1,3-Dichloropropene		542-75-6	1	34.	44,737
Epichlorohydrin		106-89-8	0.5	16.4	43,158
Dichlorvos (DDVP)	0.1	62-73-7	0.011	0.053	6,303
Diazinon	0.01	333-41-5	0.0008	0.0112	1,834
Ethylene dibromide (EDB)		106-93-4	20	11.	724
Naled	0.1 at 20°C	300-76-5	0.0064	0.002	410
Mevinphos (Phosdrin)	0.01	7786-34-7	0.0011	0.00013	157
Heptachlor	0.05	76-44-8	0.0033	0.0004	161
Fonofos (Difonate)	0.1	944-22-9	0.0099	0.0002	27
Sulfotep (TEDP)	0.1	3689-24-5	0.015	0.00017	15
Lindane	0.5	58-89-9	0.042	0.00041	13
Aldrin	0.25	309-00-2	0.0168	0.00012	9
Dichrotophos	0.05 at 20°C)	141-66-2	0.0052	8.6e-05	4.4
Chlorpyrifos	0.1	2921-88-2	0.007	1.7e-05	3
Parathion	0.05	56-38-2	0.0042	9.7e-06	3
Fenthion	0.2 at 20°C	55-38-9	0.018	3.0e-05	2.3
Endosulfan	0.1	115-29-7	0.0060	1.0e-05	2
Methyl parathion	0.2	298-00-0	0.0186	1.7e-05	1.2
Carbofuran	0.1 at 20°C	1563-66-2	0.011	5.0e-06	1
Endrin	0.1	72-20-8	0.0064	3.0e-06	6.2e-01
Dieldrin	0.25	60-57-1	0.0160	5.9e-06	4.8e-01
Chlordane	0.5	57-74-9	0.0298	9.8e-06	4.3e-01
Thiram	1	137-26-8	0.102	1.7e-05	2.2e-01
EPN	0.1	2104-64-5	0.0076	9.5e-07	1.7e-01
Fenamiphos	0.1	22224-92-6	0.008	1.0e-06	0.16
Malathion	1	121-75-5	0.074	7.9e-06	1.4e-01
Ronnel	10	299-84-3	0.76	7.5e-05	1.3e-01
Ethion	0.05	563-12-2	0.0032	1.1e-06	0.06
2,4,5-Trichlorophenoxyacetic acid	10	93-76-5	0.96	3.8e-05	5.2e-02
Warfarin	0.1	81-81-2	0.0079	1.2e-07	1.9e-02
Paraquat	0.1	4685-14-7	0.0095	1.0e-07	0.01
Methoxychlor	10	72-43-5	0.7074	2.6e-06	4.8e-03
Carbaryl (Sevin)	5	63-25-2	0.608	1.4e-06	2.9e-03
Diquat (respirable)	0.1	2764-72-9	0.0071	1.0e-08	1.9e-03
Atrazine	5 at 20°C	1912-24-9	0.566	3.0e-07	1.0e-03
Azinphos methyl	0.2	86-50-0	0.0154	7.5e-09	6.4e-04
Temephos	1	3383-96-8	0.52	7.9e-08	2.0e-04
Rotenone	5	83-79-4	0.31	2.6e-09	1.1e-05
Benomyl	10	17804-35-2	0.842	3.7e-09	5.8e-06
Picloram	10	1918-02-1	1.01	7.2e-11	9.4e-08

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ATTACHMENT E, Pt. 1

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES, AND
TOXIC SUBSTANCES

April 22, 2010

MEMORANDUM

SUBJECT: Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting on the Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment.

TO: Steven Bradbury, Ph.D.
Acting Director
Office of Pesticide Programs

FROM: Myrta R. Christian
Designated Federal Official
FIFRA Scientific Advisory Panel
Office of Science Coordination and Policy

THRU: Laura Bailey
Executive Secretary
FIFRA Scientific Advisory Panel
Office of Science Coordination and Policy

Frank Sanders
Director
Office of Science Coordination and Policy

Please find attached to this memorandum the meeting minutes of the FIFRA Scientific Advisory Panel open meeting held in Arlington, Virginia on February 2 – 4, 2010. This report addresses a set of scientific issues being considered by the Environmental Protection Agency pertaining to the "Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment."

Attachment

cc:

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SAP Minutes No. 2010-03

A Set of Scientific Issues Being Considered by the Environmental Protection Agency Regarding:

Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment

February 2 – 4, 2010
FIFRA Scientific Advisory Panel Meeting,
Held at the
Environmental Protection Agency Conference Center
Arlington, Virginia

NOTICE

These meeting minutes have been written as part of the activities of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP). The meeting minutes represent the views and recommendations of the FIFRA SAP, not the United States Environmental Protection Agency (Agency). The content of the meeting minutes does not represent information approved or disseminated by the Agency. The meeting minutes have not been reviewed for approval by the Agency and, hence, the contents of these meeting minutes do not necessarily represent the views and policies of the Agency, nor of other agencies in the Executive Branch of the Federal government, nor does mention of trade names or commercial products constitute a recommendation for use.

The FIFRA SAP is a Federal advisory committee operating in accordance with the Federal Advisory Committee Act and established under the provisions of FIFRA as amended by the Food Quality Protection Act (FQPA) of 1996. The FIFRA SAP provides advice, information, and recommendations to the Agency Administrator on pesticides and pesticide-related issues regarding the impact of regulatory actions on health and the environment. The Panel serves as the primary scientific peer review mechanism of the EPA, Office of Pesticide Programs (OPP), and is structured to provide balanced expert assessment of pesticide and pesticide-related matters facing the Agency. FQPA Science Review Board members serve the FIFRA SAP on an *ad hoc* basis to assist in reviews conducted by the FIFRA SAP. Further information about FIFRA SAP reports and activities can be obtained from its website at http://www.epa.gov/scipoly/sap/ or the OPP Docket at (703) 305-5805. Interested persons are invited to contact Myrta R. Christian, SAP Designated Federal Official, via e-mail at christian.myrta@epa.gov.

In preparing the meeting minutes, the Panel carefully considered all information provided and presented by EPA, as well as information presented by public commenters. This document addresses the information provided and presented by these groups within the structure of the charge.

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A Set of Scientific Issues Being Considered by the Environmental Protection Agency Regarding:

Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment

February 2 – 4, 2010
FIFRA Scientific Advisory Panel Meeting,
held at the
Environmental Protection Agency Conference Center
Arlington, Virginia

Steven G. Heeringa, Ph.D.
FIFRA SAP Chair
FIFRA Scientific Advisory Panel
Date: April 22, 2010

Myrta R. Christian, M.S
Designated Federal Official
FIFRA Scientific Advisory Panel
Date: April 22,2010

Federal Insecticide, Fungicide, and Rodenticide Act
Scientific Advisory Panel Meeting
February 2 – 4, 2010

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INTRODUCTION

The Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP) has completed its review of Scientific Issues Associated with the "Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment." Advance notice of the meeting was published in the *Federal Register* on November 18, 2009. The review was conducted in an open Panel meeting held in Arlington, Virginia, from February 2 – 4, 2010. Dr. Steven G. Heeringa chaired the meeting. Myrta R. Christian served as the Designated Federal Official.

Data from epidemiology studies and human incident reports contain valuable information, and contributes to a weight of evidence analysis in the characterization of human exposure, response to pesticides, and human health risks. Epidemiology and incident data do, however, pose challenges with respect to characterizing human health risks. EPA convened this meeting of the FIFRA Scientific Advisory Panel (SAP) to discuss science issues related to using epidemiology and human incident data in human health risk assessment. The Office of Pesticide Programs (OPP) solicited comment on a draft framework for implementing the use of such data into human health risk assessment in conjunction with several case studies. OPP's draft framework prescribes a weight of the evidence approach using the best available science for mode of action, exposure, pharmacokinetics, animal and human data determined by in vivo or in vitro studies, and physiologically-based pharmacokinetic models. Three case studies evaluated by the SAP were intended to illustrate the draft framework and to highlight key science challenges with incorporating epidemiology or human incident data into a risk assessment. The Agency solicited comment on the weight of the evidence approach for evaluating and integrating the exposure, laboratory animal data, and human incident information.

One case study presented an evaluation of several ecological and retrospective cohort epidemiology studies for atrazine. OPP, in collaboration with EPA's Offices of Water and Research and Development (ORD), solicited comment on the strengths and weaknesses of these types of epidemiology studies, and sought advice on the appropriate use of such studies in the atrazine human health risk assessment. This case study is also the first step in EPA's atrazine science re-evaluation plan as described previously at the November 3, 2009 FIFRA SAP (http://www.epa.gov/scipoly/sap/meetings/2009/110309meeting.html).

A second case study illustrated the analysis of reported human incident cases. This case study was based on diazinon, a pesticide used historically in residential settings.

A third case study represented the collaborative work by scientists from OPP, ORD, the National Cancer Institute (NCI), and the National Institute of Environmental Health Sciences (NIEHS) on the Agricultural Health Study (AHS). This case study compared the exposure algorithms used by OPP and the AHS, and considered the temporal relationships for multi-chemical exposure in the AHS. The purpose of the comparison of the OPP and AHS exposure algorithms is to better understand the differences and similarities in how the two approaches estimate worker exposure. Temporal relationships for multi-chemical exposure in the AHS involved the timing and combined uses of pesticides, with particular emphasis on pesticides sharing common modes of

action. The Agency solicited advice on the types of evaluations conducted to date and those being proposed with the third case study.

The FIFRA SAP will advise the Agency on approaches for integrating diverse types of experimental toxicology and epidemiology data. The SAP input will be considered for characterizing atrazine's human health risks to be presented to the SAP in September 2010.

Drs. Steven Bradbury, Acting Director, Office of Pesticide Programs, EPA, and Tina Levine, Director, Health Effects Division, OPP, provided opening remarks at the meeting.

The agenda for this SAP meeting included presentations from the Health Effects Division in the OPP, the National Cancer Institute and public comments.

PUBLIC COMMENTERS

Written statements were provided by:

Mark Schultz of Land Stewardship Project, Kathryn Gilje of Pesticide Action Network, Lorette Picciano of Rural Coalition/Coalición Rural and members of other organizations
Kenneth Racke, on behalf of Dow AgroSciences
Dan Campbell, on behalf of Syngenta Crop Protection, Inc
Erik Janus, on behalf of CropLife America
Jere White on behalf of Kansas Corn Growers Association and other organizations
Michele Marcus, Ph.D., Emory University
Lisa Kelley, on behalf of National Council of Farmer Cooperatives
Wayne Clifford, Washington State Department of Health

Oral statements were presented by:

Jennifer Sass, Ph.D., on behalf of Natural Resources Defense Council
Gerard Swaen, Ph.D., on behalf of Dow AgroSciences
Erik R. Janus, M.S., on behalf of CropLife America
Dominik Alexander, Ph.D., on behalf of Exponent Inc.
James Swenberg, Ph.D., Noel Weiss, Ph.D., Sir Colin Berry, Tim Pastoor, Ph.D., Charles Breckenridge, Ph.D., on behalf of Syngenta Crop Protection
Mr. Tyler Wegmeyer, on behalf of the American Farm Bureau Federation®
Mr. Scott Slaughter, on behalf of the Center for Regulatory Effectiveness
Jessica Johnson Bennett, on behalf of the National Corn Growers Association
Gary J. Burin, Ph.D., on behalf of The Triazine Network
R.L. Silken Jr., Sielken Associates, on behalf of Syngenta

SUMMARY OF PANEL DISCUSSION AND RECOMMENDATIONS

This report of the FIFRA Scientific Advisory Panel (SAP) addresses several key issues relating to the integration of data from epidemiology studies and human incident reports into the human health risk assessment process. The report is based on the draft framework prepared by the Office of Pesticide Programs (OPP) and responses to specific questions framed by the Agency. The draft framework included three case studies selected to identify key issues and scientific challenges in incorporating the results of epidemiology studies and human incident data for pesticide exposure characterization into the risk assessment process.

The Panel commends EPA for its effort to incorporate human data in human health risk assessments in a transparent manner, based on the draft framework. Overall, the Panel was impressed with the documentation presented in the draft framework and the oral presentations by OPP staff that complemented the written information. The Panel commended OPP staff for its work in developing the highly detailed draft framework. Because of the extensive information presented in the draft framework and the clarity of the charge questions, the Panel was able to conduct indepth discussions and provide EPA with specific recommendations for difficult scientific challenges.

Much work remains to be done before the changes in toxicity testing proposed by National Research Council of the National Academy of Science can be implemented. During the transition period from the current practice to that of measuring perturbations of molecular pathways due to exposure to environmental agents, it will be necessary to evaluate all available information. Epidemiologic studies have the potential to inform both the experimental toxicologist, and the regulatory manager of possible sources of harm in human populations. However, like all information considered in risk assessments, the quality and reliability of the information provided by epidemiologic studies needs to be closely scrutinized. This SAP report is intended to provide specific guidance to OPP with respect to incorporation of epidemiologic data into risk assessment.

The Panel recommended that OPP conduct a broader analysis and revision of the framework regarding the extent and nature of reliance on epidemiologic data in risk assessment to include a range of environmental chemicals including arsenic, disinfection byproducts, benzene and solvents such as trichloroethylene and perchloroethylene in addition to those described in the draft framework and associated case studies. Review of the nature and extent of reliance upon human data in risk assessments more broadly within the Agency and as a basis for development of relevant guidance in other regulatory programs is encouraged. Acquisition of epidemiologic expertise for the review and interpretation of human data, including assessment of exposure in relevant studies is also strongly recommended.

Early recognition of the likely contribution of human data in scoping/problem formulation will increase transparency, facilitate peer engagement and conserve resources. For example, lack of adequate characterization of exposure-response relationships in epidemiological studies may preclude the need to do an extensive weight of evidence analysis for these data, since they cannot contribute significantly to dose-response analysis and hence, risk characterization.

The value of framework analysis in coordinating assessment and research has not been emphasized fully in the documentation. For example, there is repeated reference to problem formulation in the draft framework without indication of how the toxicological and epidemiological databases might be considered in an integrated fashion to identify uncertainties and critical data gaps. This would be an appropriate way to identify limitations of available human data to focus additional research. Targeted additional data might include, for example, *in vitro* studies in human tissues or cell lines and focused epidemiological studies to address specific questions in potentially susceptible subgroups by evaluation of early biomarkers of effect.

Based on experience in studies of toxicity of industrial chemicals and environmental contaminants, weighting of different types of human data varies and depends on the nature of the studies and on the results (i.e., the weight of evidence of an effect in humans). Where sufficient weight of evidence for causality of an adverse outcome in humans exists based on robust epidemiologic study designs and data with well-characterized exposures, these data would be preferred to those obtained from laboratory animals for dose-response characterization. Results of epidemiologic studies in which exposure was well-characterized for a relevant effect in humans could be used as a basis to "bound" dose-response estimates from animal studies.

However, incorporation of data from epidemiologic studies into risk assessment clearly poses challenges to assure that the quality of the data is adequate for risk assessment. Therefore, particular attention must be paid to the quality of epidemiologic studies. Relevant considerations include, but are not limited to: 1) the quality of the exposure assessment, including validation measures if available; 2) sample size and statistical power to assure that a meaningful effect could be detected with reasonable probability if one exists; 3) careful definition of the outcome and assessment of the accuracy of classifying persons as diseased or not diseased; 4) attention to possible sources of bias including selection bias, information bias and confounding; 5) adequate consideration of and control for confounding and identification of effect modifying factors; and 6) external validity or the potential for the study to be generalized to other populations. As discussed further in the body of the report, the Panel recommends that OPP consider the full range of epidemiologic study designs that are used for hypothesis testing and not limit consideration to prospective cohort studies. Historical cohort studies, case-control studies, cross-sectional studies and hybrid designs all have potential to be used in the weight of evidence approach in a quantitative manner when they meet the requisite criteria, particularly for exposure assessment.

The Agency proposes a weight of the evidence approach (WOE) for evaluating human and experimental animal data. The approach described in the draft framework incorporates the modified Bradford Hill criteria as a tool for integrating a variety of types of data, including human epidemiologic data, in risk assessment. The Panel recommends that the modified Bradford Hill criteria are highly appropriate for framing the likelihood of a specified consequence of exposure to a particular chemical in humans. These criteria are well-accepted for assessing evidence of causation in the field of epidemiology and in public health. It is important, however, that the criteria not be viewed as a checklist, but rather as characteristics that collectively provide a systematic way to evaluate the evidence, aggregate observations and guide assessments and conclusions. They require flexible interpretation. Particular attention should be

focused on the criteria of strength of association, dose-response, temporal relation, consistency of findings and "biological plausibility", but with the realization that the biology often has not been established when the epidemiologic evidence becomes available. The draft framework should include a clear discussion of the meaning of biologic plausibility because several interpretations are possible. Emphasis should also be placed on evidence of dose-response relationships in epidemiologic studies, as well as on the temporal sequence of events, the strength of the associations and the consistency of findings across studies and populations.

The Panel commends the Agency for their efforts in developing the draft framework for integration of animal and human data. Future application of the framework for evaluation of weight of evidence has substantial potential for improving risk assessment and for the transparent and appropriate integration of human and toxicological data. The use of the "source to adverse outcome pathway" can also be important in identifying critical data gaps and as a basis to focus additional research. The approach moves the focus in toxicology and risk assessment from late adverse effects to earlier biomarkers of exposure and effect, so that more informative human data at relevant dose levels can be collected. Further, the framework is helpful in directing attention to dose-response relationships for early key events in the assessment of available data; it is these dose-response relationships that are critical in the subsequent risk characterization. However, for epidemiologic data to be most useful in risk characterization, exposure must be robustly and quantitatively addressed, preferably with inclusion of appropriate biomarkers of exposure and effect based on identification of key events in a mode of action context.

The draft framework provides a description of the strengths and limitations of several epidemiologic study designs. The Panel recommends that the framework clearly separate ecologic studies from other designs, since their inherent limitations and inability to estimate risk at the level of the individual render them inherently weak for quantitative purposes. Further, the Panel recommends that the term "retrospective studies" as used in the Agency report be replaced with a more complete and accurate description of the study designs that are used in epidemiology to assess risk, including prospective and historical cohort studies, case-control studies, cross-sectional studies and hybrid designs.

In response to the Agency charge, the Panel identified several criteria for "robust, well-designed epidemiology studies. It is inappropriate to attempt to use these criteria for ecologic studies due to their inherent limitations as described in detail in the body of the report. For the hypothesis-testing designs, the paramount requirement in environmental epidemiology is a well-characterized, quantitative exposure assessment that minimizes exposure measurement error and decreases the likelihood of introducing misclassification in categorical or continuous data analyses. The exposure assessment should be evaluated for accuracy, precision and reliability and should include validation where feasible. Incorporation of biomonitoring and biomarkers of exposure is particularly helpful in this regard. Exposure metrics can represent dose estimates (for example, average daily dose or peak dose), duration of exposure or a combination of these in a cumulative exposure metric.

Other characteristics of robust, well-designed studies identified by the Panel are those that are applicable to all types of epidemiologic studies and include the general criteria of study quality described in a preceding paragraph. The study should have a well-defined population that

includes exposed persons with a wide range of exposures as well as unexposed persons to be maximally informative. Investigators should recognize and attempt to control for selection bias, information bias and confounding and to identify effect modification, particularly as relevant to susceptible subsets of the population, including children. The use of explicit, well-defined criteria for ascertainment of outcome is also important, with recognition of sources of error in data bases such as birth certificate files.

Prospective and historical cohort studies, case-control and cross-sectional studies have significant potential for use in a WOE approach for risk assessment. The extent to which data from these epidemiologic designs can be applied in a quantitative context will depend on the methods used for exposure assessment and the ability of the investigators to make relatively accurate and precise measurements of dose and outcome. The potential weaknesses of each study design are well-described in the literature; thus, the Agency needs to remain cognizant of these when considering the use of data from any single study or an aggregation of studies for a specific pesticide. Well-designed case-control and historical cohort studies may have quantitative value in the risk assessment process, depending on their ability to establish dose-response relationships. Well-designed and carefully executed prospective cohort studies, such as the Agricultural Health Study (AHS), provide maximal opportunity for incorporating epidemiologic data into risk characterization. Prospective cohort studies collect exposure data prior to the onset of disease thus minimizing several forms of potential bias. Further, the depth and extent of exposure assessment, including the potential for incorporating biomonitoring measurements and biomarkers of exposure and effect in selected samples of the cohort, present unique advantages for dose estimation and assessment of multiple biologically and toxicologically relevant endpoints.

The Panel recommends that the Agency consider study quality as the primary factor in selection of studies for incorporation in the atrazine WOE analysis. An important issue is how the Agency decides whether to use particular sets of data. In the interests of transparency, the Panel recommends that the Agency establish a set of criteria for determining the acceptability of epidemiologic studies. These criteria may be based on quantitative criteria, scientific judgment, or a combination of these. Inevitably, it will be necessary to exercise some degree of scientific judgment in this assessment. The Panel recommends that epidemiologists participate actively in the process. Observational research is subject to potential error due to the nature of the science. However, the presence of uncertainty in epidemiologic research does not necessarily imply that the study cannot be used. Epidemiologists routinely make judgments about the extent of potential biases, such as an inability to measure exposure precisely or to arrive at valid estimates of dose, as well as the probable effects of these uncertainties on the risk estimates. In particular, epidemiologists consider the possibility that exposure misclassification may have biased a dichotomous categorization of exposure toward the null and distorted an exposure-response relationship or that differential misclassification, e.g., recall bias, biased the risk estimate away from the null.

The Panel also recommends that novel observations derived from epidemiologic studies should not be dismissed on the basis of a lack of concordant findings in animals. The potential for variation in the MOA across species suggests that such findings be subjected to further exploration. At the same time, the possibility that some statistically significant associations may be due to chance should be borne in mind by Agency reviewers. Similarly, the Panel recognizes

the importance of null findings in epidemiologic research, despite the potential for publication bias. The Panel recommends that a comprehensive literature search be performed, followed by the interpretation of findings by well-trained epidemiologists and others with specific expertise for the relevant exposures and outcomes. With respect to the reproductive outcomes evaluated in the draft framework and case study A, the Panel recommends that future assessments include the full suite of adverse reproductive outcomes including potential effects on the male.

Case Study A

A second goal of the SAP's work was to assess the evidence for an association between exposure to atrazine and adverse reproductive outcomes in five papers published since the 2003 IRED decision on atrazine. This case study is also the first step in EPA's atrazine science re-evaluation plan as described in the November 3, 2009 FIFRA SAP report.

In general, OPP performed an accurate and thorough analysis of the five published studies included in Case Study A and has captured most of the limitations of these studies. The descriptions of potential bias, methods of exposure assessment and statistical analysis were accurate. The Panel made several additional observations relevant to specific studies that are included in the body of the report.

The first weakness of Case Study A was limiting the criteria for inclusion of studies published since 2003 as the Panel was unable to examine the full weight of the epidemiologic evidence for atrazine and reproductive outcomes. All reports in the case study used either an ecological or a retrospective cohort design. Thus the Panel did not have the opportunity to explore how other epidemiologic designs that used cross-sectional, case-control, or other approaches might be incorporated in the risk assessment process. Second, the overall quality of these studies was relatively low, thus limiting their applicability to the upcoming review of atrazine or the more general issue of incorporating epidemiology in risk assessment. Third, two of the five published studies used an ecologic design (Mattix et al., 2007; Winchester et al., 2009). At best, these studies might contribute to hazard identification and problem formulation, but better studies of atrazine and reproductive outcomes are available to meet this goal, and the Panel recommended a comprehensive literature review be undertaken to include all epidemiologic studies of the potential effects of atrazine.

The Panel recommended that the draft framework be expanded to include: 1) background information on the target health effects (definitions, overall incidence rates and rates by age, sex and race, established risk factors for each reproductive outcome, including tobacco and alcohol use, body mass index, nutritional status, reproductive history, medications and recreational drug use); 2) a summary of accuracy and completeness of reporting of endpoints from birth certificates; 3) a discussion of temporal and spatial variations in incidence for reproductive endpoints; 4) a discussion of methods used to handle analyses below the limit of detection; 5) an assessment of whether the endpoints are specific and whether their definitions are precise enough to distinguish specific endpoints resulting from different modes of action; 6) an analysis of whether the observed endpoints were compatible with the known reproductive effects of atrazine observed in animal studies; 7) a discussion about feasible modes of action for chemicals in inducing adverse reproductive outcomes; and 8) examples of well-characterized associations

between environmental exposures and reproductive outcomes from epidemiologic studies of other chemicals. Specific comments regarding each of the five studies are presented in the body of the report.

Case Study C

A case study on integrating human incident data into regulatory risk assessment was included to illustrate an analysis of reported human incident cases using diazinon, a pesticide that has historically been used in residential settings (Attachment C). The Panel's consensus was that little weight should be placed on self-reported incident data in risk assessment. Although human incident data can sometimes be useful in providing information on trends or differences in the frequency and severity of symptoms and whether human effects are consistent with those observed in toxicological experiments or epidemiologic studies, the limitations of using human incident data for risk characterization and risk assessment outweigh the advantages. The major limitations include: 1) likely under-reporting of cases due to the lack of mandatory reporting other than for registrants; 2) uncertainty regarding the exact exposure conditions and amount of the chemical to which the individual was exposed; 3) the fact that human incident data largely capture only acute events and not events with long latent periods or those associated with long-term exposures; 4) the lack of specific training for persons recording information; 5) the non-specific nature of some symptoms and the possibility that these are associated with other illnesses or physiological responses to stress, and 6) the utility of self-reported human incident data only being applicable to pesticides with notable acute toxicity.

The recognized strength of this type of data, in contrast to information from animal toxicity studies, is that responses in humans are detected under real-life situations, with conditions of differential individual sensitivity, modifying factors and other influences possible in the human population. Surveillance for unanticipated effects in incident reports could be useful in identifying alternative mechanisms of action not previously described.

However, the Panel felt that the limitations of the incident data for diazinon out-weigh the possible application of such data for risk characterization. The diazinon case study, as presented by the Agency, is unique because of the distinct symptoms resulting from cholinesterase inhibition and because of the risk mitigation measure of removing diazinon from residential use and the consequent reduction in incidents. Other pesticide groups, such as the triazine herbicide family, that do not produce symptoms of acute toxicity would probably not generate usable incident data for the following analyses.

In conclusion, the Panel recommended that incident reporting data, such as those considered in the diazinon Case Study, could be used qualitatively for problem formulation and hazard identification in the risk assessment process, but their application in risk characterization is very limited unless follow-up information and or laboratory data from individual incident cases become available.

Case Study B

A case study concerning the Agricultural Health Study (AHS), a prospective cohort study of approximately 89,000 licensed pesticide applicators and their wives in Iowa and North Carolina

conducted by the National Cancer Institute (NCI) [Michael Alavanja, principal investigator] was also developed as part of the draft framework. Overall, the Panel concluded that the use of data from a well-designed and carefully executed prospective cohort study, such as the AHS, provides the best opportunity for incorporating epidemiologic data into risk assessment. Further, they also may be useful in comparing dose-response data between humans and laboratory animals for some outcomes. The AHS has the potential to extend the use of epidemiology to risk characterization for agricultural chemicals. Such advances in risk assessment methodology could enhance the usefulness of the risk assessment paradigm for the eventual protection of public health.

However, substantial challenges exist in incorporating exposure data from even a well-conducted prospective cohort study such as the AHS into the risk characterization paradigm. The eventual resolution of large discrepancies between epidemiologic and animal studies in apparent dose-response relationships, the form in which the data are used (categorical assignment of exposure in the AHS versus dose estimates on a continuous scale in OPP) or substantial differences in types of responses between animal and epidemiologic studies, is unclear from the draft framework. If epidemiologic data are to be used to derive quantitative values in risk assessment, determining a process for decision-making in cases in which wide differences are observed in dose-response relationships between animal and epidemiologic studies could clarify the framework and its implementation. At present, it can be argued that in most cases, animal studies can more "robustly" describe dose-response relationships and therefore may currently provide a more reasonable approach for characterizing dose-response relationships and for evaluating mode of action than studies in humans. This does not rule out the possibility that in specific situations epidemiologic data could play an important role in either defining or directly contributing to estimates of departure points and risk.

Therefore, the Panel supports the efforts of OPP and ORD to collaborate with scientists at NCI and the National Institute of Environmental Health Sciences to: a) compare the exposure algorithms used by OPP and the AHS, and b) consider temporal relationships for multi-chemical exposure in the AHS. This work is at a relatively early stage of development but appears to be on track with respect to integrating the exposure data obtained from the AHS with the Agency approaches to quantifying worker exposures.

The Panel recognized the merit of finding commonalities between the Agency's exposure assessment methodology and the AHS exposure metrics. The Agency's method results in estimates of workers' exposure as an input into risk assessment that form the basis for setting exposure limits for workers. Finding commonalities to the AHS exposure metrics would provide a way to extend the usefulness of this large and growing database for the protection of pesticide users.

However, the Panel recommends that this section of the document be revised and streamlined. The AHS exposure metrics and scores, and methods of calculating these values are available in the literature (Dosemeci et al., 2002). However, the Agency's method is less clear. The calculation of exposure (dermal, as expressed in mg a.i./day) using the PHED database could be clarified. Further discussion of the unit exposure parameters can also be added. The Panel also recommended that discussions of the variability and uncertainty associated with the foundational

databases be included for each method, e.g., PHED, AHS's self-reporting and input parameter values.

PANEL DISCUSSION AND RESPONSE TO CHARGE

Agency Charge

1. Draft Framework for Incorporating Human Epidemiologic & Incident Data in Risk Assessment

OPP's draft framework describes a proposed weight of the evidence (WOE) evaluation that integrates science on exposure, pharmacokinetics, and mode of action derived from experimental animal and human *in vivo* and *in vitro* studies. This proposed WOE uses the "source-to-adverse outcome pathway" and the modified Bradford Hill criteria like that in the Mode of Action (MOA) Framework (Section IV of Draft Framework) as tools for organizing and evaluating these diverse types of data to determine the evidence available on the potential human health consequences of pesticide exposures.

Question 1.1 Section II of the draft framework describes the major types of *epidemiology studies* along with their strengths and limitations, factors to consider when reviewing epidemiology studies, and ways to use epidemiology data in risk assessment. Please comment on the soundness and completeness of these discussions. If appropriate, please include comments on additional factors for OPP to consider when evaluating the quality and weighing the utility of epidemiology studies in risk assessment/characterization.

Panel Response 1.1

General Responses for Section II

The Panel commends EPA for its effort to incorporate human data in human health risk assessments in a transparent manner, based on the draft framework. An expanded analysis and rewrite of the extent and nature of reliance upon epidemiologic or incident data within OPP for a range of pesticides in addition to the draft framework and associated case studies on individual pesticides would be informative. Review of the nature and extent of reliance upon human data in risk assessments more broadly within the Agency and as a basis for development of relevant guidance in other regulatory programs is also encouraged. Acquisition of epidemiologic expertise for the review and interpretation of human data, including assessment of exposure in relevant studies, is also strongly advised.

Based on experience in studies of toxicity of industrial chemicals and environmental contaminants, weighting of different types of human data varies and depends on the nature of the studies and on the results (i.e., the weight of evidence of an effect in humans). For example, if sufficient weight of evidence for causality of an adverse outcome in humans exists, and this is based on robust epidemiologic data with well-characterized exposure data, these data would be preferred to those obtained from laboratory animals for dose-response characterization. Further, results of epidemiologic studies in which exposure was well-characterized for a relevant effect in humans could be used as a basis to "bound" dose-response estimates from animal studies.

The lack of pesticide-specific context within the text of the draft framework document (with the possible exception of reference to pesticide-specific exposures) results in rather a general (non-context specific) overview of the strengths and weaknesses of various types of epidemiologic studies. This often involves referencing of generic sources of information on the advantages and disadvantages of various types of epidemiologic studies. However, more recent editions of many of the references and other frequently used sources are available (e.g., Rothman, Greenland and Lash, 2008; Gordis, 2008; Rothman, 2002; Szklo and Nieto, 2007).

Presentation and interpretation of study results need to be considered in evaluating evidence from epidemiologic studies. Authors often base their interpretation on whether a finding is statistically significant, regardless of the magnitude of the association as measured by the odds ratio, prevalence ratio, relative risk, or regression coefficient and regardless of the statistical power for evaluation of each adverse outcome. Therefore, very small but statistically significant associations may be emphasized while larger, but non-significant effects may be ignored. Variations based solely on whether a finding is statistically significant may be emphasized, rather than on the basis of the magnitude of the effect. For example, in at least one investigation in the Case Study B included with the draft framework, an effect measure (correlation coefficient) was not reported because it was not statistically significant. This is inappropriate; focusing solely on statistical significance has the potential to lead to misinterpretation of consistency of reported associations between exposure and effect.

Sensitivity analyses are not uniformly conducted in most epidemiologic studies. Sensitivity analysis can be used to estimate the impact of biases, such as exposure misclassification and potential confounding by known but unmeasured risk factors. EPA should incorporate sensitivity analyses in its list of criteria for reviewing epidemiologic data for risk assessment purposes. When reviewing studies, it is not sufficient to state simply that the study may have suffered from unmeasured confounders or the failure to control for measured confounders. One must first make the case that the risk factors in question are actually confounders, and if it is likely that a confounder may have affected the study results, and that the impact is important enough to affect the interpretation of the findings. In general, the Panel felt that substantial confounding, of a magnitude adequate to change a moderately strong or strong risk estimate to a null finding, is rare in epidemiology.

Specific Responses for Section II: Reviewing Epidemiology Studies for Use in Pesticide risk Assessment

EPA should consider the following issues in reviewing epidemiologic studies for use in risk assessment:

a. Was the epidemiologic study conducted primarily in a hypothesis generating or a hypothesis testing mode?

b. Was the method of assessing exposure accurate and reliable?

c. Were inclusion and exclusion criteria clearly stated and reasonable to provide a representative sample with regard to exposure and health outcome so as to provide a relatively unbiased and representative estimate of effect?

d. Was the method of assessing and the criteria for determining the health outcome clearly stated and valid and reliable; e.g., confirmed with histopathology, and were they designed to detect newly diagnosed (rather than prevalent) cases so that it was reasonably possible to determine that exposure preceded disease?

e. Was appropriate information on potential confounding factors, such as socio-demographic, behavioral and dietary factors collected for both exposed and unexposed groups or for cases and controls in the same way, and were they appropriately controlled in the analyses of the data? Were data on co-morbid conditions collected? (i.e., factors that are associated with the health condition of interest as well as factors associated with exposure)

f. Did the study sample the population or individuals of interest? (i.e., was selection bias minimized and generalizability optimized?) How does the study population relate to the universe of potentially exposed populations?

g. Did the study examine individuals with a wide range of exposures? (i.e., ability to detect a dose-response and to generalize to other populations) Did the study include unexposed populations or individuals?

h. Did the exposures examined in the study relate to past or current situations? (e.g., acute versus chronic exposures and the targeted health end points)

i. Did the study have adequate statistical power to detect meaningful differences for outcomes between the different groups of exposed and unexposed or less exposed individuals while controlling for important confounding factors? Does the sample size take into account the expected incidence of the target health effect in the study populations? (e.g., Page 13, 7th bullet of the EPA Draft Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment [January 7, 2010] – specify the statistical power of the sample size to detect an effect after adjusting for confounders). Was the study powerful enough to detect as statistically significant meaningful differences while adjusting for confounding variables and exposure measurement error which typically reduce statistical power?

Comments Relevant to Types of Epidemiologic Studies

Case-control studies – These studies generally involve fewer participants than studies of cohorts or dynamic populations. Controls are not always "without a particular disease" (page 14 of draft document). Some control series consist of participants with other diseases not of interest, and some control series consist of a random sample of the study population without regard to status of the disease of interest (so that it is possible for a case to also be a control).

The appropriate selection of cases and controls is important to avoid selection bias. Specific considerations in the assessment of case-control studies include:

a. Determine whether a clear case definition and inclusion and exclusion criteria for the case group have been provided. Case definitions and criteria provide a basis for the minimization of bias and must be sufficiently specific so that cases of specific diseases that have disparate etiologies are not included. For example, although cervical cancer and uterine cancer are both cancers of the reproductive system, they have different etiologies and risk factors so that in a study of reproductive system cancers, the lack of specificity will result in the decrease in risk estimates for a cancer when a true association exists when other cancers for which no association exists are included in the analysis.

b. Similarly, determine whether inclusion and exclusion criteria and sampling techniques result in a control group that is representative of the population that gave rise to the cases.

c. In the selection of cases for inclusion, determine whether the case group includes newly diagnosed (incident) cases or existing (prevalent) cases. Incident cases will be more useful in establishing the likelihood that exposure preceded disease onset and was not related to survival.

d. Determine whether the collection of data on exposures and confounding factors is identical for cases and controls. Where possible, the data collected should enhance the likelihood that the exposure preceded disease onset, e.g., collecting biological samples from cases and controls to ascertain exposure levels may reflect recent exposures that may be due to cases modifying their behaviors because of illness, treatment or altered metabolism due to disease and would thus be less relevant. Validation of exposures and measurement of potential confounding variables to minimize recall bias strengthens case-control studies.

e. Case-control studies, as well as historical cohort studies and cross-sectional studies, may estimate exposures in the past using monitoring data combined with sophisticated modeling techniques ("historical exposure reconstruction"), or in the case of occupational studies, individual, area or plant monitoring data integrated into a job-exposure matrix.

f. Determine whether participation rates of both cases and controls were sufficiently high to minimize the likelihood of participation bias. Eligibility and participation rates and comparison of important covariates (e.g., socioeconomic status, demographics, employment, comorbidity, etc.) should be compared between cases and controls to assess the potential for selection bias.

Cohort Studies – Cohort studies can be conducted retrospectively (historical) or prospectively. Occupational cohort studies most generally take the form of the historical cohort study. The Agricultural Health Study (AHS) is a longitudinal, prospective cohort study in which members are followed over time until they die or the study ends.

An important distinction must be recognized between prospective, population-based and occupational cohort studies. In the former instance, the cohort participants are drawn from a sample of the general population, ideally representative at the time of selection, and then followed over time. In the latter case, members of an occupational cohort are drawn from worker records and do not represent the general population.

"Population-based cohort studies" are those in which that the study group is obtained from a dynamic population such as the population of a state or municipality. Some cohort studies evaluate cohort members only during the period of their residency, leading to ascertainment error. For example, cancers occurring in former residents of a state are not captured by that state but by the state in which they now live. If they leave and are not followed up, this may be a source of selection bias.

Some of the considerations relevant to the assessment of cohort studies include:

a. Determine the extent of attrition or loss to follow-up (especially differential attrition which could lead to selection bias). Determine whether a high rate of participation was maintained longitudinally with minimal attrition so as to minimize participation bias.

b. Potential observer bias can occur if observers are not masked to the exposed/unexposed status of individuals and/or the hypotheses under study, particularly when the outcome(s) involve(s) some subjectivity. Determine whether observers or those who determined the health outcomes of interest were masked as to exposure status of individuals so as to minimize observer bias.

c. Determine whether the criteria for diagnosis/identification of cases are applied consistently in exposed and unexposed populations, assessed with the same frequency in both groups and handled consistently over time.

d. Determine whether appropriate analyses that maximize the use of data for participants who withdraw, move or are otherwise lost to follow-up have been conducted.

e. Determine whether the assumptions of longitudinal analytic approaches are actually met and that these analytic approaches are used appropriately.

f. Determine whether exposed and unexposed individuals were monitored or measured at the same intervals and in the same ways.

The deletion of 'for rare diseases' in the description of cohort studies is suggested. Prospective cohort studies are not efficient for studying rare diseases even when they contain large study populations such as the Agricultural Health Study (AHS), which will have adequate power to evaluate risks for frequently occurring or less rare cancers but substantially less power for rare cancers and other uncommon outcomes.

On page 14 of the EPA Draft Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment (January 7, 2010), the text states that groups of exposed and unexposed cohorts are studied over time. However, some cohort studies compare the exposed cohort to U.S. (or state) populations rather than unexposed individuals to calculate standardized mortality or morbidity ratios.

Cross-sectional studies – Exposure information is not necessarily obtained "at the same point in time" as the outcome nor is it always just a characterization of exposures around the time that the outcome is measured (page 14). Many cross-sectional studies use historical information on

exposure, which is relevant in relation to temporality. The key distinguishing feature of a cross-sectional study is the measurement of prevalence of disease (e.g., birth defects, SGA), symptoms, biological/physical and physiological response measurements (e.g., pulmonary function tests, blood pressure, chest x-ray, clinical examinations, liver and kidney biomarkers). The ability to conduct screening to make measurements which would not be routinely collected is an advantage of the cross-sectional study.

Prevalence is the proportion of individuals in a population that has disease (Rothman, 2002). Prevalence can be determined as "point prevalence" (e.g., measuring PFTs at a particular workplace at a particular time), or as "period prevalence (e.g., the proportion of cases of autism among those residing in a town during 1998). Cross-sectional studies do not involve a follow-up period (prevalence is a proportion not a rate). Cross-sectional studies are not longitudinal unless one does a series of cross-sectional studies of the same population. In contrast, incidence (the number of new cases divided by the total number of persons at risk for that disease in a population during a specified time period) involves a follow-up period (i.e., it is longitudinal).

A cross-sectional study includes only those who are present when the event takes place (e.g., the pregnancy must result in a live birth; the worker must be currently employed; people who moved out of the town prior to 1998 are not evaluated). A major drawback is that such studies involve "survivor populations" and do not evaluate those who, for example, have left a workplace because they became ill from the workplace exposure. Another major drawback involves the nature of prevalence. Prevalence is a function of incidence and duration. For diseases of long duration, it may not be clear whether the exposure of interest increases the risk of disease or prolongs the duration without increasing the risk of the disease. Incidence is usually more relevant for the types of effects that are generally considered in relation to long term effects of chemical exposures.

Hybrid Designs – Some study designs in which case and control samples are drawn from cohorts as they proceed over time ("case-cohort") or at the end of follow-up ("nested case-control") are not mentioned in the text. These designs can be very efficient and have considerable potential for detailed exposure assessment. Investigators may have collected biologic specimens for evaluation of biomarkers before cases occur. Because these designs are nested within cohorts, exposures can be measured and biologic samples collected at baseline and repeatedly over time before follow-up for disease occurrence. Therefore, there is the potential for less bias in assessing exposure. Another study design that may become important for pesticide research is the case-crossover study design.

Ecologic Studies – First, a single ecologic study cannot provide strong enough evidence to establish a causal relationship (page 15 of the EPA Draft Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment [January 7, 2010]). This is the fundamental premise underlying the Bradford Hill considerations for weight of evidence, particularly the criterion of consistency. Rather, ecologic studies (especially those with good design), suggest hypotheses for further research (i.e., are "hypothesis-generating"). In practice, ecologic studies are sometimes conducted as a crude approach for testing hypotheses by evaluating correlations but their limitations are well-recognized.

Second, an important distinction must be made between ecologic (group-based) and individual-based studies, particularly with reference to the exposure assessment. One can do a study of individuals and use ecologic data (group data for county, town, or census tract) to assess exposures. However, in ecologic studies, no information is available on whether the people who are diseased are the ones with exposure, while in individual studies, that information is available. For example, a study of disease rates by contamination levels in water on a state basis might be ecologic with respect to the exposure assessment, but the outcome has been obtained at the level of the individual. Thus, the term "semi-ecologic" is sometimes used to describe studies in which the outcome is measured at the level of the individual but the exposure is measured at the level of the group.

The situation in which disease is determined at the level of the individual but exposure is defined by characteristics such as water supply may create confusion in determining the study design. For example, in Case Study A of the draft framework, the Villanueva study (2005) was interpreted as ecologic. In an ecologic study, exposure is assigned to population groups, not individuals. The exposure variable is not representative of any individual's exposure. Instead, the exposure variable characterizes an entire population or geographic area. The ecologic exposure variable is usually of the form of "% of the population with a certain characteristic" (e.g., % unemployed) or of the form "average income". In the case of the two ecologic studies in Case Study A, exposure was characterized on the basis of areas. On the other hand, the Villanueva study assigned exposures to individuals by defining unique water distribution systems with known water sources in which all the population served by each system received similar levels of atrazine (and other contaminants). Although exposure misclassification was likely to exist (e.g., due to bottled water consumption by some women), this was an individual level study.

Ecologic studies are not necessarily geographically-based. For example, a study might compare average rates of exposure in persons who have had special training vs. those who have not had such training or disease rates before and after a major change in exposure.

In addition to the ecologic fallacy mentioned in the text, an additional bias arises from the inability to control properly for confounding factors at the individual level when information is available only at the group level. Finally, in ecologic studies it may be difficult to determine whether exposure preceded the disease.

The results of an ecologic study in which a hypothesis is tested will provide only weak evidence for a causal relationship. This is because of possible ecologic biases, weaknesses in the group-level exposure characterization and uncertainty regarding temporality. However, it is important to assess ecologic studies on the basis of the quality of their design (i.e., some ecologic studies can provide much stronger evidence than others, though the designs in Case Study A are relatively weak). Useful information may be gleaned from an ecologic study if it is well-designed.

Cluster Investigations – The text does not describe this form of investigation. Cluster investigations can be the first alert to a major health issue e.g., vinyl chloride and angiosarcoma of the liver. They can also lead to epidemiologic studies that provide important new information

on the health effects of environmental or occupational exposures, though they are notoriously difficult to interpret, without follow-up.

Meta-analyses and pooled analyses – These are also not described but may be conducted when the body of epidemiologic evidence is sufficient. Such collective analysis can make additional contributions, particularly when the statistical power to detect meaningful effects in individual studies is low.

Important factors to consider when evaluating epidemiologic studies for use in risk assessment

Exposure Assessment

It is important to distinguish approaches which can lead to quantitation of exposure from approaches to exposure assessments that are more qualitative in nature. The quality of the exposure assessment is the principal determinant of the impact of epidemiologic studies in dose-response analysis and risk characterization in risk assessment.

This section in the draft framework (page15) appropriately emphasizes the difficulty of evaluating low exposures. However, it is equally important to stress the need for exposure information sufficient to characterize the time period when the exposure would be likely to have its effect on the outcome of interest. In other words, the timing of exposure may be equally if not more important than the level or duration of exposure. In most instances, the relevant exposures occurred in the past (maybe distant past for cancers and other chronic diseases). "Direct" approaches, such as biomonitoring and personal monitoring, are generally not useful for characterizing prior exposures unless the contaminants of interest are very persistent (i.e., bioaccumulate, long half-lives of excretion). However, historical exposure reconstruction based on available data and sophisticated modeling techniques often contribute significantly in estimating past exposures sufficient for risk assessment purposes and additional consideration of these approaches in this section of the draft framework is suggested.

Historical records and questionnaires – Most often, these approaches do not estimate quantitative levels of exposure but rather, serve as the basis to assign categorical levels. Occasionally, however, quantitative levels of exposure are estimated on the basis of proxy measures (e.g., duration of exposure, pounds of pesticide applied over a particular time). It seems important to emphasize that even when quantitative measurements are available (e.g., personal monitoring), these are often categorized in order to avoid use of a continuous variable in an exponential model (i.e., to avoid the strong model assumption that risk increases exponentially with each unit increment over the full range of exposure). This can lead to measurement error and bias, and risk estimates may be biased in unpredictable ways.

Environmental monitoring – These data can be used to estimate quantitative levels of exposure but there should be some understanding of the relationship between potential exposure and dose. Another advantage not mentioned in this paragraph is that often a large amount of monitoring data is available over time, including data obtained in the past when the exposure was relevant to the outcome under evaluation. In contrast, although biomonitoring reflects internal exposure, it

may provide only a "spot" evaluation of current exposure, but may be more closely related to the adverse outcome of interest in the source to outcome pathway.

Personal monitoring – One disadvantage of personal monitoring not considered in the draft framework is the lower likelihood of having information to characterize exposures during the relevant time period, usually in the past. Also, it is unlikely that the full range of exposures over time will be represented, and sampling may not be maintained over a sufficiently long period to capture peaks and fluctuations. People may change their behavior when they wear personal monitors, and there will be behavioral differences between individuals that will affect personal exposure monitoring results. In other words, personal monitoring may need to be supplemented by environmental monitoring, biological monitoring and/or interview or questionnaire information (e.g., daily diaries).

Biomonitoring – The same disadvantages noted for personal exposure monitoring apply here and some of the disadvantages mentioned in this section also apply to personal monitoring (e.g., sampling over short duration or at one point in time, possibly not measuring the right chemical or metabolite, measuring other chemicals or metabolites, past exposures)

Biomarkers– The selection of relevant biomarkers of exposure and/or effect, based on the toxicological properties of the pesticide (particularly on mode of action) enables much greater likelihood of meaningfully integrating the epidemiological and toxicological databases. (See also comments above on environmental monitoring).

Confounding Factors

The discussion on page 18 of the draft framework is very general and limited, with inadequate detail to enable the reader to understand how "OPP will consider whether relevant confounding factors are properly identified, described, measured and analyzed so that an unbiased estimate of the specific association under study can be made". What aspects and factors will be considered? Some comments with potential relevance to this discussion, particularly in relation to addressing confounding in the evaluation of relevant studies for risk assessment purposes, are presented below.

It is important to define and distinguish clearly between confounding, effect modification, synergy and other mediating effects of covariates. Definitions of confounding should explicitly address the requirement that the variable produces a distortion in the effect estimate either towards or away from the null value. For a variable to act as a confounder it must satisfy three criteria: (1) it must be associated with the disease of interest; (2) it must be associated with the exposure under analysis; and, (3) it does not lie on the causal pathway between exposure and disease. Further, the relationship between the confounder and the exposure or outcome of interest does not have to be statistically significant to have an impact on the risk estimate for the main effect.

The potential for confounding is often mentioned in critiques of epidemiologic studies, but rarely is an argument presented on the likely size of the impact of the bias. In practice, substantial confounding occurs only rarely. The classical case, in which smoking acts as a confounder of an

occupational exposure-lung cancer association, has been found to produce little effect (usually a change of 20% or less in the relative risk). It should be emphasized that a confounder must be a relatively strong risk factor for the disease and be strongly associated with the exposure of interest to create a substantial distortion in the risk estimate. It is not sufficient simply to raise the possibility of confounding; one should make a persuasive argument explaining why a risk factor is likely to be a confounder, what its impact might be, and how important that impact might be to the interpretation of findings.

If unmeasured confounders are thought to affect the results, researchers should conduct sensitivity analyses to estimate the range of impacts and the resulting range of adjusted effect measures.

When deciding whether to include a potential confounder in a regression model, it is important to make sure that the factor is actually a risk factor on its own and not only related to the exposure of interest. Adjusting for a factor that has an association with the disease of interest wholly or partly because of its association with the exposure of interest will attenuate an exposure-disease association, if one truly exists. Adjusting for season of conception may be such an example. Although several reasons are possible for the seasonality of adverse birth outcomes (e.g., disinfection byproducts in drinking water, nitrates, other fertilizers, air pollutants, SES factors), it is also possible that some of the association between season of conception and adverse birth outcomes may be due to, for example, exposure to a pesticide itself. If this is the case, adjusting for seasonality will attenuate the pesticide-adverse birth outcome association (i.e., bias toward the null). In an investigation included in the Case Study A, a variable for "farming exposure" consisting of the proportion of crop-planted land around a home was added to the regression models. Again, this could lead to attenuation of an exposure-disease association because, if it is associated with the outcome of interest, this is likely to be due to it also being associated with the exposure of interest and not a risk factor on its own.

Additionally, the lifestyle factors mentioned in this section as potential confounders are rather limited in scope. For example, confounders may include dietary factors other than high energy diets; physical activity may not be just inactivity; and other factors (e.g., genetics, comorbidities, medications, alternative therapies, alcohol consumption, etc.) might be potential confounders.

When collinearity exists between contaminants (e.g., trichloroethylene and perchloroethylene often occur together in drinking water, or pesticide mixtures containing several chemicals), it is difficult to disentangle the effects of the individual contaminants. If a chemical that is correlated with the chemical of interest is also a risk factor for the disease being studied, then it can create confounding. "Residual confounding" occurs when the additional exposure(s) or other confounding factors are not measured and controlled in data analyses. Studies should include a discussion of residual confounding (and possibly a sensitivity analysis) if the impact may be important enough to affect the interpretation of the findings.

Finally, it should be emphasized that errors in the measurement of a confounder (i.e., confounder misclassification) may result in the inability to provide adequate control for its confounding effects.

Effect Modification

It is important to separate issues of confounding from effect modification. Confounding is a bias that investigators seek to minimize. Effect modifiers are variables that change the magnitude of the association across strata of the study population, e.g., age, race/ethnicity, socioeconomic status, genetic polymorphisms. Investigators evaluate effect modification by the examination of interaction terms in multivariable models or by stratification of the data across levels of the modifier. Effect modifiers may or may not be confounders; e.g., smoking may be a confounder in studies of airways disease but may also be an effect modifier if the risk of exposure to an air pollutant is higher among smokers than non-smokers. Investigators search for evidence of effect modification because differences in risk across strata of the population may provide important information in evaluating the association between exposure and the effect of interest and may be important in identifying susceptible populations. Unless a study is designed specifically to evaluate effect modification (e.g., smoking-asbestos interaction for lung cancer), it may lack statistical power to evaluate such an effect if a sufficient number of individuals are not available in each stratum.

Misclassification of Exposure and Outcome

The text on page 19 of the draft framework should make a clear distinction between differential and non-differential exposure misclassification. With respect to non-differential exposure misclassification, care should be taken to differentiate the direction of the distortion of the effect estimate toward or away from the null depending on the categorization of exposure (dichotomous vs. multiple levels of exposure). A table to illustrate the effects of non-differential exposure misclassification, such as those contained in several texts, would be helpful to illustrate the changes in the odds ratio or relative risk that occur with modest, but anticipated and relatively frequently occurring degrees of misclassification in the range of 10 to 20 percent.

Methods for assessing the impacts of exposure misclassification bias, selection bias, and confounding bias exist. Inclusion of these in relevant studies should be encouraged. The "healthy worker effect" is not mentioned in the text and can create an important bias in occupational studies. The "healthy worker effect can lead to bias toward the null and below, creating the interpretation that the exposure is "protective".

Disease misclassification bias should be elaborated in the text. In the studies involving birth defects included in Case Study A, the disease misclassification resulting from the use of birth certificates to ascertain birth defects was probably non-differential, resulting in a loss of statistical power. However, it is possible that the bias could have been differential with respect to exposure status leading to bias either toward or away from the null.

Issues in Statistical Analysis

The discussion on page 19, related to statistical analysis, should be expanded to include several additional points. When an outcome under evaluation is rare or the sample size is relatively small, it is possible to introduce "statistical bias" in the analysis by including too many covariates in the model. The resulting effect estimate may be biased in either direction but is

unlikely to reflect the true estimate of effect. Statistical bias is also possible when conditional methods are used (e.g., conditional logistic regression when the design includes matching of the comparison group to the group under study). If too few discordant pairs (or discordant sets) are observed (i.e., the number of discordant pairs is small, thus analogous to the situation of a small sample size), statistical bias can also occur, producing a biased estimate of effect. Although it may be important to control for potential confounders, one must take care not to over-control or to end up with cell sizes that are so small that statistical bias is introduced as a result. In such situations, it may be more important to seek parsimonious models that adjust only for the most influential confounders so that the effective sample size is adequate to address the research question without introducing bias due to the statistical modeling.

In evaluating statistical results, it is important to consider the magnitude of the association as measured by the relative risk, odds ratio, risk ratio, regression coefficient, etc. Strong relative risks are unlikely to be due to unmeasured confounding, while weak associations may be due to residual confounding by variables that the investigators did not measure or control in the analyses.

The draft document should also include a discussion of statistical significance in the context of the clinical/biological/scientific significance of the result and the difference between biological and statistical significance. Some statistically significant associations may have little clinical or biological relevance; conversely, some associations that fail to meet the criteria for statistical significance (which are somewhat arbitrary) may be important clinically or from a public health perspective and merit further investigation, especially when the association is strong (but imprecise).

Interpretation of null studies

Exposure measurement error resulting in exposure misclassification is the most likely cause of null findings when an association truly exists, followed by lack of statistical power due to inadequate sample size to detect small effects or to include sufficient numbers of individuals with the health outcome(s) of interest (page 20). In the studies of birth defects considered in Case Study A, ascertainment was based on birth certificates, likely resulting in substantial under-ascertainment that would reduce statistical power to detect meaningful differences.

In addition to the factors described above, a study may be "null" because the exposure is below a threshold at which an effect would occur or be detected. An assessment of the study's statistical power to detect the magnitude of effect of interest is important in interpreting null results. Information on mode of action as a basis to assume site concordance is critical in interpretation of results for risk characterization. If adequately addressed, such results can be used to "bound" dose-response estimates from toxicological studies.

Publication bias related to exclusion of null studies from the literature is discussed on page 24.

External validity

The issue of generalizability in etiologic research (page 20) concerns whether exposures were similar (dose, timing, duration, etc.) and whether important effect modifiers (e.g., sensitive or vulnerable populations) were considered. It is not only an issue of whether a sample is "representative" of the larger population of which it is a sample. Therefore, if exposures are similar, the results found for agricultural workers in NC and Iowa should be fairly generalizable to other agricultural workers in other states, unless important effect modifiers are present that are distributed differently among agricultural workers in NC, Iowa and/or other states (e.g., CA), for example, racial/ethnic distribution when genetic predisposition may modify the effect of exposure. The results of the AHS may not be generalizable to migrant farm workers and their families, because these populations may have a different distribution of risk factors than the AHS population, including race/ethnicity and potentially lifestyle factors and comorbidities, that can act as effect modifiers in addition to their exposures being different (e.g., they may have more intense exposures to pesticides).

Benefits and uses of epidemiologic data in human health risk assessment

This section describes well some uses and benefits of epidemiologic data for risk assessment (i.e., hazard identification/characterization, exposure characterization, and dose-response characterization). Among the unmentioned advantages of epidemiologic studies is that they evaluate the actual conditions of exposure in human participants. On the other hand, in animal studies drinking water exposures are simulated often solely by oral administration, sometimes at unrealistic doses for humans, and occupational exposures are similarly simulated by a single exposure route, usually inhalation.

On page 20, the first paragraph under (C) contains a statement about how "high quality studies with robust exposure assessment may be used to estimate risk quantitatively". This is subsequently qualified to indicate that "most epidemiology studies suffer some limitations in size, scope, exposure assessment or data analysis which prevent their use in quantitative risk assessment" (referenced to Calderon, 2000). Although some epidemiologic studies may not be useful to quantify risks, others have been used for this purpose (e.g., occupational and drinking water studies have been used in trichloroethylene risk assessments, studies of occupational exposure to radon have been used to assess lung cancer risk). As indicated in the general comments included at the beginning of the response to this subquestion, it would be informative to provide an analysis of in what circumstances and how epidemiologic data have been used in risk assessment (for pesticides, specifically and more generically, for broader U.S. EPA program mandates). One example of data from an epidemiologic study having been used to inform the outcome of risk assessment in a quantitative manner is the NIOSH dioxin study (Fingerhut et al., 1991).

Question 1.2 Section III of the draft framework describes the major sources of *human incident data* along with their strengths and limitations. Section III also describes ways to use human incident data in risk assessment. Please comment on the soundness and completeness of these discussions. Please include comments on additional factors to consider when evaluating the quality and weighing the utility of human incident data in risk assessment/characterization.

Panel Response 1.2

Information on how OPP uses human incident data is very general. Case Study C is also fairly unique and as a result, has limited applicability. As indicated in the general comments in the response to Question 1.1, it might be helpful to include an analysis of how incident data have been used elsewhere in the Agency. In particular, some examples as to how this information has indicated the need for a new risk assessment or risk management (as specified in the report) would be helpful.

The description in this section presenting the toxicity data within these human incident reports seems mainly focused on their severity ranking. However, when data are used in risk assessment, the nature of the toxicity endpoints is important, and, while this was discussed in the draft document with respect to the diazinon case study, greater clarity in the text of Section III would be helpful. Most importantly, it should be emphasized that the utility of these data within the framework of risk assessment can be maximized when evaluated in the context of possible mode(s) of action and any related *in vitro* data.

In this context and in relation to potential other uses of the human case reports and surveillance of acute poisonings, incident data can be helpful in considering similarities of site concordance of target organs between animals and humans. This is important in mode of action/human relevance analysis. Use of these data is not restricted to hazard identification, but extend to hazard characterization. Other possible uses of the data include identifying vulnerable or sensitive population subgroups (e.g., age, gender, occupation and demographics), albeit not at molecular levels. This may be helpful as a basis for refining the risk assessment to ensure adequate protection for all subgroups or possible exposure scenarios, or for targeting safety policies or outreach for the reduction or prevention of poisoning events.

In view of their limitations, however, reliance on incident reports in quantifying risks is necessarily limited. Incident reports usually involve high doses and frequently involve illegal or accidental exposure, and, as a result, they will not be reflective of normal use exposures. The incident data are frequently of limited detail and largely the observations of non-medically trained individuals, based on short term exposure with only limited follow-up. The exposure estimates in these cases are normally semi-qualitative at best, and of limited reliability. In addition, they generally involve exposure to products rather than single chemicals, so the possible interactions of the main active ingredient with other chemicals are unknown. Also, probably little, if any, information is available in incident reports to indicate the nature of other contributing factors or confounders. In addition to those mentioned above, the uses cited for incident data (i.e., need for changes in risk management, monitoring success of mitigation

measures and targeting enforcement activities), are all reasonable, assuming their reliability has been assessed critically.

Of special concern in the interpretation of incident data is the reporting of symptoms by medically untrained individuals and without specific disease criteria. When an accidental exposure occurs at a potentially high dose, it is likely that reported signs and symptoms may reflect physiologic responses to the fright induced by the situation rather than the effects of the chemical. Caution is urged in the interpretation of symptoms that could be attributed to physiologic stress reactions if these are not consistent with the plausible toxicological effects for a chemical. The classic flu-like symptoms that are frequently cited as an acute adverse consequence of exposure to some pesticides could also be attributed to some non-chemically-induced causes.

The several sources of incident data described by OPP also vary substantially in their completeness, level of description and geographic scope. EPA has reasonably evaluated the utility and reliability of these five data sources, and the summary in Table 3 is particularly helpful. Additional factors to take into consideration when evaluating the quality of human incident data and their utility in risk assessment/characterization are:

a. Whether the reporting system is active (i.e., the repository agency for such information seeks out reports or incidents by contacting the relevant parties [e.g., officials, health care providers, workers, manufacturers and owners] on a regular basis) or passive (i.e., depends on the relevant parties to report to the agency or repository body).

b. Whether reporting is mandatory, (i.e., requires certain officials, physicians and other health care providers, applicators, manufacturers, farm owners, etc. to report any incident) or voluntary and who is required to report.

c. Greater clarity about how the data are used for estimating "trends over time"; more weight might be given to incident reporting that tracks pesticide use over time so that more or fewer pounds of use should correspond to more or fewer incident reports (assuming all other things such as protective gear, weather, characteristics of users or those exposed remain unchanged).

In addition to the need for analysis of validity and reliability for incident reports, and considering the limitations mentioned above, it is possible that a few individual sets of incident data from which the exposure level can be reliably estimated might contribute to modifying, improving, or confirming the certainty (or uncertainty) of an existing risk assessment based only on animal studies with default uncertainty factors.

Missing from Section III is an explicit statement of intention to estimate or quantify exposure and/or dose for the incident reports. For example, the Panel encourages the Agency to explore linking the California Pesticide Illness Surveillance Program (PISP) data to that in other related databases, such as information in site-specific pesticide use data, to see if the exposure scenarios can be better characterized or confirmed.

Throughout this section, recognition should be explicit that in part, the objective of OPP's work is to avoid incidents. The limited contribution of incident data to quantitative risk assessment needs to be considered in this context.

Question 1.3 Section IV of the draft framework describes a proposed WOE approach for evaluating human and experimental animal data from *in vitro* and *in vivo* studies. This proposed approach makes use of the "source to adverse outcome pathway" and the modified Bradford Hill criteria (like that in the MOA Framework) as tools for organizing, evaluating, and describing the human health consequence of a particular chemical based on the available data. Please comment on the proposed use of modified Bradford Hill criteria in the context of the source to adverse outcome pathway for integrating a variety of types of data at different levels of biological organization including human incident and epidemiologic data in risk assessment.

Panel Response 1.3

The nature and application of the Bradford Hill criteria or viewpoints[a] in the framework addressing source to adverse outcome pathway for integrating a variety of types of data at different levels of biological organization vary from their more traditional consideration in assessing the weight of evidence of epidemiologic data, alone. For the framework integrating human and animal data in the context of mode of action, the proposed criteria or viewpoints appropriately represent those related principally to weight of evidence rather than consideration of individual studies. They have also been appropriately modified to be more relevant to the intended context.

Framed in this context, it is important to keep in mind the original purposes of the "Hill" criteria. As first proposed by Bradford Hill, the principles were meant to bring some structure and rationality to the difficult art of interpreting observational data affected by confounders and other sources of uncertainty. Thus, their adaptation to the specific issue at hand is appropriate because they are rarely used exactly as Hill first proposed them.

Prior to considering the weight of evidence of human and animal data in the context of the framework, the weight of evidence for causality in epidemiologic studies is generally addressed. This takes into account the Hill criteria or viewpoints relevant to consideration of individual studies (e.g., temporality, strength; dose-response). Overall weight of evidence of causality is then considered on the basis of those considerations /viewpoints (e.g., consistency) relevant to the collective database. Some of the criteria/viewpoints are relevant to both (e.g., strength; dose-response).

Prior to conducting a WOE analysis, the investigators should assure themselves that they have accessed the complete body of relevant epidemiologic literature available from peer-reviewed sources. A plan should be developed for the literature search that incorporates second and third level searches in the published literature as well as using the standard approaches of literature searching such as PUBMED (http://www.ncbi.nlm.nih.gov/pubmed/). See further discussion on page 52.

[a] While referenced as the "Bradford Hill criteria", in fact, these criteria or viewpoints first appeared a few months earlier, in nearly the same form, in the 1964 Surgeon-General's report on the health effects of cigarette smoking. (Smoking and Health: Report of the Advisory Committee to the Surgeon-General of Public Health Service. U.S. Dept. of HEW, Public Health Service, GPO, 1964.), and potentially even before. They might more appropriately be quoted as "frequently attributed" to Bradford Hill.

For example, point 1 on page 28 of the draft document refers to the literature search, but does not say how that search will be organized and conducted, nor does it refer to the necessary screening of papers to find those with some merit for the present purpose in, for example, problem formulation. Given that the literature on important public health issues is often vast, complex, largely of poor quality and not well-focused (Bailar 1989), it is recommended that the likely contribution of various sources of data be considered early in scoping as a basis to conserve resources.

Study quality is an important consideration in evaluating epidemiologic data for inclusion in WOE approaches. Criteria for assessing study quality should be explicitly described, though are necessarily somewhat empirical in nature. General guidelines are found in the meta-analysis literature where similar considerations are relevant for selecting studies for inclusion. Elements of study quality may include but are not limited to the following considerations: study design, the existence of an *a priori* hypothesis versus an exploratory analysis, sample size and statistical power adequate to detect the size of the meaningful effect under evaluation, ascertainment of the outcome in terms of sensitivity and specificity, quality of the exposure assessment and the potential for differential and non-differential misclassification, measurement of key potential confounders, assessment of other forms of potential bias, evaluation of effect modification, statistical analysis, the possibility of multiple comparisons unsupported by *a priori* hypotheses, or other supporting data or biological plausibility and others. Many of these issues are introduced on page 28 of the draft framework.

Studies demonstrating no association with a pesticide exposure are equally as informative in a WOE analysis as those that do, provided they meet the criteria for quality described above. Publication bias resulting from rejection or failure to submit 'null' studies is of concern. However, several journals such as Epidemiology have an explicit editorial policy of not rejecting 'null' studies on the basis of the findings.

It is often useful to distinguish between two types of evidence from epidemiologic studies: the quality of the information available and the size of the effects reported. The OPP draft might usefully point this out, and make the intended meaning clear each time the phrase is used.

In the context of assessment of the collective weight of evidence for causality in human studies, alone, the modified Bradford Hill criteria are highly appropriate for framing the description of the likelihood of a specified consequence of a particular chemical using data that are typically available. These considerations are well-accepted for assessing evidence of causation in the field of epidemiology and in public health. It is important to view the considerations not as a checklist, but rather as a group of characteristics that taken collectively provide a systematic way to aggregate observations and guide assessments and conclusions. They require flexible interpretation.

As noted at the beginning of this response, the application of these considerations in the context of assessing the weight of evidence for hazard based on integration of epidemiologic and toxicologic data necessarily varies from their more traditional consideration in determining the weight of evidence of causality from epidemiologic studies, alone. It may be important, then, to clarify terminology in the documentation and to distinguish explicitly potential variations in

interpretation of the considerations/viewpoints in the context of the framework vs. consideration of epidemiologic data, alone.

Biological plausibility needs special attention in this context. In an epidemiologic sense, for example, it sometimes means the integration of everything else one knows at the time of analysis, to develop a sense of the likelihood that a real effect is present. This is the statistician's concept of prior probability, and may be the simplest and most straightforward way to express the concept. However, other interpretations are possible.

In the context of weight of evidence for integration of toxicological and epidemiologic data, biological plausibility takes into account side by side comparisons not only of chemical specific toxicological data including that on mode of action, but integrates what is known from a broader understanding of biologic principles.

Apparent lack of biologic plausibility should not, in itself, be a reason for inaction and the Agency should leave open the possibility that new effects or different manifestations of effects might be revealed in human data. The definition of biologically plausibility evolves over time and may involve a great deal of uncertainty. Biological plausibility based on toxicity studies should not be used to negate contradictory evidence from epidemiologic studies. A potential way of further clarifying the distinction between use of these viewpoints in integrating epidemiologic and toxicologic data versus assessing the weight of evidence of epidemiologic data alone would be to consider differential weighting of the criteria. However, lack of available data may preclude this for specific applications. Establishing dose-response relationships is highly desirable in epidemiologic studies and should be weighted heavily. Temporal association is critically important in that exposure must precede effect and should be weighted accordingly. Strength, consistency and biological plausibility should also be weighted heavily. On the other hand, specificity of the exposure-response relationship has fallen out of favor, particularly in studies of chronic diseases following systemic exposure and should be considered the weakest criterion, if at all.

Based on increasing experience in application of the mode of action/human relevance framework and to avoid the mistaken idea that this addresses exposure in any way, it is suggested to consider revising reference to "dose-response relationships" to concordance of dose-response relationships between the key and end events". Earlier key events are expected to occur at lower doses, and, if this is not the case, the data do not support the hypothesized mode of action. The incidence of earlier key events is also expected to be greater than or equal to that for the end toxic effect; if this is not observed, the weight of evidence does not support the hypothesized mode of action. See Le Marchand (2005) for a discussion of how biological measurements at the cellular or molecular level are being used in cancer risk epidemiology.

Question 1.4 OPP has extensive experience applying the MOA Framework to experimental animal data. However, OPP has not yet completed a WOE approach that also includes epidemiology or human incident data like that proposed in Section IV of the draft framework. Please include in your comments what, if any, additional scientific considerations not discussed in the draft framework OPP should take into account when conducting such WOE analyses.

Panel Response 1.4

The draft framework for integration of *in vitro*, *in vivo* animal and human data has many advantages and the Agency should be congratulated for their efforts. Application of frameworks as a basis for increasing transparency and consistency in the evaluation of weight of evidence undoubtedly has potential for improving risk assessment.

The use of the "source to adverse outcome pathway" and the modified Bradford Hill criteria (like that in the MOA Framework) is also extremely helpful not only as a basis for organizing, evaluating, and describing the potential implications for human health of a particular chemical based on the available data but also in identifying critical data gaps. It is also important in moving the focus in toxicology and risk assessment from late adverse effects to earlier biomarkers of exposure and effect, so that more informative human data at relevant dose levels can be collected. Further, the framework is helpful in directing attention to dose-response relationships for early key events at a very early stage in the assessment of available data; it is these dose-response relationships that are critical in the subsequent risk characterization. The source to adverse effect pathway and framework as proposed in the documentation offers significant potential for the transparent and appropriate integration of human and toxicologic data.

Clear benefit is to be gained, however, in more clearly distinguishing the qualitative and quantitative aspects of a framework analysis as a basis for integration of human data in subsequent dose-response characterization. While pre-existing epidemiologic and incident reporting can be helpful in hazard characterization, unless exposure has been robustly and quantitatively addressed, preferably with inclusion of appropriate biomarkers of exposure and effect based on identification of key events in a mode of action context, the contribution to dose-response characterization will necessarily be more limited (Figure 1 in EPA Epi-Incident Framework Draft document).

Early recognition of the likely contribution of human data in this context in scoping/problem formulation will additionally increase transparency, facilitate peer engagement and necessarily conserve resources. For example, lack of adequate characterization of exposure-response relationships in epidemiologic studies may preclude the need to do an extensive weight of evidence analysis for these data, since they cannot contribute significantly to dose-response analysis and hence, risk characterization.

The value of framework analysis in coordinating assessment and research has also not been emphasized in the documentation. For example, there is repeated reference to problem formulation in the draft framework but without indication of how the broader toxicologic and

epidemiologic databases might be considered in an integrated fashion at this stage as a basis to identify uncertainties and critical data gaps to inform the assessment. This would be an appropriate way to identify limitations of available human data in the context of the overall database as a basis to focus additional research. Appropriate human data, might include, for example, *in vitro* studies in human tissues or cell lines and perhaps focused epidemiologic studies to address specific questions in potentially susceptible subgroups by evaluation of early biomarkers of effect.

Alternatively and/or additionally, scoping of the likely relative contribution of the existing human data (including but certainly not limited to epidemiological data), in the context of the overall database, can be helpful as a basis for engaging peers in the review of preliminary considerations of this information and for focusing resources on additional work to complete the assessment.

In the context of specific content of a framework analysis, based on increasing experience with evaluations of MOA/HR, potential alternatives for hypothesized MOAs would normally be considered at the outset as a basis for distinguishing relevant pathways and key events in an integrated fashion (Page 31, "Other MOAs").

There is also limited reference in the draft framework to how uncertainty will be considered. For example Page 29 refers to "Postulated MOA", but does not deal with the difficulties of determining the MOA with reasonable certainty, or how residual uncertainty should be dealt with in the analysis.

It is important to remember the historical context and purposes for developing the MOA frameworks, which focused originally on cancer outcomes and have only recently been extended to non-cancer endpoints. For decades, positive findings from animal cancer studies were assumed to be relevant for human hazard identification. In the late 1970s and early 1980s research programs were initiated to systematically examine the biologic events that appeared to correlate with and perhaps account for the induction of cancer in a number of common sites for tumor responses in rodent cancer studies. The original framework for mode of action for experimental animal tumor sites and types was established to ensure that the many hypotheses that were being proposed as a basis for considering their relevance were, in fact, based on a solid scientific foundation.

Extension of the framework to consider human relevance represented a logical progression from consideration of the nature of key events in animals to address both qualitative and quantitative aspects of kinetics and dynamics. This proved to be an illuminating exercise for those who participated, and highlighted the extent of erroneous perception that certain animal tumor types were not relevant for humans based on the expectation that humans would not be exposed at sufficient levels to develop tumors, rather than on true differences in physiology or biology.

The most important aspect of the framework for assessing the human relevance of tumors in animals is that in the absence of sufficient weight of evidence that an animal cancer MOA could not occur in humans, the default assumption is that the animal cancer finding is relevant for human health assessment. In this context, it is important that the proposed framework for

incorporation of data from *in vitro, in vivo*, human incident and epidemiologic data does not imply that inconsistent findings in any one area could lead to inaction on the part of the Agency.

Consider, for example, the relevancy of a particular endpoint in a high throughput screening assay to the toxicity pathway that is under consideration or the observation of a strong tumor response in an organ that doesn't exist in humans; e.g., the Harderian gland, Zymbal's gland or forestomach in rodents. The uncertainty associated with the relevance to humans could be considered comparable to that associated with confounding in an epidemiologic study. Confounding is often used to minimize the relevance of elevated risk estimates, when in fact confounding can attenuate or exaggerate a true association. The Agency should guard against inappropriate conservatism in the face of uncertainty when attempting to combine different types of data in reaching public health decisions. By considering all of the "relevant" data in a framework analysis, the EPA cannot raise the bar so high that nothing is recognized as a threat to public health.

While the Panel was given assurance that strong epidemiologic signals would not be ignored, having more data can lead to confusion as easily as to clarity, especially when the data are inherently variable in quality or in relationship to the endpoint of interest. Professional judgment of the strength of the data in the separate areas in the context of both hazard characterization and dose-response analysis will still be necessary before a decision can be reached on the collective cohesiveness or biologic plausibility.

The draft framework notes that consistency may not exist between human and animal model responses. In such situations, it is proposed that the most sensitive endpoint in animal models be used to ensure protection of humans. By way of example, consider that pesticide X inhibits an enzyme resulting in cardio excitation as the most sensitive endpoint in rodents. A similar effect occurs in humans, but at lower exposure concentrations, pesticide X causes headache, slight confusion and nausea. These endpoints are not detected in animal studies. In such situations, it is the relationship between enzyme inhibition and the most sensitive apical effect in humans, which is critical. If, for example, a 25% inhibition results in headaches, etc. in humans, then a 25% reduction in enzyme activity should be used as the appropriate endpoint in animal experiments.

Epidemiologic studies may also suggest <u>other MOAs and/or key events</u>. This needs to be taken into account in relative weighting of information.

The Agency also needs to recognize the difference between poor quality epidemiologic data and epidemiologic data in which results do not fall within the comfort zone whose boundaries were established by the Bradford Hill criteria. Both have limited use in the risk assessment process, but while the former can be ignored (or deferred for possible future use), the latter should raise the possibility that these boundaries need to be expanded, and there should be a strategy to pursue this possibility.

Aspects not addressed within the draft framework include the following: extrapolation among species (mentioned in paragraph 1 on p. 32), and extrapolation of high-dose toxicity in experimental animals and human incident cases to environmental exposure in humans that

typically occurs at much lower doses. Individual susceptibility, genetic variation and other potential effect modifiers are also not addressed.

Additional Specific Comments on the Draft Document

Page 8, Figure 1: It would be helpful to emphasize that the critical connector is mode of action. Biomonitoring data, biomarkers of exposure and effect need to be selected based on what we know from the animal studies. This enables meaningful integration of the toxicologic and epidemiologic data.

Page 9, Table 1: Provides a nice overview of relevant documentation from a range of agencies on which OPP has drawn to develop the framework

Page 10, last paragraph and Figure 2: The current description is not very informative. It would be helpful to describe the nature of the information from a framework analysis that is considered in problem formulation since this is likely to play a key role in the integration of epidemiologic and toxicologic data. This would be the appropriate step, for example, to bring together relevant MOA information as a basis to inform epidemiologic design.

Page 21, paragraph 1, lines 5-7: Some Panel members felt that the NRC (2007) vision relates to a much broader range of human data, than the type currently collected in epidemiologic studies. In fact, it seems that *in vitro* studies in human tissues or cell lines are more likely to be informative in a more predictive mode of action context. In the future, epidemiologic studies will be much less exploratory in nature but focused to address specific questions in identified subgroups defined through consideration of early biomarkers of effect.

Page 21, paragraph, 4, line 2: Animal toxicology studies can be designed to cover a broad range of exposure levels. Unfortunately, in the vast majority of cases, they are not.

Page 31, paragraph 1: The intended meaning of the second full sentence of this paragraph is not clear. "When animal and epidemiological data do not provide a consistent toxicological picture of a particular pesticide more weight would likely be given to those studies with robust study design and availability of replication or confirmatory data." Would more weight be assigned to the human epidemiologic studies?

Page 31, paragraph 1: This paragraph references multiple effects. An adverse effect is considered to be a function of one mode of action. There are key events leading to this MOA with several potential pathways for perturbation.

Page 31, paragraph 2: In the interest of better integrating epidemiologic and toxicologic data, what we should be seeking are biomarkers of exposure or early biomarkers of effect that can be measured in humans, and are based on an understanding of mode of action for critical effects. One Panel member did not fully understand (or agree with) the conclusion that by selecting a biologically plausible and sensitive endpoint from animal studies, the risk assessment is also protective of human health. Why can we assume that the dose-response curves are similar?

ATTACHMENT E, Pt. 2

Agency Charge

2. Case Study A: Retrospective and Ecologic Non-Cancer Epidemiology Studies

OPP has a dual purpose for developing the Case study A on recent ecologic and retrospective epidemiology studies reporting adverse birth outcomes associated with atrazine exposure. First, the case study illustrates key methodological issues that OPP must consider when integrating ecologic and retrospective epidemiology studies in risk assessment/characterization. Second, this case study reviews several recent studies that will be considered in the re-evaluation of atrazine. Building on the feedback from the SAP at the February, 2010 meeting, these studies will be incorporated in the overall WOE analysis and risk characterization for atrazine. The atrazine WOE is scheduled for review by the FIFRA SAP in September, 2010.

Question 2.1 As discussed in Question 1.1, the draft framework provides general descriptions of the strengths and limitations of ecologic and retrospective epidemiology studies with respect to human health risk assessment. Please describe what you consider to be characteristics of robust, well-designed ecologic and retrospective epidemiology studies.

Panel Response 2.1

General Comments

Clarity of terms must be provided in the Framework document. Ecologic studies (which are based on group level data) must be separated from retrospective studies (which are based on individual level data) because their potential use in risk assessment is very different. Greater clarification is needed, particularly for the term "retrospective epidemiology studies" because many epidemiologic investigators use this term to describe case-control studies in which information about exposure is assessed retrospectively. In Case Study A, the term "retrospective" is also applied to "retrospective cohort studies" (better labeled historical cohort studies) but with a different design. In historical cohort studies the exposed and unexposed members of cohort are ascertained retrospectively, and these persons are followed up to determine incidence rates of the health outcomes.

Case-control studies and retrospective cohort studies share some of the same challenges of accuracy and completeness of retrospectively ascertained exposure information, but they may determine exposure using different methods. In case-control studies, participants are usually asked about prior exposures, so the information gathered may suffer from inaccuracy and incomplete recall. In retrospective cohort studies, the exposed and unexposed cohorts are often identified from existing records on prior exposure; e.g., in an occupational setting. Thus, retrospective cohort studies have the potential to provide more accurate and complete assessment of exposure than participant recall of exposures in case-control studies. It should be noted, however, that even though the Agricultural Health Study is a prospective cohort study, much of its initial exposure assessment that determined exposure groups was based on retrospective recall of exposures, thus making it somewhat of a hybrid retrospective and prospective cohort study.

It should further be noted that nested case-control or case-cohort designs, which were not included in Case Study A are efficient designs that may provide useful information for risk assessment. For example, a nested case control study within the Agricultural Health Study could

provide less potential for bias in ascertainment of exposure than traditional case-control or retrospective cohort designs.

Finally, the use of the term "predictors" in this section is inappropriate in ecologic (and in some cross-sectional) studies because the exposure of interest is usually assessed at the same time as the health outcome so that the temporality of the relationship cannot be determined and thus it is unknown if the exposure is truly a risk factor.

Due to the inherent differences between ecologic studies and other study designs, retrospective studies of various types will be considered separately in response to questions 2.1 and 2.2.

The Panel recommends that this OPP report acknowledge that ecologic studies are inherently weak vehicles for quantitative estimation. Data from ecologic studies may have considerable strengths in other ways: generating hypotheses, supporting smaller and inconclusive data of stronger inherent character, providing "floors" to the size of some effects to support legislation or regulation. However, the strength of ecologic studies should not be overstated in the interpretation and analysis of problems. The quality of ecologic studies spans a spectrum of strengths; not all ecologic studies are equally informative.

Ecologic studies are considered to be of most use in hypothesis generation and are rarely suitable for hypothesis testing because both the exposure and the health outcome data for such studies are collected at the group, rather than the individual level. Thus, they are not useful in quantitative risk assessment. An inherent problem in ecologic studies is an inability to control for potential confounding at the level of the individual when exposure and outcome are assessed at the level of the group. Therefore, adjustment for confounding factors at the population level may not sufficiently remove confounding effects. This in turn, may create disparate findings from studies where adjustment for confounding is performed on an individual basis to obtain summary statistical results comparing groups. This shortcoming, often termed the "ecologic fallacy" renders ecologic studies less useful in the problem formulation stage of risk assessment as well as in risk characterization.

Additional considerations for evaluating the information in Table 1 of Case Study A on page 41 of the draft framework should include whether information on potential confounding variables was obtained and used for adjustment in the statistical analyses. This is particularly important when comparing rates between geographic areas or between a state and the United States as a whole because differences in the population distributions for variables including age, racial/ethnic, socioeconomic status, lifestyle factors (e.g., smoking, diet), and family history, as well as other pesticide, chemical and air pollution exposures, could affect the rates and thus influence the results.

In this respect it is important to note that the CDC natality database that was used for the ecologic studies by Mattix et al. (2007) and Winchester et al. (2009) include data on potential confounding factors, such as maternal demographic variables and behavioral risk factors such as tobacco and alcohol use. However, some of these data are missing for certain states where recording of this information is not required (e.g., tobacco in California, Pennsylvania and Washington) or are not comparable over the years or across the states. This can result in significant amounts of missing data on potential confounding factors so that significant residual confounding cannot be ruled out, and furthermore results may not be directly comparable across

states after adjustment for confounding because the adjustment is based on inclusion of different confounding factors.

However, not all ecologic studies have similar potential for bias, and therefore the strengths and weaknesses of such studies should be considered individually as to their potential to inform risk assessment. The group level data may be more or less refined. For example, a study of disease rates by contamination levels in water on a state basis might be ecologic, but more information is available in comparing counties, still more in towns, or water districts, and still more in comparing rates with persons classified by household, all without individual data, though the last might be better described as individual-based. Some investigators refer to studies in which outcome is assessed at the level of the individual and exposure (such as concentration of a pesticide in the municipal water supply) at the group level as "semi-ecologic".

In rare instances, such as examining census tract or county rates of disease prior to and following a well-defined event such as introduction or removal of a pesticide from use, or following widespread contamination of the environment following an industrial accident, ecologic studies may be informative enough to incorporate in hazard identification. Such relatively informative examples of ecologic studies compare rates of disease in populations before and after events (with consideration for latency) or across geographic strata of well-defined exposed and unexposed populations.

Specific Comments for 2.1

The Panel provides the following comments regarding characteristics of robust, well-designed epidemiology studies:

Exposure measurement error is the predominant weakness in environmental epidemiology in general and in studies of pesticide exposure specifically because it is rare that individual data from biomonitoring are available to estimate internal dose. Exposure measurement error in turn, creates exposure misclassification when individuals are assigned to exposure categories using cutpoints from continuous data. Therefore, investigators should make maximal efforts to assure that their exposure data are as accurate and precise as study conditions permit, and that validation of estimated exposures be performed if conditions permit. As mentioned in the draft framework, exposure assessment is an important consideration in all epidemiologic studies, irrespective of the design.

As summarized in Table 1 of Case Study A on page 41, exposure assessment generally involves surrogate measures of exposure (e.g., levels of atrazine in the surface or drinking water or proximity to fields in which atrazine was applied), rather than measures of body burden or concentrations in drinking water and amount of tap water consumed to estimate exposure levels (noted on page 45). Robust, well-designed ecologic, case-control, historical or prospective cohort studies should use the best possible measures of exposure to estimate dose. For drinking water exposures, tap water concentrations are preferred over ground or surface water measurements, and information about individual amounts of tap and bottled water consumption at home and at work improve the accuracy of the exposure estimate. For studies that assess exposure through drinking water, sufficient measurements in the distribution system must be available to characterize monthly levels of contaminants. Alternatively, monitoring data and modeling can be used to estimate levels for the critical periods of gestation for specific outcomes. Many reproductive epidemiologic studies estimate exposures for specific trimesters

of gestation; e.g., studies of birth defects and first trimester exposures. Studies involving public water systems are easier to conduct than studies involving private wells due to availability of monitoring data for public systems. If the study focuses on public systems, the study area selected should have information sufficient to define adequately the coverage area of each public system and the source(s) of water for each system. If a study examines exposure from pesticide drift, modeling should be used to estimate levels near the home, and if possible validated by collection of dust samples from within the home. If chronic diseases or cancers are the outcomes of interest, the assessment must be able to characterize exposures in the distant past, which can present formidable challenges.

A potential source of exposure misclassification in reproductive epidemiology is the use of residential address of the mother at the time of the birth to assign exposure categories. Several studies have shown that approximately 25 percent of pregnant women move during pregnancy (Canfield et al., 2006), creating the potential for exposure misclassification when the critical period of gestation occurred prior to the relocation. Similarly, studies of chronic diseases often use the address at the time of diagnosis. Use of these addresses can result in misclassification of the relevant area of residence (e.g., residence at conception for studies of reproductive effects or residence years before disease onset in studies of chronic disease) and thus in the assessment of exposure levels. This potential source of misclassification is recognized in the draft framework. Robust, well-designed case-control and cohort studies should acquire residential histories and use the most relevant residential location(s) for assessing exposure. However, it should be noted that even obtaining relevant residential location may misclassify individual exposures because individuals spend substantial portions of their lives at work or otherwise away from their residences, and the exposures in these locations are often not considered. More recently, investigators have been collecting work address/location information in occupational histories which may reduce measurement error.

Accuracy, precision and reliability of exposure data are important components of high quality exposure assessments. These considerations may include the precision of measurement of exposure or estimated dose, the extent to which exposure measures or categories are well-defined, the incorporation of data on external exposure (e.g., from measurement in the individual's micro-environment) or internal dose through biomonitoring, the use of analogous data as an exposure surrogate, and predicted exposure estimates from validated modeling. Individually collected exposure data, preferably absorbed dose data, would be of greatest value. Exposure indices that have poor predictive ability should be avoided. If an exposure index is used, it should be validated with data. The exposure measurement must provide adequate discriminating power to detect an exposure-related hazard (at a minimum provide reliable gradations of relative amounts of exposure). Exposure metrics can represent dose estimates (for example, average daily dose or peak dose), duration of exposure or a combination of these in a cumulative exposure metric (e.g., area under the curve statistic).

Misclassification of the outcome can also occur in epidemiologic studies. Therefore, case-control, historical and prospective cohort studies should derive data on reproductive or cancer outcomes from registries with mandatory reporting and active surveillance with explicit and consistently used criteria and definitions of outcomes. Similarly, if exposed and unexposed cohort members are followed by routine, regular screening for outcomes, explicit and consistently used criteria and definitions of outcomes should be used. If self-reports are to be used, efforts to confirm diagnoses should be made by review of medical records or data linkage with birth certificate, cancer or other registries. Robust, well-designed studies of all types should

derive reproductive and cancer outcome data from registries with mandatory reporting and active surveillance with explicit and consistently used criteria and definitions of outcomes. In case-control and historical cohort studies, ascertainment should be performed in a similar manner for the exposed and non-exposed group for the outcome as well as for potential confounders. Differences in ascertainment can introduce selection and information bias.

A well-defined study population with inclusion of an appropriate comparison group to address the study hypothesis and objectives is an important component of study quality. The use of population-based registries for cancers and birth defects, and confirmed diagnoses if information is obtained from self-reports is recommended to reduce selection and information bias, respectively.

In the studies summarized in Table 1 of Case Study A on page 41 of the draft framework, much of the data on occurrence of birth defects, preterm delivery and small for gestational age was derived from birth defects registries, birth records and national datasets. Important considerations in using such data sources (i.e., also for cancer registry data) include:

a. whether reporting to the registry or on the birth record is mandatory as this would tend to make these sources of information more complete, and reporting from areas where it is not mandatory could be influenced by factors that might also be related to exposure (e.g., socioeconomic status may be related both to likelihood of reporting and of exposure to pesticides);

b. whether the registry actively identifies birth defects or depends on passive reporting of defects: an active identification system would tend to provide more complete ascertainment of cases;

c. whether reporting to the database depends on who reports, e.g., health care providers or parents or both because the more individuals who are reporting, the greater the likelihood of more complete ascertainment;

d. whether the criteria for and definitions of outcomes (e.g., birth defects, preterm delivery and small for gestational age) have been explicit and consistently used so that data are comparable across years and across regions; and

e. whether the length of follow-up is appropriate. For birth defects, some outcomes are not captured in birth records because they become manifest some time after birth, during the first year of life.

Robust, well-designed studies should make a maximum effort to assure that potential confounding is controlled to the extent possible. Thus, investigators should obtain complete information on as many potentially confounding variables and risk factors as possible from all individuals to reduce the possibility of residual confounding by unmeasured variables. Potential confounders should be evaluated with appropriate criteria and methods to determine whether they are related to the exposures and outcomes and whether their inclusion in multi-variable models produces a change in the effect estimate. Those potential confounders that meet the criteria should be retained in the final models.

Robust, well-designed studies should have sufficient statistical power and information to enable analyses for effect modification. Effect modification is present when the magnitude of the risk estimate varies across strata of another variable such as age, gender, race/ethnicity or socioeconomic status. Incorporating analysis of effect modification is an important component of good epidemiologic studies and is relevant to risk assessment for identification of potentially susceptible subsets of the population such as children. Change in the risk estimate across racial/ethnic groups could be due to genetic differences in the frequency of a polymorphism in a gene that controls transformation or metabolism of a xenobiotic. Effect modification can be detected by stratifying the analysis across levels of the variable or by including interaction terms in the model to assess statistical interaction. Therefore, sufficient sample size and information should be provided to enable stratification on the potential effect modifier(s) with sufficient statistical power to detect meaningful differences.

Robust, well-designed studies should incorporate careful consideration of appropriate analytic methods. Some epidemiologic analyses are based on categorization of exposure; e.g., into tertiles or quartiles depending on the nature of the exposure metric. Linear models are also used when data are available for continuously distributed exposure variables. Smoothing methods may be used to inform the categorization of exposures (instead of categorizing by percentiles, or to check the appropriateness of percentile categorization). When meaningful cutpoints exist (e.g., the MCL) these may be used in the analysis. In addition, when data are available, sensitivity analyses should be conducted for potential exposure misclassification bias, information bias, selection bias (including loss to follow-up), healthy worker/survivor biases, and other confounders to estimate their potential impact(s) and interpretation of the findings. All findings should be reported. Interpretation of findings should not be based solely on statistical significance testing. The precision of risk estimates is based on the confidence interval around the risk estimate.

High quality studies should account for temporal, spatial and individual variability as related to exposure. They should also incorporate a sufficiently long observation period with respect to the expected latency of health effects. For example, exposures to pesticides in water should include a complete residential history and history of water consumption habits; studies of workers should include a full occupational history with address/location information.

Statistical power is an important consideration in evaluating associations. Studies should provide sufficient sample size to examine the relation of exposure to reproductive, cancer or other chronic disease outcomes. Statistical power is the ability of a study to detect an association between an exposure and a health effect, if in fact, one exists. Statistical power should be considered in evaluating null studies to reduce the probability of failing to detect an association if one exists (false negatives). Statistical analysis should include examination of the confidence intervals for a risk estimate as well as the determination of statistical significance. For studies with low statistical power, failure to find statistically significant differences should be interpreted cautiously and should consider the magnitude of the observed effect and variability in the effect estimate.

Data from Winchester et al. (2009) [Table A-2 on page 43 of Attachment A of the draft framework] illustrate another issue in interpretation of some ecologic and cohort studies relevant to sample size. Specifically, sample sizes can be quite large when comparing populations of entire regions e.g., county level data, resulting in very small risk estimates being statistically significant. These data raise the issue of statistical versus biological or clinical significance.

Failure to consider heterogeneity in the categorization of the outcome when developing the case definition may lead to improper conclusions. Analyses that consider all forms of an outcome together (all cancers, all birth defects) are not informative since different etiologic mechanisms are likely to be involved in specific forms of the disorder. Teratogens may be responsible for specific birth defects but have no association with other defects or other adverse reproductive outcomes.

A potential issue in consideration of epidemiologic studies is that of multiple comparisons, particularly when these are not based on well-founded *a priori* hypotheses. Epidemiologists recognize that multiple analyses for multiple outcomes (e.g., multiple categories of birth defects) or analyses that classify exposures in many different forms using varying temporal or spatial constructs or measures of exposure may generate statistically significant differences, some of which may be due to chance alone due to the multiple testing. Such exploratory studies may be useful, but caution must be exercised in interpreting results. In such circumstances, investigators should recognize the potential impact of multiple testing unless a well-developed hypothesis indicates the likelihood of the exposure being related to more than one outcome. Differences in opinion exist with regard to the appropriateness of formal statistical adjustment in these situations, and these are discussed in further detail in response to question 2.4.

Finally, for some exposures and outcomes, published pooled and meta-analyses may be informative. Pooled analyses are particularly useful when original data from multiple investigators are available since study power will be increased. Meta-analyses which incorporate sample sizes and risk estimates from a number of individual studies into a single risk estimate with its confidence interval are also useful to provide proper perspective in reaching conclusions, particularly in a weight-of-the-evidence approach for epidemiologic studies.

Many of the points made by the Panel are also summarized in a paper by Swaen (2006). These characteristics of robust case-control, historical and prospective cohort studies should not be reduced to a checklist because variation occurs from problem to problem and between studies, so that thoughtful interpretation by epidemiologists will remain necessary. These criteria can also be applied to ecological studies, although most ecological studies will fail to meet many of these criteria.

A parallel to this discussion is found in a paper by Shore et al. (1992) which states in part "Good-quality epidemiological studies are those with sound methodology, lack of bias, long enough follow-up times to observe a (carcinogenic) *health effect* response, adequate exposure information, and dose-response information. Before a lack of (carcinogenicity) *health effect* can be inferred, it is essential that the exposures be of substantial duration and intensity, and that the number of exposed persons be reasonably large."

Question 2.2 Ecologic and retrospective epidemiology studies are particularly useful in identifying new hypotheses about the human health effects of pesticide exposure and may confirm the human relevance of findings from experimental animal studies. However, these types of studies do not typically include robust characterization of exposure and they do not address confounding factors as well as prospective studies. Although there may be exceptions, generally, ecologic and retrospective epidemiology studies are not sufficiently robust for use in quantitative risk assessment (i.e., for use in deriving a point of departure or in quantitatively informing extrapolation factors, etc). In light of the strengths and limitations of ecologic and retrospective studies, please comment on appropriate ways to use these types of epidemiology studies in risk assessment/characterization or their utility in problem formulation (e.g. defining additional analyses or research/testing).

Panel Response 2.2

General Comments

As described above, exposure measurement error and misclassification of exposure comprise the most important limitations of epidemiologic studies for incorporation in risk assessment. Epidemiologic studies of pesticides face challenges in assessing exposure accurately and to a lesser extent in identifying and measuring potential confounders and effect modifiers. This is illustrated by the difficulties experienced in trying to obtain reliable biomonitoring data that matches the characteristics of the exposure and the outcomes of personal monitoring studies in studies of workers. Even with some degree of control over the measurements on identified individuals, large variability and associated large uncertainties still occur.

External exposure is a surrogate for internal exposure (exposure of a target organ). For a single well-defined externally applied dose, internal exposure can vary by an order of magnitude between individuals. Even direct measurements can be misleading. For example, the study of Barr et al. (2007) indicates that atrazine mercapturate measurements (the frequently used marker of atrazine exposure) underestimate exposure to atrazine and its break-down products. In both occupationally and non-occupationally exposed individuals, other metabolites (diaminochlorotriazine, desethylatrazine) predominate. Despite these limitations, rigorous estimates (preferably confirmed by measurements; e.g., multiple 24-hour urine samples analyzed in a study of absorbed dose to 2,4-D) (Hayes and Aylward, 2009; Aylward et al., 2010) can provide validation of the external exposure estimates. Similarly, a series of studies by Harris and colleagues show that epidemiologic studies can be conducted using absorbed dose estimates of 2,4-D and other herbicides as well as external exposure estimates and self-reported questionnaire information (e.g., pesticide use) (Harris et al., 2002, 2005; Harris 2007; Harris and Wells, 2007).

Worker exposures typically result in the highest expected levels of exposure, so epidemiologic studies of occupational cohorts can be useful in identifying associations with adverse effects (if these exist). However, the use of worker exposure data does not provide an assessment of exposure of other members of the population. Spouses and children can be exposed through contact with contaminated clothing, contaminated equipment or recently treated areas. This became obvious with the families of workers exposed to asbestos. Although this exposure

should be minimal if good practice is followed, contamination can occur. Other potential sources of exposure (spray drift, water, food) need to be considered.

As with experimental toxicology, epidemiologic studies tend to concentrate on one compound in isolation. It is difficult to identify all of the important compounds in the "chemical soup" that might interact with the action at the target or that might produce similar outcomes. Humans are rarely exposed to individual chemicals, particularly in the context of agricultural chemicals. The assessment of hazard associated with chemical mixtures poses a challenge to toxicologists due to complexities associated with the selection of chemicals in the mixture and exposure concentrations of the individual chemicals. These problems are not confined to pesticides since humans are exposed to many other compounds (e.g., industrial chemicals, pharmaceuticals, components of household products, personal care products {some with seasonal use e.g. sun screen products}) on a regular basis. Well-designed retrospective epidemiologic studies may provide a foundation for assessing hazards associated with exposure to chemical mixtures. Occupationally exposed cohorts represent one opportunity for such studies. Epidemiologic studies may provide insight into exposures to mixtures of relevance to humans. Although challenging, associations with components of the mixture or with the entire mixture as measured could be evaluated using the WOE framework and, when appropriate, used to provide guidance in the design of animal toxicity studies.

Identifying potential confounding factors is an important part of the design of epidemiologic studies. It is not possible to know whether all relevant factors have been identified, and it is important to assess whether known potential confounders have been considered. Given these reservations, well-designed epidemiologic studies have the potential to contribute to the risk assessment/characterization process in a number of areas. An important area is the identification of potential health problems (previously not considered) that may be associated with exposure to pesticides. This can identify compounds that should be regarded as of concern, and provide guidance in the prioritization of research. They can provide some guidance to environmental levels of exposure that may impact adversely on health, and could inform research to determine internal exposures corresponding to observed external exposures. Well-designed epidemiologic studies may help in the identification of sets of lesions that could be investigated using toxicodynamic and molecular methods to determine the mode of action of the target compound.

A formal framework for validating epidemiologic methods, particularly for exposure, is needed. The development of such a framework has transformed the field of analytical chemistry over the last twenty years. Validation protocols have led to greatly increased reliability of analytical chemical data. Epidemiologic and toxicological studies could provide better information (for instance, improved definition of the uncertainties associated with the estimation of exposure, and identification of target sites relevant to human exposure) if investigators from varying disciplines collaborated with each other. For example, banking of bio-specimens and environmental samples could be used to increase the utility and reliability of future cohort studies and permit validation of exposure estimates.

It would be beneficial to have a framework for assessing the scientific validity of the outcomes being investigated. This would ensure that studies using categories containing multiple endpoints resulting possibly from different modes of action are identified, and evaluated

appropriately. Although the statistical methods used by the various authors were described in the case study, their appropriateness was not evaluated. A framework in which to consider the strengths and weaknesses of any analysis would be of benefit, and would focus attention on possible alternative ways of analyzing the data.

Specific Comments for Types of Epidemiologic Studies

Ecologic Studies

As described in the response to question 2.1, ecologic studies are considered to be of most use in hypothesis generation and are rarely suitable for hypothesis testing since data are collected at the group, rather than the individual level. Thus, they are not typically useful in quantitative risk assessment unless the target of interest is the larger group as is sometimes the case in descriptive epidemiology. The use of surrogate measures of exposure may create sizeable misclassification of exposure. Thus they are largely useful (arguably more so than incident data), for suggesting hypotheses to be addressed in future well-designed studies of individuals and in examining consistency of findings.

Due to the problems inherent in the "ecologic fallacy" and the inability to control for potential confounding at the level of the individual, ecologic studies are also less likely to be useful in the problem formulation stage of risk assessment. In rare instances, such as when examining census tract or county rates of disease prior to and following a well-defined event, such as introduction or removal of a pesticide from use, or following widespread contamination of the environment following an industrial accident, ecologic studies may be informative enough to incorporate in problem formulation. An example of the latter occurred at Seveso, Italy in 1976 when a substantial proportion of the residential population was exposed to dioxin - 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD). The event was followed by short-term studies of morbidity and by a long-term prospective cohort study to evaluate mortality and cancer incidence based on zones of exposure to the contaminant (Bertazzi et al., 2001). In this example, exposure was assigned based on soil measurements of TCDD and a limited number of human samples (Bertazzi et al, 1998). Such relatively informative examples of ecologic studies compare rates of disease in populations before and after events (with consideration for latency) or across geographic strata of exposed and unexposed populations.

Ecologic studies are often exploratory. However, observations made in these studies can be used to direct future hypothesis setting and analysis. They can also provide insights into future needed analyses and research, i.e., identifying gaps in knowledge and informing problem formulation and driving the research agenda. Exposure characterization is an inherent weakness of ecologic studies since it is not focused on the individual. Accordingly, these studies are of limited value in this aspect of the risk assessment process. This makes it difficult to use such studies as the basis of hazard characterization. However, the results can be corroborative of the hazard characterization obtained from toxicological studies using animal surrogates, and can identify hazards that are unique to humans, and not seen in animal models.

One important potential use of ecologic epidemiologic studies is in the monitoring of the effects of mitigation measures implemented as a result of the risk assessment process. Data at the

population level may be informative in a temporal analysis in which disease rates are compared for the same population before and after implementation in a regulation or changes in a standard (i.e., as a check on the correct identification of mitigation requirements and the effectiveness of risk management measures aimed to protect human health). Further, on the basis of published studies it may be possible to identify opportunities for validation of findings where a particular pesticide is removed from use, or restricted in use in a region (e.g., atrazine in many European countries) or where exposure is changed (e.g., due to improved drinking water treatment). However, the latter would simultaneously reduce levels of other contaminants found at low levels in treated drinking water. Additional analyses and research can be appropriately proposed based on preliminary findings from ecologic or historical cohort studies.

Retrospective Epidemiologic Study Designs

Several study designs comprise the group designated as "retrospective", including case-control studies, historical cohort studies, nested case-control studies within cohort studies and cross-sectional studies. These vary in their design characteristics and in their strengths and weaknesses. In turn, their usefulness in informing risk assessment will vary depending on the design, the quality of the exposure assessment, study power, the ability to adjust for potential confounders and other considerations described in response to question 2.1.

Case-Control Studies

The case-control design is frequently used in epidemiologic research because it is suitable for the study of rare outcomes and is relatively efficient. Well-designed and executed case-control samples can provide valid results comparable with what can be gleaned from a cohort study but at considerably reduced time and cost (Rothman, 2002). Various forms of bias can be introduced into case-control studies unless rigorous attention to appropriate design is provided. Information bias due to differential recall or reporting and selection bias due to differential participation or inappropriate selection of control groups are potential problems. However, these potential flaws are not necessarily present in and are not restricted to case-control studies, and reviews of their usefulness should examine the potential for introduction of bias and if present, the likely extent and direction of such bias. The retrospective nature of the case-control study is due to the sequence of events in carrying out the study. Typically, cases and controls are identified from suitable populations and information about exposure is collected subsequently. For studies of reproductive outcome, the length of time between identification of cases and controls and ascertainment of exposure may be relatively short, thereby reducing the extent of misclassification of exposure due to faulty recall of distant events.

A relevant example of a case-control study is one in which men with reduced semen quality (cases) were compared with a group of men with normal semen quality (controls) for biomarkers of pesticide exposure (Swann et al., 2003). The hypothesis was developed when the investigators initially found that men who lived in an agricultural area of Missouri had reduced sperm concentration and motility compared with men who lived in an urban area. They then conducted a case-control study by enrolling men with abnormal semen parameters (low concentration, lower percentage motile sperm and higher percent of abnormal sperm morphology) along with men with semen parameters within normal limits from the same areas in

the Midwest. They then analyzed urine samples from all men provided at the time of semen collection for concentrations of metabolites of eight pesticides. They found that pesticide metabolite levels in cases living in one of the states (central Missouri) were higher than those in controls for alachlor, atrazine and the insecticide diazinon (Swann et al., 2003). This study has some characteristics of a cross-sectional study because information about the disease status and exposure was collected simultaneously. Nonetheless, it illustrates the potential for obtaining quantitative data about pesticide exposure from biomonitoring using a case-control approach that may be incorporated into quantitative risk assessment.

Historical Cohort Studies

In the historical (or retrospective) cohort study, the cohort is identified from historical exposure records or data, such as biologic samples that may be available from samples collected in the past. Follow-up is conducted for cohort members, and disease status is ascertained at the time of the study, permitting comparison of rates among persons with varying levels of historical exposure. Thus, this design is more efficient and less costly than the prospective form of the cohort study, provides a better temporal sequence of exposure to outcome than ecologic or cross-sectional studies (making it more useful in risk assessment), and is often used to assess the relationship between exposures and health outcomes among occupationally exposed persons. When historical data are available for exposures to pesticides that occurred in the past, this form of retrospective study may be useful for risk assessment. Two examples of historical cohort studies are included in Case Study A (Villaneuva et al., 2005; Ochoa-Acuña et al., 2009) and are discussed further in response to question 2.3. These retrospective cohort studies are based on measured concentrations of atrazine in municipal water treatment plants and ascertainment of outcome for women who lived in the communities supplied by their respective water utility. The quality of the historical data and the degree to which exposure can be assigned to each individual will determine the extent to which the data will be valid for quantitative risk assessment.

Nested Case-Control Studies

In some instances, case-control studies may be incorporated within cohort studies and are referred to as "nested case-control studies". This design is useful when the costs of analysis for pesticide exposure are too high to study the entire cohort. A control group is selected from among all eligible controls and compared to cases that developed during the study period. For example, Krieger et al. (1994) conducted a nested case-control study of 150 women with breast cancer and 150 disease-free women, selected from a cohort of 57,040 women who had been enrolled during a multiphasic health examination in the late 1960s and had a serum sample collected and frozen at the time of examination. The women were followed up through 1990 for the development of breast cancer. Each case was matched by race to a cancer-free control and the concentrations of DDE [1,1-dichloro-2,2-bis(p-chlorophenyl)ethylene], the main metabolite of the pesticide DDT [2,2-bis(p-chlorophenyl)-1,1,1-trichloroethane]), and polychlorinated biphenyls (PCBs) were compared among the groups of white, African-American and Asian women. Matched analyses found no significant differences in the concentrations of these organochlorine chemicals between cases and controls in any of the three exposure measures. In this nested case-control study, the quantitative information about (absence of) risk associated

with development of breast cancer at specific serum concentrations of DDE and PCBs could be incorporated into a quantitative risk assessment for these chemicals.

Cross-Sectional Studies

Cross-sectional studies often assess exposure and outcome at the same point in time. Thus, their major limitation is an inability to establish the temporal sequence of events unless additional information is available. However, many cross-sectional studies obtain historical exposure information adequate to determine temporality. An additional limitation is vulnerability to healthy worker effect/survivor biases because the exposure may affect disease incidence, disease duration, or both. Their value is that they can evaluate early indicators of a health effect (e.g., effect biomarkers), and if done serially, can provide longitudinal data on changes in effect biomarkers over time. Cross-sectional studies may be informative in evaluating pesticide exposures and potential health effects. For example, Farr et al. (2004) conducted a cross-sectional study to assess the association between pesticide use and menstrual function among 3,103 women living on farms in Iowa and North Carolina who participated in the Agricultural Health Study. Premenopausal women completed two self-administered questionnaires on pesticide use and reproductive health at enrollment. They reported exposures about lifetime use of pesticides and hormonally active pesticides. The characteristics of their menstrual cycles, including cycle length, missed periods, and intermenstrual bleeding were ascertained at the same time. After controlling for age, body mass index, current smoking status and occupational physical activity, the authors reported associations between pesticide use and longer menstrual cycles and increased odds of missed periods compared with women who never used pesticides. Women who used probable hormonally active pesticides had a 60–100% increased odds of experiencing long cycles, missed periods, and intermenstrual bleeding compared with women who had never used pesticides (Farr et al., 2004).

In summary, case-control, historical cohort and cross-sectional studies have significant advantages over ecologic studies in evaluation of individual level data on exposures, health outcomes and confounding variables and are thus much more useful in risk assessment. The extent to which data from these epidemiologic designs can be applied in a quantitative context will depend on the methods used for exposure assessment and the ability of the investigators to make relatively accurate and precise measurements of dose as well as the outcome. Well-designed case-control and historical cohort studies may have quantitative value in the risk assessment process, depending on their ability to establish dose-response relationships.

Question 2.3 The atrazine case study (Case study A) provides specific examples of ecologic and retrospective epidemiology studies. Please comment on OPP's reviews of the studies discussed in Case study A. In your comments, please provide specific feedback on the OPP's descriptions of each study design, exposure assessment, use of appropriate statistical methods, and ability to address bias and confounding in addition to other factors that may be important in the interpretation of these studies.

Panel Response 2-3

General Comments

In general, OPP has performed an accurate and thorough analysis of the five published studies included in Case Study A, and has captured most of the limitations of these studies.
A weakness of Case Study A is related to limiting the criteria for inclusion to studies that examined a reproductive outcome and were published since the 2003 IRED decision. This limitation had several undesirable effects. First, all studies in Case Study A used an ecologic or retrospective cohort design. Thus the Panel did not have the opportunity to explore how other epidemiologic designs that used cross-sectional, case-control, or other approaches might be incorporated in the risk assessment process. Second, the overall quality of these studies was relatively poor, thus limiting their applicability to the upcoming review of atrazine or the more general issue of incorporating epidemiology in risk assessment. Third, two of the five published studies used an ecologic design (Mattix et al., 2007; Winchester et al., 2009). As pointed out on page 61 of the draft framework and elsewhere in this report, ecologic studies are not useful for hypothesis testing and can rarely be used to establish exposure-disease relationships. At best, these studies might contribute to hazard identification, but better studies of atrazine and reproductive outcomes are available to meet the goal of assessing exposure-disease relationships.

Overall, the approach of the Agency to evaluating the epidemiologic studies provides a useful framework, and covers the important factors that need to be considered. However, some additional general factors should be addressed in the evaluation process. First, the background material needed for a full evaluation of these studies was lacking. For instance, background information on the target health effects would have been useful. It would be useful to compare the methods used to handle analyses below the limit of detection in the various papers, because this can affect summary statistics, associated confidence intervals and any statistical testing and modeling. It would also be useful to assess whether the endpoints are specific and whether their definitions are precise enough to distinguish a number of specific endpoints resulting from different modes of action. It would be useful to see whether any feasible modes of action have been identified as underlying the observed lesions. An analysis of whether the observed endpoints were compatible with the known reproductive effects of atrazine observed in animal studies was not included. Some background material on temporal and spatial aspects of the reproductive health effects would be helpful in an assessment of studies such as those evaluated in Case Study A. Background information on the reproductive outcomes evaluated; i.e., low birth weight, SGA, preterm birth and birth defects in the general population would have been useful in evaluating the case study. This information could include total incidence rates, incidence rates by maternal age and race, recognized risk factors for each outcome (tobacco and alcohol use, body mass index, nutritional status, prescription and over the counter medications,

recreational drug use etc), accuracy and completeness of reporting from birth certificates, examples of well-recognized associations with environmental exposures from epidemiologic studies, data from experimental studies in laboratory animals and other considerations.

An evaluation of background material on the seasonality in live births would be helpful because such seasonality is strongly influenced by socio-demographic factors (Bobak and Gjonca, 2001; Darrow et al., 2009). It would be worthwhile to compare the observed periodicity in exposed populations with that in other areas with limited exposure to toxicants of interest. The Agency should consider how this sort of background information could be used in assessing the quality and utility of epidemiological studies.

The Agency and the authors of the five studies in Table A-1 of the draft document missed potentially useful analyses related to the papers in this group. Some review papers going back to the mid 1960's considered the medical significance of date of birth (Bailar and Gurian, 1964; Kesselman and Bailar, 1964; Bailar and Gurian, 1965). Three forms of congenital malformations (congenital hip dislocation, spina bifida and other neural tube defects, and patent ductus arteriosus) showed a seasonal pattern. These were also unusual in being the only three that occurred more frequently in females. This work may be important because atrazine was not in use at that time in the sixties, and evidence for or against a seasonal pattern at that time might tend to help in the interpretation of more recent data.

The five studies presented in this draft report share several problems in common. All are reporting effects that are small in relation to the background "noise", so that unrecognized or uncontrolled forms of bias may be quite important. Each of these studies had numerous sources of potential bias. Some of them also have a problem of multiple comparisons, though it appears that none of the studies addressed the issue. Multiple comparisons may create difficulty in interpreting findings, because some associations may be due to chance. The interpretations, particularly of the individual-level studies (Ochoa-Acuña et al., 2009; Ochoa and Carbajo, 2009; Villanueva et al., 2005) had flaws, according to one reviewer. The sixth study (Mohanty and Zhang, 2009) is an ecologic study based on a slide presentation. This study has not been published in the peer-reviewed literature. The Panel recommends that abstracts from scientific meetings, presentations and other data sources that have not been subject to the peer review process should not be incorporated into risk assessments by the Agency.

The retrospective cohort studies by Villaneuva et al. (2005) and Ochoa-Acuña et al. (2009) used municipal drinking water concentrations of atrazine from monitoring data to assign exposure levels for the analysis. This method of exposure assessment may lack precision at the level of the individual for several reasons described below. However, retrospective cohort studies that measure concentrations of a chemical in the municipal water supply for exposure assessment have been used frequently in epidemiologic studies of drinking water contaminants and may be used in risk assessment when study quality is high. For example, several retrospective cohort studies were conducted to examine associations between exposure to the disinfection by-products (DBPs) that are produced during chlorination and reproductive outcomes (Dodds et al., 1999). Their usefulness was limited by the quality of the exposure assessment and specifically for reproductive outcomes, the ability of the investigators to estimate exposure accurately during the critical period of gestation for each member of the cohort. In the research on DBPs,

mandatory quarterly testing requirements at the plant and in the distribution system of municipal systems serving 10,000 persons or more in the United States and Canada provided a data base for estimation of exposure. Assigning concentrations to specific trimesters of gestation period was performed by regression methods in one study (Dodds et al., 1999). With less frequent sampling, the quality of the exposure assessment will be negatively affected if exposure varies over time.

The degree to which daily, weekly or seasonal fluctuations in concentration occur, potential variability in concentration throughout the distribution system and the stability of the pesticide in water must be considered to reduce exposure misclassification (Nieuwenhuijsen et al., 2000). Important factors in estimating exposures to cohort members include the potential use of bottled water for consumption, the amount of tap water consumed daily, the use of tap or home filtration systems, whether the system is flushed prior to obtaining drinking water and other behaviors such as consumption of water outside the home. The use of tap water for making soup, cold drinks, coffee and tea and juices needs to be considered in this pathway. Exposure to volatile chemicals such as the trihalomethanes will be reduced during heating of water for preparation of soups and heated beverages. Conversely, exposures to volatile organic chemicals, such as the trihalomethanes, through showering and bathing comprise an important exposure pathway (Gordon et al., 2006). Residential mobility during pregnancy has been shown to be relatively common, affecting up to 25 percent of women in some studies. Therefore, reliance on the address found on the birth certificate may not provide an accurate assessment of exposure if the participant moved across boundaries during her pregnancy to locales where the concentration of the contaminant would be expected to vary widely.

One Panel member was concerned that all non-statistically significant findings, including findings that showed a monotonic exposure-response relationship, were ignored in the Agency review. Insufficient attention was paid to exposure-response relationships, how the studies evaluated them, and whether the evaluations were adequate (e.g., were the categorizations of exposure simply based on percentiles, were meaningful categories used [e.g., an MCL], was the use of a continuous variable in a logistic regression or binomial regression model warranted (i.e., to what extent did the model selected adequately characterize the likely shape of the dose-response curve?). Another Panel member felt that an over-emphasis was placed on confounding bias in the draft framework and that confounding rarely has a big impact in epidemiologic studies. This Panelist also felt that an under-emphasis was evident on the bias most likely to have a big effect on the estimation of the magnitude of effect and the exposure-response relationship, i.e., exposure misclassification. Further, inadequate attention was paid to the statistical power of the studies and to the impact of disease misclassification due to factors such as under-ascertainment of birth defects from information on birth certificates, and the grouping of heterogeneous birth defects that may have differing etiologies.

In the evaluations of the papers it might be beneficial to place some emphasis on the structure of the hypothesis, and on the validity of the exposure measurement. These issues were raised in comments by a number of Panel members. In evaluating the atrazine epidemiologic studies, the Agency needs to be consistent in its assessment of the possibility of concomitant exposure to other triazines and their breakdown products which have the same mode of action according to U.S. EPA's cumulative triazine risk assessment. (U.S. EPA, 2006)

One Panel Member commented that overall, the evidence presented in the case study on recent epidemiologic findings on the association between atrazine and birth outcomes was weak and inconsistent and seemed quite compatible with no effect of atrazine on birth outcomes. This individual felt that the collection of data could serve as a good example of a "negative" dataset for use in developing guidelines for the interpretation of human epidemiologic and incident data in health risk assessment. Others felt that the quality of the studies included in the Case Study was not adequate to reach any conclusion regarding potential associations between atrazine exposure and adverse reproductive outcomes. A more complete record of published papers, and or a more complete search of the literature, may or may not reverse this conclusion.

Conversely, another Panel member offered the opinion that the ecologic studies cited by the Agency may not suffer from the severe ecologic fallacy that the Agency identified. Information presented during the meeting, including analyses presented by public commenters, weight the evidence toward the environmental contribution to birth defect incidence. It was clear to this reviewer from the presentations by the Agency and the public comment (Syngenta Panel slide #44) that birth defect incidence in the US is strongly affected by seasonality both in states with and without heavy atrazine usage. This seasonal pattern coincides with the use of atrazine in those states during late spring/early summer, gradually diminishing in the fall and winter seasons that could be explained by the half-life of atrazine in the environment. This reviewer commented that the national birth defect incidence data compiled by CDC sheds light on the association of birth defect and atrazine concentrations in water (both surface and finished drinking water) in the states where atrazine has been heavily used.

Ultimately, the Agency needs to pose the question: to what degree does the epidemiologic studies decrease uncertainty associated with extrapolation from animal studies for the protection of human health? The Agency should use their answer to the question to approach uncertainty factors and the inherent limitations of observational research accordingly.

Specific Comments Regarding Case Study A

Mattix et al., 2007

The data in the paper by Mattix, Winchester, Scherer (2007) contain a gap from 1990 to 1995-2002. Two sources of data were available; no comparison was made between the CDC data for Indiana and that reported by the state. If serious discrepancies are apparent between the two sources, then one or the other (or both) must be wrong, and it would be important to find out which, and why. In the Agency's assessment of bias, confounding and other factors, the figures presented in the bottom, left-hand column on p. 948 suggest that, of the abdominal wall defects (AWD) occurring in the Riley Hospital (279 over 1990-2002) fewer than half (133, 48%) were simultaneously identified by the State Registry. This low rate of capture of AWD incidence by the State Registry raises a question about state-to-state differences in the ascertainment of AWD incidence and whether the higher rate noted in Indiana might not be due to a higher than national average rate of capture of AWD incidence. The state registry information is based on birth certificates which often fail to capture many birth defects; AWD is one that is often omitted. The critical issue with using data reported on birth certificates is not whether they contain errors, but

whether the errors are differential between states. If reporting across states is relatively constant over a specified period of time or location, then analysis of those times or locations may provide relatively useful data.

One reviewer commented that the strength of findings presented in this paper should be reduced to account for the "multiple comparisons problem". The authors noted that the elevated Indiana rate was statistically significant only in 1996, 1998, and 2001, but a critical question is whether the reported rates for all years are statistically comparable. In short, was statistical power great enough to say that an effect was present (or greater) in some years than in others, or are we just looking at the effects of having small numbers of AWDs in each year? Were any features of atrazine use unique during the higher-incidence years? It appears that the data in Figure A-2 were not adjusted for nitrates.

Winchester et al., 2009

One reviewer commented that the relationship shown in figure A-1 is far from striking, especially when one views this in the context of the overall scale of cases (per 100,000), a maximum variation of perhaps 6% is observed. This is even more concerning due to numerous possible season-related confounders. The peak incidence in terms of last menstrual period (LMP), roughly, date of conception, was May-June. The data would be more convincing if the authors had found a lack of such a pattern in mothers who had been drinking ground water. Also, the text provided no evidence that the authors adjusted for other seasonally changing chemical exposures, nor did they look at concurrent data from other states with lower atrazine exposures to see whether the reported patterns were unique to atrazine exposure. It was appropriately noted in the Agency's critique that chemical concentrations were measured in surface water and not drinking water, and that these were population-level data based on rates.

According to one reviewer, as shown in Tables A-2 and A-3, all but one of the birth defect types occurred more frequently in April-July than in other months, and the exception ("Nervous", not further specified) barely fell below a ratio of unity. About half of the differences were statistically significant. However, chemical teratogens tend to be more specific with effects targeted to the organ that was in a critical stage of development at the time of exposure. The lack of specificity and broad pattern of the evidence suggests a pervasive bias related to some other seasonally changing factor.

Ochoa-Acuña and Carbajo, 2009

The retrospective cohort study of limb defects by Ochoa-Acuña and Carbajo (2009) identified birth defects among 48,216 singleton births between 2000 and 2004 in rural Indiana. Although not stated explicitly, it appears that the investigators compared cases of birth defects with unaffected infants to calculate odds ratios for exposure to cornfields or soybean fields within 500m of the residence. Unfortunately, the exposure analysis relied on proximity to these fields and as an exposure surrogate and was not validated by other methods.

For some birth defects, e.g., neural tube defects (NTD) and abdominal cavity, the statistical power to detect meaningful differences was low. The category "heart defect" combines defects

with very heterogeneous etiologies and should be analyzed, if sufficient numbers of cases exist, by subgrouping (e.g., conotruncal heart defects). The study controlled for the variable "farm exposure", i.e., the percent of cropland around a home, which likely led to a bias toward the null. This factor is part of the exposure of interest and not a confounding variable. If distinguishing farming exposure effects from "drift" to bystander populations is a concern, then the analysis could be stratified by this variable. The authors noted that when the models did not include this variable, respiratory defects "appeared increased".

In the evaluation of this study, the statistical methods were described but not evaluated. Limb defects showed an odds ratio with a 95% CI above 1.0 for <3.4 ha vs. >3.4 ha. Other defects also had high odds ratios, but the lower limit of 95% CI fell below 1.0. Elevated odds ratios were observed for several birth defects and soybeans, which were not discussed because they were not statistically significant. The strongest relationship in Table 3 was the finding for NTD (odds per unit increase in exposure = 1.72) and soy area, but was not discussed. It would have been useful to have comments on whether additional or different statistical methods could have been used to analyze the data. For instance, it may have been better to divide the crop areas into three or four categories rather than using an exposed group and not-exposed group comparison. The methods used may have been too conservative. Multiple comparisons were performed; this is of concern because the effect estimate is modest and the confidence bounds on the adjusted ratio for fields of corn (OR = 1.22, 95% CI: 1.01, 1.47) barely exclude unity. The odds ratio for soybean fields does not suggest an effect (OR = 1.04, 95% CI: 0.85, 1.28). The use of a continuous variable in an exponential model (Table 3) is problematic because it assumes an exponential increase in risk for each increment of exposure (Hosmer and Lemeshow, 2000).

Villaneuva et al., 2005

This is a retrospective cohort study with exposure assigned to individuals and covariates measured at the individual level (not an ecologic study as designated by the Agency). As shown in Table A-6 of Attachment A, a non-significant association was present for preterm delivery and exposure to atrazine in finished water which was not discussed by the authors or the Agency. An exposure-response relationship was observed (ORs 1.0, 1.22, 1.93 across the tertiles), and the finding in the higher exposure group (i.e., OR = 1.93) was stronger than any other finding in the study. A monotonic exposure-response relationship was apparent; therefore, this finding should have been thoroughly examined. In table A-7 the ORs for first trimester exposure and preterm delivery (1.36, 95% CI 0.95, 1.95) and third trimester exposure and SGA (1.37, 95% CI 1.04-1.61) are nearly identical. The finding for SGA is statistically significant due to larger numbers of cases. This example demonstrates the limitations in relying solely on p values in interpreting epidemiologic data.

The Agency's assessment of the exposure data included the issues of using drinking water concentrations averaged across several years, while the birth effects were for a single year, the relatively low concentrations of atrazine, and the fact that the concentrations of atrazine in the three exposure groups were not adequately heterogeneous to discriminate potential associations. The latter point was also discussed by the authors. Breakdown products of atrazine were measured in the study, which constitutes a strength in the exposure assessment for that

investigation. Exposure was transformed into geometric mean values without any explanation. If it was because of skewness in the distribution, then it was inappropriate to use this parameter because it is the high-exposure points that are of concern, and it is counter-productive to reduce their impact on the analysis. Only one year was examined, and so any possible year-to-year patterns cannot be studied. No analyses were included of possible covariates correlated with distribution units, such as ground vs. surface water, or local contamination by known sources of toxic chemicals. The authors were thorough in their identification of potential bias and confounding factors, which included selection and recall bias, maternal smoking and alcohol consumption, exposure to agents with known effects on abnormal birth parameters (disinfection byproducts, air pollution, PCB's and lead) that may have seasonal patterns that coincided with that of atrazine, and atrazine exposure other than by drinking water.

Table A-9 contains a statement that the study by Villaneuva et al. (2005) is limited because it was conducted in France and the results may not be generalizable to the U.S. The rationale for OPP's statement is unclear.

Ochoa-Acuña et al., 2009

In this study of atrazine in drinking water systems in Indiana, study data were adjusted for season. This adjustment may have led to a bias toward the null if the seasonal effect on SGA or preterm birth was partly due to atrazine exposure. The study used a continuous variable in an exponential model, which makes the strong assumption that the risk increases exponentially with each increment (log atrazine level) of exposure. Moreover, use of a log transformed exposure variable may not be appropriate for characterizing the exposure-response curve. The categorization uses percentiles which may or may not be appropriate to characterize the exposure curve. The SGA effects were statistically significant although small. They fall in the range of 1.06-1.2.

In the study by Ochoa-Acuña et al. (2009), the availability of atrazine sampling data at 7 to 14 day intervals constitutes a strength of the exposure assessment. Some issues involved with interpolation of atrazine concentrations and the quality and comparability of the four drinking water monitoring programs were addressed. However, a weakness in the exposure assessment was that estimates were based on sparse data, especially for the winter months. The latter is critical since it is this period that comprises the unexposed months of gestation.

Roughly 70% of the birth records available to Ochoa-Acuña et al. (2009) came from one mid-sized community, which raises questions about selective effects on reporting. It is not clear why Fort Wayne predominated in the data, or whether unmeasured confounders occurred more frequently in Fort Wayne.

Weak evidence of an association between atrazine exposure and SGA was detected for exposures in the third trimester and "entire pregnancy". No association with preterm delivery was evident. LBW was not reported in this part of the analysis. The range of the confidence bounds was smaller for SGA than for preterm delivery with larger sample sizes available for the third trimester and entire pregnancy than for the first or last month of gestation in the preterm delivery analyses.

One Panelist introduced several additional factors to be considered in the WOE analysis, particularly with respect to the ecologic studies in Case Study A:

a. Window of susceptibility – In the atrazine case study investigators looked at either the association of atrazine concentrations in surface water and the months of LMP in relation to the national birth defect data (Winchester et al., 2009), or the association of atrazine concentrations in surface water during the 3rd trimester and SGA (Ochoa-Acuña et al, 2009). This Panelist felt that the data presented provide a convincing link between birth defects and atrazine concentrations in surface or drinking water at the critical points of time during development. If the window of susceptibility for a birth defect is not taken into account (as presented in a public comment) the correlation between month of LMP and atrazine concentrations in water disappeared.

b. Longitudinal or temporal variations of atrazine exposure and the correspondence with birth defect outcomes – In the case of atrazine exposure and birth defects, temporal variation of atrazine concentrations in either surface water or drinking water is critical for assessing birth defect risks. If atrazine concentrations in water were to remain constant throughout the year while birth defect incidence rates varied, the WOE of this association would be non-existent. The converse is also true.

c. Evidence for protecting public health. – One of the missions of the Agency is to safeguard the public from unnecessary pesticide exposures. In the opinion of this Panelist the evidence for an association between atrazine and birth defects is adequate to consider it as "Some Evidence For", and thus it would be prudent not to dismiss it. As this framework is developed and evolves, and better quality epidemiologic and/or incident data become available, the Agency will be able to change the designation toward or away from "Some Evidence For" with greater confidence without endangering public health.

Question 2.4 In light of scientific issues discussed in Questions 2.1-2.3, OPP requests input from the SAP on factors to consider when integrating these studies in the atrazine WOE analysis currently under development.

Panel Response 2.4

Of primary concern to the Agency in integrating the results of epidemiologic studies in the atrazine WOE analysis is an assessment of study quality. The criteria for assessing the quality of such studies are discussed in detail in response to question 2.1. Study quality must be a key component in selection of studies to incorporate into the analysis. An important issue is how the Agency decides whether to use particular sets of data. It is not uncommon for the Agency to be criticized by some experts for *excluding relevant* data from their risk assessments and criticized by others for *including poor quality data* in the same risk assessment. The Agency should establish a set of criteria for determining the acceptability of epidemiologic studies. These criteria may be based on quantitative criteria, scientific judgment, or some combination of these. Inevitably, it will be necessary to exercise some degree of scientific judgment in this assessment. The Panel recommends that epidemiologists participate actively in the process. Observational research is subject to potential error due to the nature of the science. However, the presence of uncertainty in epidemiologic research does not necessarily imply that the study cannot be used. In practicing their "art" epidemiologists make judgments about the extent of potential biases, such as an inability to measure exposure precisely or to arrive at valid estimates of dose. They also make judgments about the probable direction of these uncertainties, i.e., whether the misclassification of exposure is likely to have biased the risk estimate toward or away from the null. In particular, epidemiologists consider the possibility that exposure misclassification may have biased a dichotomous categorization of exposure toward the null and distorted an exposure-response relationship or that differential misclassification, e.g., recall bias biased the risk estimate away from the null.

The interpretation of new information about the effects of pesticide exposure must be considered carefully. If a novel observation is made in a study, but the decision was made not to use the study based on other criteria, the data should be archived "pending further investigation". This will add transparency regarding the use of data and preserves the possibility that novel data will be resurrected and used if corroborated by additional studies. Epidemiologists generally believe that no single observational study should be considered "definitive" and that the findings from well-conducted studies still require confirmation in other populations (the consistency criterion of Bradford Hill). Further, novel findings should have a biologically plausible framework if they are to be considered in the WOE. The possibility that individual statistically significant associations may be due to chance should not be ignored by the Agency or other reviewers.

Studies demonstrating no association with a pesticide exposure are equally informative in a WOE analysis as those that do so provided they meet the criteria for quality described above. Publication bias resulting from rejection or failure to submit 'null' studies is of some concern. However, several epidemiology journals, such as Epidemiology, have an explicit editorial policy of not rejecting 'null' studies on the basis of null findings alone when the study is otherwise well-conducted and address an important health concern. Publication bias is addressed in detail in the literature for meta-analytic epidemiologic analysis and methods, such as the use of funnel

plots, for detection of potential publication bias are routinely used in meta-analyses (Greenland, 1998).

The process for selection of studies in the WOE analysis for atrazine or more generally for incorporation in risk assessment begins with a comprehensive literature search to identify the full array of available studies. In conducting a WOE analysis the investigators should be assured that they have accessed the complete body of relevant epidemiologic literature available from peer-reviewed sources. A plan for the literature search should be developed that incorporates second and third level searches in the published literature as well as using the standard approaches of literature searching such as PUBMED (http://www.ncbi.nlm.nih.gov/pubmed/).

Careful evaluation of the findings should be performed by trained epidemiologists. For example, if drinking water studies are under evaluation, then epidemiologists experienced in conducting these kinds of studies, and scientists with expertise in water modeling and drinking water exposure assessment should be asked to review them. Similarly, review of occupational retrospective cohort studies should be conducted by researchers familiar with the issues of occupational exposure assessment and the statistical methods used for analysis of this form of cohort study. The field of epidemiology is diverse; therefore, those who conduct reviews and make expert judgment regarding inclusion of studies and eventual use of the data should have the training and experience required to do so.

Epidemiologic studies of reproductive outcomes have substantial strengths as summarized previously (Savitz and Harlow, 1991). As widely recognized, the fetus represents a susceptible subset of the population that may be exquisitely sensitive to the effects of environmental contaminants. The events that encompass conception and gestation and the exposures that may affect the processes of implantation, development and growth of the fetus occur in a relatively short time frame of one year or less. Thus, in the evaluation of potential adverse effects of *in utero* exposures to atrazine and other pesticides, epidemiologic studies focus attention on the critical temporal windows of exposure for each outcome; for example a single birth defect, spontaneous abortion or growth retardation. The truncated time frame of interest provides opportunity for more precise exposure assessment and reduces the probability of recall error when questionnaires are used to obtain information from parents. In studies of reproductive effects of disinfection byproducts, investigators were able to focus the analysis on exposures during specific months and weeks of late gestation in assessing associations with low birth weight, intrauterine growth retardation and pre-term birth (Hinckley et al., 2005).

When animal data show that a pesticide affects pathways essential to human reproduction and thereby establish biological plausibility for an effect, the Agency should examine the full suite of endpoints that may be perturbed (Moses, 1994). Epidemiologists have studied human fecundity by using time to pregnancy as a marker of success (Baird et al., 1986). This marker has been applied to pesticide exposures (Thonneau et al., 1999) and was described in early studies of agricultural workers exposed to dibromochloropropane on banana plantations in Costa Rica (Whorton et al., 1979).

Spontaneous abortion is an endpoint frequently examined in human studies of chemical exposures. For example, Arbuckle et al. (2001) studied the effects of pesticide exposures on the

risk of spontaneous abortion in a Canadian farm population. Studies of spontaneous abortion can be limited by the introduction of selection and reporting bias (Wilcox et al., 1984) and by the fact that in 20 to 25% of reproductive failures fetal loss is not manifested clinically (Wilcox et al., 1988). Late fetal loss, after 20 weeks of gestation can be assessed using data on stillbirth and neonatal mortality. Perturbations in fetal growth incorporate studies of birth weight (continuous variable), low birth weight (< 2500 grams), very low birth weight (< 1500 grams) and preterm delivery (37 weeks of gestation). Intrauterine growth retardation or small-for-gestational-age is assessed by comparing the infant's birth at a specific week of gestation to the norms for that racial/ethnic group using the 5^{th} or 10^{th} percentile as the basis for designating each birth. Examination of fetal growth parameters is frequently performed by analysis of birth certificate data that can be accessed readily and linked to environmental exposure data for the cohort or case and control samples.

The hormonal control of processes such as the onset of menarche, the patterns of menstrual cycle activity and the menopause provide the biologic framework for epidemiologic studies of these endpoints vis-a-vis pesticide exposure. Examples of studies which assessed associations with atrazine are available in the scientific literature (Farr et al., 2004; Farr et al., 2006). These studies should be incorporated into the WOE analysis.

Finally, it is widely recognized that approximately 50 percent of impaired fertility in humans is attributable to the male. Therefore, studies that assessed exposure to atrazine and semen quality (sperm concentration, percent motility, percent abnormal sperm) are important components for the WOE analysis. In particular, the study of Swan et al. (2003) should be informative. Some of the studies of reproductive outcomes that were not included in Case Study A should be relevant to the Agency's review of atrazine later this year.

Several issues in analysis and interpretation of findings from epidemiologic studies are often the source of discussion among epidemiologists, biostatisticians and others. First is the consideration of the interpretation of statistical significance and the sole reliance on the use of the p value for decision making (e.g. < 0.05). A series of papers exists in the literature in which epidemiologists and others have made a strong case for interpretation of the precision of risk estimates using confidence intervals in lieu of a strict interpretation of the p value (Savitz, 2003; Rothman and Greenland and Lash, 2008). It is important to evaluate all findings that show an elevated or reduced risk estimate or an exposure-response relationship regardless of statistical significance. Reviewers should consider the likelihood that the study lacked adequate statistical power and if needed, conduct the appropriate power calculations to assess the magnitude of risk that could have reasonably been expected to be detected if a true association existed.

A second issue that has been widely discussed by epidemiologists is that of multiple comparisons and the possibility that some findings in epidemiologic research may be due to chance as a result of multiple statistical tests. Epidemiologists warn against making adjustments to p values or confidence intervals that are inappropriate, overly conservative and wasteful of information (Rothman, 1990; Savitz and Olshan, 1995; Savitz, 2003; Rothman, Greenland and Lash, 2008). In considering this issue, reviewers should distinguish between analyses that incorporate multiple exposures and or outcomes in searching for any and all associations from those that explore *a priori* hypotheses in databases that permit multiple analyses to be conducted. The results of a

study should not be discounted simply because it efficiently and comprehensively evaluates multiple outcomes and multiple exposures (or multiple exposure indices). Several approaches to the multiple comparisons issue are discussed in detail in the literature (Steenland et al., 2000; Rothman, Greenland and Last, 2008). Multiple inference procedures involving hierarchical models are useful if the research interest concerns a joint hypothesis (i.e., a "family" of similar or "exchangeable" exposure-disease associations) or the purpose is simply exploratory (e.g., to answer the question, "Which, if any, of a "family" of exposure-disease associations should be followed-up in future investigations?"). However, in most instances the research question concerns a separate, single exposure-disease comparison or hypothesis. In the latter case, each comparison should be evaluated as if it were the only comparison in the study (this approach is also appropriate when there is doubt about whether the research interest is in a joint comparison or single comparisons – see Rothman, Greenland & Lash 2008, page 237).

Epidemiologists recognize the problems inherent in such analyses and the possibility of chance findings. Epidemiologists are cognizant of the inherent problems in conducting multiple statistical analyses and are trained to interpret these findings carefully and to employ many of the guidelines suggested in the Bradford Hill criteria discussed elsewhere in the report. The strength of an association, presence of a dose-response relationship, consistency of the finding across studies, coherence with available biologic information and other criteria are routinely employed in interpretation of data. Reviewers should also consider the reproducibility of observations among studies in terms of the direction of the effects observed, the magnitude of the effect and the concentrations at which these effects occur. The latter consideration is often ignored. In addition, the criterion of specificity of effect in the original Bradford Hill criteria has largely fallen into disfavor due to the systemic effects of many environmental exposures. Furthermore, the lack of evidence regarding biologic plausibility is not sufficient reason to discount or ignore that the remaining criteria may constitute sufficient weight of evidence in assessing an exposure-outcome relationship.

Epidemiologists often categorize exposures categorically, using tertiles or quartiles to evaluate potential dose-response relationships. Cutpoints for such analyses are typically developed from exposure data from the unaffected members of the cohort, or in case-control studies from the distributions among controls. Incorporating these studies in the WOE analysis will require considerable thought since the model departs from the traditional examination of linear dose-relationships. The shape of the dose-response curve in humans may not be linear if a threshold exists below which the chemical has no effect. The Agency should examine dose-response models and human data for other chemicals (non-carcinogens) to determine the optimal methods for integration of categorical exposure data from human studies into the WOE analysis.

Agency Charge

3. Case Study C: Human Incident Data-- Retrospective Case Study Using Diazinon

EPA is undertaking an effort to more systematically and transparently review and use human incident data in risk assessment/characterization or in problem formulation than has been done previously. As part of this effort, a case study using human incident data on diazinon is included.

Question 3.1 Case study C describes various analyses and evaluations that can be conducted when evaluating human incident data. Please comment on ability to use incident data for the following types of analyses: trend of incidents over time, frequency of reported symptoms, common product clusters, frequency of repeated exposure scenarios, and assessment of children vs. adult symptom profiles), in the diazinon case study and suggest alternative and/or additional analyses, if appropriate.

Panel Response 3.1

The majority of the Panel members agreed that little weight should be placed on self-reported incident data in routine risk assessment. Although human incident data can sometimes be useful in providing information on trends or differences in the frequency and severity of symptoms and whether human effects are consistent with those observed in toxicologic experiments or epidemiologic studies, the limitations of using human incident data for risk characterization and risk assessment outweigh the advantages. The major limitations include: 1) likely under-reporting of cases due to the lack of mandatory reporting other than for registrants; 2) uncertainty regarding the exact exposure conditions; 3) capture of largely only acute events and not events with long latent periods or events associated with long-term exposures; and, 4) the applicability of self-reported human incident data only to pesticides with notable acute toxicity.

The diazinon case study, as presented by the Agency, is unique because of the distinct symptoms resulting from cholinesterase inhibition and because of the risk mitigation measure of removing diazinon from residential use and the consequent reduction in incidents. Other pesticide groups, such as the triazine herbicide family, that do not produce symptoms of acute toxicity would probably not generate usable incident data for the following analyses.

Trend of Incidents over Time

Incident data have value in assessing effects resulting from changes in use patterns and implementation of use restrictions, and thus can serve as a good measure of the success of risk management procedures for minimizing acute toxicities. For diazinon, the reduction of reported incident cases appears to reflect its restricted access to the general public. It is unclear, however, whether exposures at lower levels would trigger incident self-reporting and whether these incidents would also have been reduced. For pesticides that do not pose marked acute toxicity potential, incident data are presumed to be sparse and inconclusive and of limited use in risk characterization/assessment.

It is worth noting that the different incident reporting systems seem to be generally reliable, particularly for evaluating frequency of incidents over time, such that all five systems showed a relatively similar decrease in incidents over time with the removal of diazinon. The data are also collected in a relatively uniform manner (e.g., product information, severity rankings and symptoms) among the different data sources. However, increased communication and coordination among the different reporting systems to make the data collection instruments more uniform could improve the collective data generated.

Frequency of Reported Symptoms and Frequency of Repeated Exposure Scenarios

The reporting of similar symptoms following exposure to a particular pesticide product by different individuals within a defined time period should raise concern for the use pattern of this particular product. Reporting of common symptoms by exposed persons may also indicate a pattern of acute toxicity previously undetected in experimental studies or in reports by individual registrants. Under circumstances in which incident data reveal a health outcome that was not previously observed in toxicology or epidemiologic studies, such human incident data could be valuable in terms of exploring biologic plausibility associated with specific pesticide exposures. However, it is very likely that although these incident reports can be effectively collected, the available data may not adequately discriminate between high and low level exposures.

Common Product Clusters

Common product clusters resulting from incident reports occur, but are more important as an immediate public health concern, rather than serving a risk assessment purpose. For instance, methyl parathion poisoning cases in the southeast in the 1990s resulted from misapplication of the product. Such incident data would be inappropriate for consideration in risk analysis. This is also true for cases of abuse or suicide in which the data would not be relevant for risk management or risk assessment because the exposures for those abuse/suicide cases would be expected to exceed label recommendations or be by ingestion. If clusters of incidents point to a risk management failure, then certainly incident data should be used to protect those individuals who might be at unanticipated elevated risk.

One Panel member thought that a cluster is likely to indicate a more severe problem than an isolated case, but not more severe in proportion to the number of persons reported. This is because reporting by one individual is likely to stimulate reporting by others, so that a cluster is artificially created in excess of what would happen if reporting were independent. However, the reporting of clusters has contributed to the risk assessment process, for example in the aldicarb contaminated watermelon episodes in California (Goldman et al., 1990). In this instance, the reported cluster played an important role in the risk assessment for this particular pesticide.

Assessment of Children vs. Adult Symptom Profiles

Comparisons of the distributions of symptoms in children and adults can provide supportive evidence of similarity of effect, but lack of similarity does not necessarily mean that the mechanisms are different because they could reflect: 1) different levels of sensitivity of reporting (e.g., effects in children may be more likely to be reported than similar effects in adults); 2) different routes of exposure; and 3) different sizes of populations exposed (e.g., small numbers

exposed persons might result in less certainty in the distribution of symptoms). It is also likely that the magnitude of exposure (or dose) to certain pesticides that would trigger the reporting of the incident would be very different between adults and children.

Other limitations of using human incident data for risk characterization and assessment further reduce its utility. For instance, follow-up on these incident reports is typically minimal, limiting the information on possible long-term consequences. Since more subtle, long-term effects are typically more difficult to detect in animal studies, better follow-up of reported incidents may be beneficial in that regard. It is also very likely that self-reported incident data may consist of anecdotal or emotional observations that have limited factual evidence of connection to a specific exposure. Because the quality of the self-report incident data is extremely difficult to determine, it will no doubt introduce bias and uncertainty in future analyses.

Overlap of self-reported cases among the five different incident databases is also a concern. It is unknown whether the overlap is concentrated on severe poisoning cases or in certain geographic areas. It appears that these sources of data are studied independently until the later stages of analysis, when results are compared across databases to identify signals of a problem. It is, therefore, necessary to consider how these five sources might be used in combination at earlier stages, not necessarily by matching cases, but at least by organizing the data in ways that draw on the strengths of the various sources. This might be accomplished by focused study in one or two areas where three, four, or even all five reporting systems operate.

One Panel member recommended that the Agency should clarify the presentation of human incident data contained in Attachment C with respect to how the data were compiled for the tables in Appendix B. In addition, several other Panel members suggested that before further considering the utility of human incident data, potential confounders or other exposures that may have been responsible for the symptoms reported should be identified and controlled in the analysis.

Question 3.2 OPP plans to conduct analyses of human incident data like that described in Case study C for other pesticides undergoing registration review. In light of scientific issues discussed in Questions 3.1, OPP requests input from the Panel on factors to consider when evaluating the reliability of human incident data and determining the relative weight that should be placed on such data in risk assessment/characterization or in problem formulation.

Panel Response 3.2

In general, very little weight should be placed on incident report data in routine risk assessments because of their diverse nature with regard to estimated dose levels, product characteristics, and the ability of the observer to assess symptoms accurately. If the numbers of incident reports are large, the exposures are well-estimated and the symptoms are highly consistent, then perhaps incident data would be useful. If incident clusters point to a risk management failure, then certainly incident data should be used to protect those individuals who might be at unanticipated elevated risk. In cases of abuse or suicide, the data would not be very helpful for overall risk management because these exposure levels would be well beyond label recommendations or by another pathway such as ingestion. Reports that contain vague or subjective information, including flu-like symptoms or those that could arise from physiological stress should be interpreted with caution. These reports could represent general symptoms from a variety of illnesses or conditions including infectious diseases or stress. It may be impossible to distinguish pesticide effects from other conditions that could mimic those due to exposure to the product. Fear of poisoning could lead to neurobehavioral symptoms, with the typical reactions associated with stimulation of the sympathetic nervous system. The diazinon case study was uniquely suited for such an analysis because of the well-defined set of acute symptoms due to its anticholinesterase activity. The availability of data before and after introduction of risk mitigation in removing diazinon from residential uses and the consequent reduction in incidents were also relatively unique. Most other pesticides would probably not be adaptable to such a clear presentation.

The incident reporting systems described in Attachment C seem to be generally reliable. This is particularly clear when evaluating frequency of diazinon incidents over time, such that all five systems showed a relatively similar drop in incidents over roughly the same time period. Considering the lack of specific training for persons recording information and the non-specific nature of some symptoms, relatively good reliability was observed among the reporting systems for different classes of signs/symptoms associated with reported diazinon exposure. The recognized strength of this type of data, in contrast to information from animal toxicity studies, is that responses in humans are detected under real-life situations, with conditions of differential individual sensitivity, modifying factors and other influences possible in the human population. A major weakness of this type of information, however, is the uncertainty regarding the exact exposure conditions, concentration, or amount of the chemical to which the individual was exposed. Even the specific chemical may not be known with certainty, and it is likely that no information exists on inert ingredients or co-exposures that may have been involved. Follow-up for these incidents is typically minimal, limiting the information on possible long-term consequences. As more subtle, long-term effects are typically more difficult to detect in animal studies, better follow-up of reported incidents may be beneficial in that regard. As noted in the draft framework, the various incident reporting systems may report different symptoms and signs

(or the same signs called something else), or differential severity rankings for symptoms of toxicity. Considering the weaknesses of this type of information, the weight given to use of incident data in risk assessment process should be low, with a more qualitative than quantitative influence on the process. Surveillance for unanticipated effects in incident reports could be useful in suggesting alternative mechanisms of action or toxicities not previously described for a pesticide.

The limitations of the incident data for diazinon out-weigh the possible benefits of the use of such data for risk assessment/characterization. One possible enhancement to the self-reported incident data would be to implement the collection of appropriate specimens or samples, where feasible, from individuals who call in to report symptoms in the future. Laboratory analyses of such specimens and sample would serve to validate the reported human incident data and also provide critical information about the levels of exposure (dose) that are responsible for symptoms among exposed individuals. Such data would also be useful in differentiating symptom profiles and exposure levels in children versus adults. Although logistic issues, costs and feasibility of implementing specimen collection may be currently beyond the Agency's capability, the idea could be discussed further among the agencies collecting human incident data. Perhaps a limited pilot study may be feasible.

Although reliable data are generally lacking for most case studies, the Agency is encouraged not to overlook the rare cases with sufficient documentation, or clinical case reports published in the open literature with extensive follow up after poisoning. Some of these reports may uncover new toxicity endpoints of concern and should be added to risk assessment. For example, chronic neuropsychological sequelae were manifested among those who appeared to recover from cholinergic signs and symptoms after acute organophosphate pesticide poisoning which involved a different mode of action (MOA) than cholinesterase inhibition.

The Panel concluded that incident reporting data such those considered in the diazinon Case Study (Attachment C) have some value for problem formulation and hazard identification in the risk assessment process, but their application in risk characterization is very limited unless follow-up information and or laboratory data from individual incident cases become available.

Agency Charge

4. Case Study B: The Agricultural Health Study Comparison of Exposure Assessment Approaches

The Agricultural Health Study (AHS) is a large long-term prospective epidemiological study that is collecting data on the health and work practices of licensed pesticide applicators in Iowa and North Carolina. The AHS is focusing particularly on the exposure of applicators to 50 chemicals, including many of the most widely used pesticides. The study also collects information on other possible agricultural exposures, and many lifestyle factors. Investigators with the AHS have published over 100 publications on a variety of topics including characteristics of the cohort and cancer and non-cancer health outcomes that have been observed in the cohort (http://aghealth.nci.nih.gov/).

Question 4.1: The Agency believes prospective epidemiology studies with robust exposure assessment, like the AHS, have the greatest potential for use in risk assessment especially for enhancing problem formulation and risk characterization. Please comment on appropriate ways to use of these types of epidemiology studies in risk assessment.

Panel Response 4.1

General Considerations

The Agency is urged to review other situations where epidemiological data have been used in risk characterization, e.g., arsenic, as these may prove useful in developing the framework. Considerations and review of data in risk assessment of chlorpyrifos (SAP, 2008) may provide a case study of how epidemiological data can be used in risk characterization. In this evaluation, a weight-of-evidence approach was used in the final determination. In the case of developmental neurotoxicity from chlorpyrifos exposure(s), prospective epidemiologic studies, with individual measures of chemical exposure, suggested that the dose-response relationship may be much different in humans than in animals. These prospective studies suggested neurodevelopmental effects may occur in humans with early exposures to chlorpyrifos, but with possibly different types of neurodevelopmental outcomes, and at potentially much lower levels of exposure, than in animal studies. The Panel concluded overall that data from both epidemiologic and animal studies suggested a connection between chlorpyrifos (and possibly other chemicals with anticholinesterase activity) and neurodevelopmental outcomes, but that dose-response relationships, and even mode of action, may not agree between these different ways of "looking" at end effects. One caveat to the conclusions was that the several anticholinesterase agents would have been acting on the same target enzyme, so sorting out the impact of any single compound would have been extremely difficult. This same concern also is present for the AHS in that any one pesticide may be present concurrently with one or more others acting on the same target system, so conclusions need to take the mixture into consideration.

The eventual resolution of large discrepancies between epidemiologic and animal studies in apparent dose-response relationships, or substantial differences in types of responses between animal and epidemiologic studies, is unclear from the draft framework. How does the weight-of-

evidence concept rationally weigh-in on decision making in this type of situation? On page 31 of the draft framework, it is stated that "when animal and epidemiological data do not provide a consistent toxicological picture…more weight would likely be given to those studies with robust study design and availability of replication or confirmatory data". Further, it asserts that "in most situations, the epidemiological study may not be sufficiently robust for deriving quantitative risk assessment values". If epidemiologic data are used to derive quantitative values in risk assessment, determining a process for decision-making in cases in which wide differences are observed in dose-response relationships between animal and epidemiologic studies could clarify the framework and its implementation. At present, it can be argued that in most cases animal studies can more "robustly" describe dose-response relationships, using the least amount of time/resources, etc., and therefore may currently provide a more reasonable approach for characterizing dose-response relationships, for evaluating mode of action, and for quantifying points of departure. That does not rule out the possibility that in specific situations, either epidemiological data or possibly even incident data (e.g., aldicarb intoxications from watermelon consumption) could play an important role in either defining or directly contributing to estimates of departure points.

Clarification of Study Designs

In an attempt to simplify the task of assessing the utility of epidemiologic studies for risk assessment, the EPA has grouped these studies into ecologic, retrospective and prospective designs. The SAP discussed design features of different types of epidemiologic studies, methods to qualitatively evaluate them, their potential limitations, and how to make efficient use of the information that is obtained from these studies in the risk assessment and/or risk management process. Although prospective cohort studies can offer many advantages, it is typically not possible to generalize about what study design is best or most appropriate.

A clear description of different study designs and their strengths and limitations for testing hypotheses or evaluating the weight of evidence for a particular cause-effect association is needed. A presentation of study designs from weakest to strongest could include a description of: case reports (i.e., acute poisoning incidents, physician case reports), case series, ecologic studies over time and/or place, clusters, case-control studies, retrospective or historical cohort studies, prospective cohort studies, and mixed designs (e.g., nested case-control, case-cohort, case-crossover). The most appropriate study design will depend on the question being asked and the data requirements (e.g., need for cross-sectional biomonitoring data, historical data on changes in exposures over time, risk estimates for known carcinogens, hypothesis generating studies in populations where cancer incidence has increased, investigation of clusters of potential occupational or environmental origin).

The EPA draft framework should provide additional clarification on the different types of retrospective studies, with a distinction made between case-control studies and retrospective (or historical) cohort designs. Historical cohort studies offer many of the same advantages of prospective cohort studies, with the added advantage of providing much quicker answers to research questions. Further, nested case-control studies offer many advantages and because of the smaller sample needed, they are much more cost-efficient than cohort studies in studying rare outcomes. Recognizing this, it is typically assumed that the prospective cohort is the strongest

observational study design. This is in part because they provide the "opportunity" to collect the most valid and reliable exposure and/or absorbed dose data and because the characterization of exposure occurs prior to the development of disease or other outcome, thus clearly establishing the temporal sequence.

In research on the health effects of exposure to pesticides, regardless of the health outcomes, the methods of ascertaining cases, classifying diseases, selecting controls etc., exposure assessment is and will remain the most challenging aspect. The Agricultural Health Study (AHS) is an example of a prospective cohort study that has set the standard for future investigations, has developed and evaluated innovative methods of exposure assessment, and will be producing data for many years that are extremely relevant for the assessment of health risks associated with pesticide exposures.

Prospective Epidemiologic Studies and Use in Risk Assessment

The main advantage of prospective cohort studies is that individuals are followed forward in time and exposure is determined prior to the development of disease. This presents advantages and opportunities for data collection relevant to the risk assessment process. These include:

a. Single and multiple/mixtures exposures that represent environmentally relevant concentrations (and associated absorbed doses) can be measured prior to the development of the outcome.

b. Changes in exposures and/or dose can be measured over time. Cumulative exposures can be estimated as well as peak exposures and variation within and between individuals over time.

c. Biologic markers of exposure can be evaluated in relation to measured and/or predicted exposures using alternative methods, models, records or questionnaire data.

d. Dose validation studies are possible.

e. Biological markers of susceptibility (gene-environment interactions) can be measured that may modify relationships between pesticide exposures and health risks. This information can help to characterize risk.

f. Early biomarkers of effect, which may be precursors to clinical disease, can be measured; this information will be relevant for evaluation of the proposed mode of action/mechanisms in humans.

g. Quantitative exposure data and biomarkers that are intermediate on the pathway from exposure to disease can be collected.

h. Information on lifestyle, other behavioral factors, and other occupational or environmental exposures that may modify exposure response relationships or act as confounders can be obtained prior to the outcome.

Although prospective study designs have several clear advantages, they may have limited power to look at rare outcomes and can take many years to obtain results. Further, even though biomonitoring is considered the preferred approach (gold standard) to obtain valid and reliable dose estimates in these studies, these types of measures are often collected at only one point in time, and may be available only in a subsample of a cohort. Most often a spot sample (as compared to 24 hour samples) of urine or a single sample of serum is collected. Depending on the characteristics of the pesticide, the concentration may reflect only the most recent exposures.

The AHS Study: Considerations for Risk Assessment

Several important considerations were explored with Dr. Michael Alavanja, principal investigator of the AHS, after his presentation and later during the Panel's deliberations. These topics are relevant to exposure assessment and exploration of potential effects of pesticides on cancer and reproductive outcomes.

Exposure Assessment

Exposure assessment for the AHS is substantially improved over many previous epidemiologic studies of pesticide exposure. As part of the joint efforts of the EPA and AHS investigators, the PHED (Pesticide Handlers Exposure Database) data will be used to estimate exposures for cohort members (occupational exposures of licensed applicators) and compared with exposure assessments (i.e., intensity scores) for the AHS. As the Agricultural Handlers Exposure Task Force accumulates data over the next few years, these should be employed. These newer data are designed to replace the PHED to reflect changes in the equipment and newer protections of present day agricultural practices. However, in terms of estimating lifetime exposures, the more historical residues would have been accumulated with the conditions prevalent during the PHED data accumulation. Therefore, there should be consideration of the appropriate database for long-term exposure estimates. Caution is urged in the use of self-reports for historical exposure assessment when the length of recall is very long since memory may not be accurate.

Additional refinement of AHS exposure assessment may be feasible depending on the availability of banked samples and resources to analyze these samples that may be collected in the future. Biologic and environmental sampling and laboratory analyses can be conducted on population subsets (i.e., occupational groups or bystanders including spouses, farm workers and children) and to ensure cost-effective and representative data (Bakke et al., 2009).

A second issue explored during the discussion involved collection of water samples from farm residences for assessment of pesticide contamination. Although it is not clear to what extent such samples are currently available or could be collected in the future, they could be used to ascertain exposure through the drinking water pathway which does not appear to be taken into account in the current exposure assessment. Family farms typically rely on private wells for domestic consumption. Because many agrichemicals are known to contaminate superficial aquifers in regions of heavy pesticide application, this pathway should be evaluated as a potentially significant contributor to exposure.

Refinement of exposure assessment to include domestic exposure to water and house dust, along with occupational exposures may improve exposure assessment and reduce misclassification. The domestic water and/or dust pathway may be particularly important for the female members of the cohort in considering potential adverse reproductive outcomes. For women who work outside the home or do not participate in farming activities these pathways could be significant sources of exposure to pesticides.

Reproductive Outcomes

Prospective studies of the nature of the AHS may take a long period of time to compile results (cohort studies of cancer may run for 20-30 years or longer). This can substantially delay the usefulness of these data for current risk assessments. However, there are short term opportunities for evaluation of reproductive outcomes.

Although a substantial number of women is included in the cohort (approximately 31,000 enrollees), these women are now an average of 56 years of age and therefore beyond the age of childbearing. However, for a portion of the female cohort members who delivered an infant during the study, data linkage with the birth certificate files in the states of Iowa and North Carolina may provide an opportunity to explore hypotheses relating pesticide exposures to outcomes such as birth weight, low birth weight, intrauterine growth retardation and premature birth. Data linkage may also facilitate comparison between self reported reproductive outcome data and self reported data for women who gave birth during the study.

Many adverse reproductive outcomes are prevalent among live births, affecting 5 to 20 percent of women and their children. Therefore, although the numbers of women who gave birth during the follow-up period will be substantially smaller than the total number of enrollees, sufficient statistical power may exist to evaluate these outcomes for specific pesticide exposures. Further, because the relevant period of exposure for reproductive events is likely to be no more than one year, reducing the likelihood of information bias, further analyses of reproductive endpoints is recommended.

Conclusion

Overall, the use of data from a well-designed and carefully executed prospective cohort study such as the AHS should provide the best opportunity for reaching the ultimate goal of incorporating epidemiologic data into risk assessment. These data may eventually be useful for the risk characterization stage of the process. Further, they also may be useful in comparing dose-response data between humans and laboratory animals for some outcomes. Other epidemiologic designs such as mortality analyses and case-control studies, have already identified farmers and agricultural workers as high risk groups for specific cancers and other disorders. These early studies can be used to inform the problem formulation stage of risk assessment. The AHS has the potential to extend the use of epidemiology to risk characterization for agricultural chemicals. Such advances in risk assessment methodology could enhance the usefulness of the risk assessment paradigm for the eventual protection of public health.

The importance of involving relevant stakeholders and experts from all of the appropriate areas in both the planning and assessment stages was stressed. This would provide greater confidence in the proposed work plans, and in the validity and utility of conclusions. The use of interdisciplinary collaborations in this work would add value to the deliberations and would provide an opportunity to establish a sound way of operating when assessing the reliability and relevance of risk assessments in which both epidemiologic and classical toxicologic information bases are combined. The joint assessment of data sets by experts from different areas would help to ensure objectivity of the interpretations, and the quality of the data used.

Question 4.2: The Agency uses a predictive, scenario-based approach to calculate risks associated with the registered use patterns of pesticides. Estimates of risk based on varying levels of protective equipment, application methods, and use conditions are presented. The results of these assessments are used to specify label conditions that are required to support the new registration or continued registration of pesticides. In contrast, the goal of epidemiologic exposure assessment within the AHS is to develop a relative exposure ranking of individuals who are actual pesticide users within a cohort. It is not feasible to directly measure actual exposure in observational analyses such as the AHS. The AHS exposure information is ascertained from questionnaires completed by individual cohort members. Because the AHS and the Agency have different purposes for evaluating pesticide applicator exposure, there are inherent differences in the occupational handler exposure methodologies between the AHS and Agency. How to reconcile these differences in order to make optimal use of the AHS in developing regulatory policy is under investigation by a collaborative effort between EPA's OPP and investigators involved with the AHS. Case study B details a three step analysis plan for accomplishing this goal. Please comment on the proposed plan for comparing the exposure assessment approaches between the Agency and the AHS. Please include in your comments the scientific value of this comparison along with additional and/or alternative analyses which could be conducted.

Panel Response 4.2

The Panel recognized the merit of finding commonalities between the Agency's exposure assessment methodology and the AHS exposure metrics. The Agency's method results in estimates of workers' exposure as an input into risk assessment that form the basis for setting exposure limits for workers. Finding commonalities to the AHS exposure metrics would provide a way to extend the usefulness of this large and growing data base for the protection of pesticide users.

The Agency presented a single illustration for a step 1 comparison between the ground boom and air blast application. The comparison between the Agency and AHS methods is reasonable as they share a similar goal of characterizing the external exposure, and both can assess the relative exposure between work tasks. Some common grounds for the two approaches were recognized, e.g., use of PHED data, reduction of exposure by PPE. The Panel agreed with the Agency that extensive scenario-by-scenario comparisons are needed to characterize more fully the similarities and differences between the two methods before bridging methodologies for the two methods can be achieved.

The Panel suggested that the document be revised and streamlined. The AHS exposure metrics and scores, and methods of calculating these ordinal values are transparent and available in the published literature (Dosemeci et al., 2002). However, the Agency's method is less clear. The calculation of exposure (dermal, as expressed in mg a.i./day) using the PHED database could be clarified. Further discussion of the unit exposure parameters can also be added.

The Panel recommended that discussions of the variability and uncertainty associated with the foundational databases be included for each method, e.g., PHED, AHS's self-reporting and input parameter values. As far as possible, distributional analyses, not merely the point estimates

could be added to the comparison and exposure models. It is difficult to compare the two methods in that both have a series of steps. Thus, it is anticipated that the elements of the two metrics have to be "picked apart" to identify which of the elements measure the same or similar facets of exposure.

The second step involves using biomonitoring datasets from the AHS to compare the exposure metrics of both methods. This step can further elucidate factors that contribute to the differences between the two exposure estimation methods. In general, evaluation based on biomonitoring data would be less complex for shorter-term than long-term exposures. Attempts to compare exposure predictions from the AHS and the PHED generated models will be most fruitful if a common measure is used to connect the two. For example, a biologic monitoring validation study could be conducted to obtain by questionnaire PPE information and application activities (to feed into the AHS algorithm), pesticide application information (i.e., volume used and active ingredient) and other required variables for the PHED model. Exposures can be calculated using both methods and compared with the "gold standard" urinary metabolite values. The uncertainties associated with the biomonitoring data are likely to be large, and it is important that they are well-defined because this dataset is to be used as the link between the two exposure models. Measurement error, and/or bias can be assessed. When assessing the biological significance of the biological data it would be more helpful to think in terms of the coefficient of determination (R^2) that is interpreted as the proportion of variation explained, rather than the correlation coefficient (r) and its associated level of significance. Sensitivity analysis can be performed at each step. However, this will be more readily achieved for the AHS model than the PHED model. However, it would be worthwhile, because it would allow the identification of major discrepancies and align the focus for further investigations.

In comparing the two sets of metrics, it is not necessary to classify each of the thousands of subjects. A proper random sample (not necessarily a simple random sample; stratified random or other variations might work) could be used to get at the general structure of the relationships, including individual correlations. Even when concern is focused on a small group with some specified outcome, a random sample should be adequate if it is supplemented by 100% sampling of the affected subgroup. It was recognized that classifying the subgroup at a later time might raise difficulties of consistency, but the savings in time and effort could be great. Truly random sampling of some kind will be critical to the use of any sampling approach.

The proposed third step involves a larger comparison of the exposure metrics as applied to atrazine and alachlor users in the AHS database. Additional complexity can be anticipated in this step. Thus, the Panel is supportive for the Agency's feasibility analysis before proceeding to this step.

Overall, the Agency sets out to achieve two goals. One is the evaluation of exposure and risk for specific chemical(s). The other is to strengthen the Agency's current method to estimate the exposure using PHED data. It may be necessary for the Agency to establish the priority for these two goals should the task for achieving both become unattainable within the Agency's operational timeline. The Agency indicated that a plan is underway to update PHED. Accordingly, the latter goal of strengthening the Agency's exposure assessment approach may need to be modified or re-defined in the future.

Question 4.3: The Agency has a long-term goal to understand the extent to which findings from the AHS are generalizable to other populations, such as pesticide applicators in states other than North Carolina and Iowa or those who may be exposed to pesticides through other pathways and under different use conditions. Please provide suggestions for analyses which could be conducted to make best use of the results of AHS in a broader regulatory context.

Panel Response 4-3

Direction from EPA staff was provided to include a discussion on the generalizability for pesticide applicators and handlers from other states and post application workers. Further, a discussion of the generalizability of results for bystanders such as farm family members and children and the general population was requested.

Although the states of Iowa and North Carolina were deliberately selected for the AHS to provide some diversity of pesticide use patterns across the United States, other agricultural environments may not be adequately represented in the exposures found in the AHS. Therefore, great caution needs to be exercised in generalizing the health risk findings of this study to other agricultural populations, or even more so to the population at large.

That said, certain standard epidemiologic analyses can be performed to ascertain potential applicability of findings to other populations or population segments. These include analyses of the same pesticide between Iowa and North Carolina as well as comparison of findings across racial and ethnic groups when the data are available to make such comparisons. Analyses by gender, age group and by type of applicator (commercial, private) also may be informative and are likely to be conducted by the AHS investigators during their analyses.

The range of human biologic responses to a chemical agent in affecting a specified demographic subgroup in State A is likely to be virtually identical to the range, at the same doses or exposures, in State B. Therefore, extrapolation from state to state must focus mainly on exposure. Further, state boundaries are, for this purpose, artificial and almost irrelevant to exposure, though they may be important for such things as data sources, local customs (e.g., use of PPE) and possible state or local regulations (e.g., aircraft spraying), types of workers (e.g., education, supervision) but not types of work.

Results should be generalizable to "pesticide applicators" in other states if populations (racial/ethnic composition) and socio-demographic factors are similar. Factors that may modify the relationship between exposure and outcome are important (e.g., genetic factors that influence susceptibility, carcinogen activation/metabolism and other factors that may act synergistically such as smoking and other behavioral characteristics). It was recommended that a clear understanding of these and other geographic factors (such as local weather conditions and pesticide use patterns) is needed and that generalization of results to other states, based on geopolitical boundaries only, may not be appropriate.

The Panel considered exposure via other pathways and under different use conditions. Because of the broad nature of this question, which is beyond the applicator exposure scenarios, it would be clearer if the Agency presented a set of schemes and metrics for the exposure scenarios for

which the Agency is planning to apply the knowledge learned from the AHS. The AHS Intensity Level metric may have the flexibility to handle multiple exposure pathway and use conditions. It appears that if novel situations arise, it would be feasible to modify or add to the determinants of the Intensity Level calculation, but this is relevant primarily for occupational exposures.

It may be difficult to generalize the occupational exposures and associated health risks to the environmental/residential setting where exposures may be primarily via drinking water, dust, or bystander exposures. It would certainly be necessary to invest in expensive biologic assessment programs to check on the reliability of exposure models modified to use with "bystander" populations. The sample sizes need not be too large providing that the samples are representative of the strata within the general population.

Analyses to Make Use of the AHS Data in a Broader Regulatory Context

Quite early in the document a clear, succinct statement should be made about how analysis for regulatory purposes differs from analysis for scientific purposes. This matter comes up repeatedly (especially in the addendum to Case Study B), but it is never explained in a way that is clear for a non-scientist.

The Panel considered uncertainty associated with extrapolation of animal data to humans in comparison to the uncertainty associated with extrapolation from one human population to the next. National Toxicology Program (NTP) and International Agency for Research on Cancer (IARC) classifications do rely on human as well as animal data to determine categories of carcinogens, whereas EPA will on occasion use only animal data for quantitative risk assessment. EPA scientists indicated that the default, of using animal data only is not preferable to using human data. These comments prompted a discussion on the relative uncertainty of dose-response assessment based on toxicologic vs. epidemiologic studies. It is important to distinguish between qualitative and quantitative aspects in discussing the utility of epidemiologic data for risk assessment. While epidemiologic data are frequently weighted in classification systems, such as those of International Agency for Research on Cancer (IARC) which are based principally on hazard identification, their contribution to risk characterization is determined by the extent to which they inform dose-response relationships. As a result, the extent to which epidemiologic studies contribute to risk characterization varies as a function of this aspect. Uncertainty associated with characterization of dose-response relationships in human populations will depend on the differences in exposures between the populations studied and other characteristics of those populations.

One potential opportunity for analysis was described by Dr. Alavanja. HPEE, or high pesticide exposure events, have been reported by a number of applicators, and approximately 20% had symptoms (chronic neurologic, respiratory (wheeze) and detached retina), but less than 5% were reported to health care providers. It would be of interest to see how many of these events were captured in the incident databases described in question 3.

Overall, to achieve the goal of generalization, it will be necessary to use interdisciplinary teams of experts and end users at both the planning and evaluation stages. Further, the success of the process will depend on the establishment of transparent frameworks for evaluating the quality of

all data (including animal toxicological, human incident, and epidemiologic data). Poor quality information can add to the noise, and reduce the ability to discern real effects and to make accurate predictions.

ATTACHMENT E, Pt. 3

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ATTACHMENT F

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
CHEMICAL SAFETY AND
POLLUTION PREVENTION

MEMORANDUM

Date: 6/30/2011

SUBJECT: Chlorpyrifos: Preliminary Human Health Risk Assessment for Registration Review

PC Code: 059101
Decision No.: NA
Petition No.: NA
Risk Assessment Type: Single Chemical Aggregate
TXR No.: NA
MRID No.: NA

DP Barcode: D388070
Registration No.: NA
Regulatory Action: Registration Review
Case No.: NA

CAS No.: 2921-88-2
40 CFR: 40 CFR§180.342

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ATTACHMENT F

Background

Attached is the USEPA ("the Agency") Health Effects Division's (HED) preliminary human health risk assessment for the pesticide chlorpyrifos. Chlorpyrifos is a currently being evaluated under the FIFRA section 3(g) registration review program which requires the re-evaluation of pesticides on a 15 year cycle. This preliminary assessment is provided in support of the registration review process for chlorpyrifos.

Chlorpyrifos (O,O-diethyl O-(3,5,6-trichloro-2-pyridyl) phosphorothioate) is an organophosphate (OP) insecticide, acaricide and miticide used to control a variety of insects. Chlorpyrifos was first registered in 1965 for control of foliage and soil-borne insect pests on a variety of food and feed crops. Currently registered uses include food and feed crops, golf course turf, greenhouses, non-structural wood treatments such as utility poles and fence posts, ant bait stations, and as an adult mosquitocide.

In June 2000, during the reregistration process, the Agency released its revised human health risk assessment (D.Smegal, 6/8/00, *Human Health Risk Assessment, Chlorpyrifos*, U.S. EPA). Subsequently, the technical registrants voluntarily cancelled and phased out certain uses of chlorpyrifos. The voluntary cancellation/phase out expeditiously addressed the food, drinking water, residential and non-residential uses posing the greatest risks estimated for children. Risk mitigation measures include eliminating use on tomatoes, restricting use on apples, phasing out termiticide use, canceling all homeowner use product registrations (except insect bait stations), and canceling uses in schools and parks where children may be exposed.

An Interim Reregistration Eligibility Decision (IRED) was issued in February 2002. The IRED included additional mitigation measures addressing occupational and ecological risks not addressed by the 2000voluntary cancellation/phaseout. To mitigate worker risk estimates of concern, a combination of reduced application rates and seasonal maximum limits, increased retreatment intervals, increased PPE and/or use of engineering controls were required as well as increased REIs for a number of crops. Upon completion of EPA's assessment of the cumulative risks from the organophosphate class of pesticides, the chlorpyrifos IRED became final (as a RED) in July 2006.

The June 8, 2000 HED human health risk assessment for chlorpyrifos was largely based on adult laboratory animal data for cholinesterase inhibition and the application of default uncertainty factors, including the retention of the 10x FQPA Safety Factor. Since 2000, there has been extensive and ongoing research on various aspects of chlorpyrifos including its neurological effects in *in vitro* and in animals and humans following gestational and post-natal exposures, and its pharmacokinetics. In 2008, the Agency developed a draft issue paper reviewing the science available for chlorpyrifos which was reviewed by the FIFRA Scientific Advisory Panel (SAP; September 2008). Since the SAP, new studies have been submitted to EPA including a special acute inhalation study, an immunotoxicity study, and acute and repeat dose comparative cholinesterase assays (CCA) in juvenile and adult rats. The CCA studies examined toxicity for both chlorpyrifos and chlorpyrifos oxon. This preliminary hazard characterization and risk assessment for chlorpyrifos includes existing data, findings of new studies made available since the 2000 assessment, and considers comments from the 2008 SAP reviews. This assessment is

considered preliminary and presents risk estimates from both the 2000 assessment (based on toxicity studies using adult animals) and risk estimates based on benchmark dose (BMD) analyses, where appropriate, from sensitive studies which use ages relevant to human exposure. For the final chlorpyrifos human health risk assessment, including determination of the most appropriate toxicological points of departure and FQPA factors, the Agency will consider the weight of the evidence of all available data and take into consideration any comments received.

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1.0 Executive Summary

Use Profile

Chlorpyrifos (0,0-diethyl-0-3,5,6-trichloro -2-pyridyl phosphorothioate) is a broad-spectrum, chlorinated organophosphate (OP) insecticide. Registered use sites include the following: food crops, including fruit and nut trees, many types of fruits and vegetables, and grain crops; and non-food crops such as forage, golf course turf, industrial sites, greenhouse and nursery production, sod farms, and wood products. Public health uses include aerial and ground-based fogger treatments to control mosquitoes. There are currently no homeowner uses except for roach bait products. Permanent tolerances are established (40 CFR§180.342) for the residues of chlorpyrifos in/on a variety of agricultural commodities (including meat, milk, poultry and eggs). There are also tolerances for use in food handling establishments. Chlorpyrifos is manufactured as granular, microencapsulated, soluble concentrate/liquids, water dispersible granular in water soluble packets (WSP) and wettable powder packaged in WSP formulations, as well as impregnated paints, cattle ear tags, insect bait stations and total release foggers. There is a wide range of application rates and methods.

Hazard Identification

The toxicology database for chlorpyrifos is substantially complete (40 CFR 158.340 guideline studies have been submitted) and has been used to characterize toxicity and for selecting points of departure for purposes of the current risk assessment. Chlorpyrifos, like other OPs, binds to and phosphorylates the enzyme, acetylcholinesterase (AChE), in both the central (brain) and peripheral nervous systems leading to accumulation of acetylcholine and, ultimately, to clinical signs of toxicity. In 2000, the Agency concluded for chlorpyrifos that inhibition of cholinesterase (ChE) was the most sensitive effect in all of the animal species evaluated (rats, mice, rabbits dogs) and in humans, regardless of exposure duration. The Agency is maintaining at this time, based on available data, that cholinesterase inhibition (ChEI) provides the most sensitive dose-response information for deriving points of departure for chlorpyrifos. In animals, significant inhibition of plasma and red blood cell (RBC) ChE occur at doses below those that cause brain ChE inhibition.

The toxicity database of laboratory animal studies spans multiple routes of exposure (oral, dermal, inhalation), animal species, lifestages and durations. The database consists of studies ranging from a single exposure (acute) to subchronic and chronic toxicity. Guideline studies on developmental toxicity and specifically developmental neurotoxicity toxicity, and reproductive toxicity. The metabolism and pharmacokinetics of chlorpyrifos is well-characterized due to a variety of studies in different species and lifestages. Recently, a comparative cholinesterase assay (CCA) was submitted which provides information on comparative sensitivity in adult and juvenile rats from acute and repeated exposures to both chlorpyrifos and its oxon. Special studies have been submitted including an acute neurotoxic esterase rat study, cognitive rat study, and recently an acute inhalation study. Chlorpyrifos is not likely to be carcinogenic to humans, based on the lack of evidence of carcinogenicity in studies in rats and mice and the absence of a mutagenicity concern. There was no sign of immunotoxicity in the guideline study at the highest dose tested.

In addition to the extensive body of data on cholinesterase inhibition, there is a growing body of literature with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent behavioral effects into adulthood. There are supporting concurrent changes in brain neurochemistry based on both *in vivo* and *in vitro* studies that may underlie these behavioral changes into adulthood. These behavioral effects are seen at doses that typically result in inhibition of ChE *in vivo*. Although there are several biological plausible hypotheses being investigated by researchers, the mode/mechanisms of action resulting in such effects are not known at this time.

In addition, there are three major epidemiology cohort studies evaluating pre- and post-natal pesticide (chlorpyrifos or OPs) exposure in mother-infant pairs with birth outcomes, and childhood neurobehavioral and neurodevelopment outcomes in neonates, infants, and young children. Although there are challenges in interpreting these studies in the context of human health risk assessment, there is consistency across the animal behavior and epidemiology studies, such as delays in cognitive achievement, motor control, social behavior, and intelligence measures. Because ChE inhibition provides the most sensitive dose-response data available, the Agency has focused the preliminary risk assessment on this effect.

Chlorpyrifos has been issued an order to conduct Tier 1 screening phase of the Endocrine Disruption Screening Program.

Points of Departure and FQPA Safety Factor

The focus of the 2011 preliminary risk assessment is on the cholinesterase inhibiting potential of chlorpyrifos. Consistent with this focus, EPA has evaluated the extensive database of ChE data for multiple lifestages and has selected the most sensitive studies which use ages relevant to human exposure. The toxicological points of departure (PoDs) are based on the results of benchmark dose (BMD) analyses where appropriate, and weight of the evidence (WOE) consideration of all reliable data. There are no residual uncertainties in the exposure database. The dietary risk assessment is conservative and is not expected to underestimate dietary exposure to chlorpyrifos and chlorpyrifos oxon.

Similar to risk assessments conducted for other ChE-inhibiting pesticides, juvenile pups aged PND11 provide the sensitive lifestage and endpoint (RBC ChE inhibition) for the acute dietary PoDs of both chlorpyrifos and the oxon. The chronic dietary PoD for chlorpyrifos is based on RBC ChE inhibition from a repeated dosing study in pregnant rats (developmental neurotoxicity, DNT). The cPOD for chlorpyrifos oxon is based on 11 day repeated exposures in adult female rats (CCA study), which is protective of effects in juvenile pups. The acute and dietary PoDs for both chlorpyrifos and the oxon were derived from benchmark dose analyses.

For the dermal route (all durations) the PoD is based on RBC and plasma ChE inhibition in adult rats (NOAEL =5 mg/kg/day). For acute inhalation the PoD is based on lung ChE inhibition in rats. A NOAEL was not identified. For repeated inhalation, the PoD is based on RBC and plasma ChE inhibition (NOAEL = 287 ug/m^3 or 20 ppb from 2 inhalation studies in rats).

A 1x FQPA safety factor (SF) is being proposed for this preliminary assessment for acute and chronic oral exposure for chlorpyrifos since the PoDs are selected from sensitive endpoints (RBC ChE inhibition) in sensitive lifestages/sexes (juveniles and/or pregnant rats). A 1X FQPA SF is also proposed for all dermal durations and repeated inhalation chlorpyrifos exposures. For acute inhalation exposure, a 10X FQPA database uncertainty factor (UF) is applied to account for LOAEL to NOAEL extrapolation (a NOAEL was not identified in the acute inhalation study). A 1X FQPA SF is also proposed for acute and chronic oral exposure for chlorpyrifos oxon because the PODs are based on the most sensitive age group in the CCA study.

Due to the preliminary nature of this assessment the Agency is presenting assessments reflecting both the retention of the 10X FQPA Safety Factor as in the June 2000 chlorpyrifos risk assessment (USEPA 2000) which was largely based on adult animal data, and a preliminary proposal to reduce the FQPA SF to 1X based on more recently available ChE toxicity studies and analyses. Given the focus of this preliminary assessment on ChE inhibition, the Agency believes the ChE data support reduction of the FQPA SF to 1x for most exposure scenarios. EPA is conducting ongoing analyses of newly published literature studies on a variety of challenging scientific issues such as response relationships among different endpoints at lower exposures, animal to human extrapolation, lifestage dependent toxicities, evaluation of the non-cholinergic effects, inter-individual variation, and interpretation of epidemiology studies in the context of the entire database for assessing human health risk to chlorpyrifos. EPA will continue to evaluate all the data/studies to determine the most appropriate FQPA SF in the revised risk assessment and to determine if the new PoDs based on ChE inhibition are adequately protective of neurodevelopmental effects. This final determination will also consider the 2008 SAP comments and the public comments received on this preliminary risk assessment.

Total Uncertainty Factors for Preliminary Assessment:

A total uncertainty factor of 100X was applied to the chlorpyrifos endpoints selected for the acute and chronic dietary, and incidental oral exposures [10X for interspecies extrapolation, 10X for intraspecies variation and a proposed 1X FQPA factor based on a sensitive lifestage and endpoint selected]. Similarly, a total uncertainty factor of 100X was applied to the chlorpyrifos oxon endpoints selected for the acute and chronic dietary exposures to the oxon.

For dermal exposures a total uncertainty factor of 100X was applied [10X for interspecies extrapolation, 10X for intraspecies variation and a proposed 1X FQPA factor].

For acute inhalation exposures, a total uncertainty factor of 300X was applied [3X for interspecies extrapolation, 10X for intraspecies variation and a 10X FQPA database uncertainty factor (for extrapolation from a LOAEL to a NOAEL). The interspecies extrapolation is reduced from 10X to 3X because the RfC methodology for inhalation is used to determine an HEC (human equivalent concentration) and takes into consideration the pharmacokinetic differences between animals and humans.

For short-term and intermediate- term inhalation exposures a total uncertainty factor of 30X was applied [3X for interspecies extrapolation (reduced from 10X because RfC/HEC methodology

used), 10X for intraspecies variation and a 1X FQPA. The repeated inhalation PoDs are considered protective of sensitive lifestages (LOAEL is based on DNT study with pregnant rats).

Exposure/Risk Assessment and Risk Characterization

Dietary

Highly refined acute and chronic dietary (food and water) exposure and risk assessments were conducted for chlorpyrifos. USDA Pesticide Data Program (PDP) monitoring data and percent crop treated estimates were used for most foods. Processing factors from studies were incorporated when available.

The residues of concern for chlorpyrifos in food are for the parent only. Residues of concern in water include both parent chlorpyrifos and chlorpyrifos oxon, a known degradation product of chlorpyrifos. There are limited environmental fate data available for the oxon. The maximum amount of chlorpyrifos transformation to chlorpyrifos oxon (i.e.100%) was used as a conservative assumption based on empirical data that indicate chlorpyrifos quantitatively oxidizes to form chlorpyrifos oxon in a short period of time during water purification and minimal degradation of chlorpyrifos oxon is expected prior to consumption of the treated drinking water. It is possible that some drinking water treatment procedures such as granular activated carbon filtration and water softening may reduce the amount of chlorpyrifos oxon in drinking water; however, it is unlikely that these treatment processes significantly reduce the amount of chlorpyrifos oxon in drinking water. In addition, these treatment methods are not typical practices across the country for surface water. For these reasons, chlorpyrifos oxon is the residue of concern for drinking water.

Environmental Fate and Effects Division (EFED) has provided chlorpyrifos oxon estimated drinking water concentrations (EDWCs) from PRZM-EXAMS modeling for chlorpyrifos use on grapes, corn/soybean and sugar beets in order to provide a range of possible EDWCs representing the many registered chlorpyrifos uses. In general, these grape, corn/soybean and sugar beet uses represent a broad range of higher end, middle, and lower end EDWCs, respectively, modeled for all chlorpyrifos uses. For each of these three crops, the Agency modeled both an average typical application rate, and a maximum application rate. These particular uses were selected as representative crops for this preliminary drinking water assessment because there is a large amount of chlorpyrifos applied to these crops per year, a large portion of these crops are treated with chlorpyrifos, and/or the use locations are distributed throughout the United States.

The EFED drinking water assessment also takes into account non-targeted water monitoring data from the USGS National Water-Quality Assessment Program (NAWQA), USEPA/USGS Pilot Reservoir Monitoring Program, and USDA Pesticide Data Program (PDP) and the California Department of Pesticide Regulation (CDPR). The reported monitoring data concentrations are less than the estimated concentrations derived from modeling recommended for use in the risk assessment. This result is attributed to 1) water monitoring sampling programs do not specifically target chlorpyrifos use areas and may not represent high use areas; therefore, peak concentrations of chlorpyrifos and chlorpyrifos oxon may not be detected, 2) sampling

frequencies in high chlorpyrifos use areas are not designed to capture peak concentrations and 3) there are limited sampling data available for some areas in the United States. Because currently available monitoring data likely underestimates chlorpyrifos and chlorpyrifos oxon concentrations, monitoring data is not an appropriate estimation of the potential exposure resulting from chlorpyrifos use and are not used in this preliminary assessment. (See R. Bohaty, June 2011, D368388 and D389480, *Preliminary Registration Review Chlorpyrifos Drinking Water Assessment* for the complete drinking water characterization.)

For food alone, the preliminary acute dietary risk estimates for all populations assessed were below the level of concern. The most highly exposed subpopulation were children (1-2 years) at 9.0% aPAD.

For water alone (using the chlorpyrifos oxon PoD), the preliminary acute risk estimates using the lower end representative water scenario (sugar beet) were below the level of concern for all populations assessed at the maximum application rate except for infants at 210% aPAD. At the average typical rates for sugar beets, exposures were also of concern for infants (340% aPAD), and children (130-140% cPAD). Using the mid-range representative scenario (corn) the acute risk estimates for all populations assessed were above the level of concern for the maximum application rates; the risk estimate for the most highly exposed subpopulation, infants, was 770% aPAD. However, for typical application rates, the risk estimates were much lower (<120% aPAD) for all populations assessed. Using the higher end representative water scenario (grapes) the acute risk estimates were below the level of concern for all populations assessed at the typical application rates (<59% aPAD for infants), but were above the level of concern at the maximum application rates assessed (2700% aPAD for infants).

The preliminary chronic dietary risk estimates (food alone) for all populations assessed were below the level of concern. The most highly exposed group were children (1-2 years) at 8.4% cPAD [excluding food handling establishment (FHE) uses] and children (1-2 years) at 11% cPAD (including the FHE uses).

For water alone (using the chlorpyrifos oxon PoD), the preliminary chronic risk estimates span a large range, depending on the representative crop and application rate assessed. Using the lower end representative water scenario (sugar beets), risks were below the level of concern for all populations assessed based on the maximum application rates (<69% cPAD) however there were some risks of concern for typical rates assessed for infants and children (110-270% cPAD). Drinking water risk estimates for the mid-range and high end representative water scenarios (corn and grapes), were below the level of concern at the typical application rates (<49% cPAD) for the highest exposed subpopulation, infants (<1 yr), but exceeded the level of concern at the maximum application rates (ranged from 280-890% cPAD) for infants (<1 yr).

Comparison of the Chlorpyrifos Dietary Assessment (June 2000 Assessment and the 2011 Preliminary Assessment)

The acute and chronic PoDs and resulting dietary risk estimates (for the most highly exposed subpopulations only: young children and/or infants) are compared for the June 2000 chlorpyrifos risk assessment and for the current 2011 preliminary assessment.

In 2000 the acute and chronic dietary PoDs were based on NOAELs (plasma and/or RBC ChEI) from oral studies using adult laboratory animals (including pregnant females). The same PoD, based on toxicity of parent chlorpyrifos, was selected for both food and water. A 10x FQPA factor was retained.

For the 2011 preliminary assessment, the acute and chronic PoDs for *food* exposures were based on the toxicity of parent chlorpyrifos (BMDs for RBC ChEI) to juvenile and pregnant animals, respectively. The acute and chronic PoDs for *water* exposures were based on the toxicity of the chlorpyrifos oxon (BMDs for RBC ChEI) from studies where juvenile and adult animals were directly dosed with the oxon. A 1x FQPA factor is proposed.

The acute dietary (food only) risk estimates for the most highly exposed subpopulation were 82% of the aPAD (2000) and 9% of the aPAD (2011).

In 2000 the acute EDWC was not included in the dietary analysis (water residues not incorporated directly into DEEM analysis) and a % aPAD result was not calculated. Instead a Drinking Water Level of Concern (DWLOC) method was used. An estimated ≤18% aPAD value for 2000 water was estimated herein for comparison purposes only and reflects the exposure amount allowed for water in the 'risk cup' after food exposures are subtracted. In the 2011 preliminary water assessment, a range of representative scenarios was assessed (higher end, mid-range, and lower end). The resulting acute drinking water risk estimates (for infants) ranged from 59% to 2700% aPAD, depending on the crop and application rate.

The chronic dietary (food only) risk estimates for the most highly exposed subpopulation were 51% of the cPAD (2000) and 11% of the cPAD (2011).

As in the 2000 acute water assessment, the 2000 chronic water assessment used a DWLOC approach. A ≤ 49% cPAD value was estimated for 2000 water. In the 2011 preliminary water assessment, a range of representative scenarios was assessed (higher end, mid-range, and lower end). The resulting chronic drinking water risk estimates (for infants) ranged from 26% to 890% cPAD, depending on the crop and application rate.

It is important to note that, aside from differences in the PoDs and FQPA factors, there have been changes in the dietary input assumptions since 2000. For example, updated food monitoring data and percent crop treated data were used in the 2011 preliminary assessment. For water, in 2000 EDWCs were based on parent chlorpyrifos and were derived from the SCI-GROW model for groundwater and monitoring data for surface water. It is now believed that the existing water monitoring data are not representative of the potential exposure in drinking water and is not recommended for use in quantitative risk assessment. Groundwater EDWCs are expected to be low relative to surface water based on environmental fate characteristics of chlorpyrifos. Therefore, the SCI-GROW modeling results used in 2000 likely underestimate the potential exposure. The 2011 preliminary risk assessment has used a range of surface water EDWCs derived using PRZM-EXAMS modeling. In 2000 the residue of concern in drinking water was assumed to be parent chlorpyrifos. Empirical data indicate the rapid conversion of chlorpyrifos to chlorpyrifos oxon during typical drinking water treatment; therefore, this preliminary assessment

considers the oxon as the residue of concern in treated drinking water and assumes 100% conversion of chlorpyrifos to oxon. The chlorpyrifos oxon is more toxic than parent chlorpyrifos.

Residential

To date, all homeowner use product registrations have been cancelled, except for roach bait station products, which are not expected to result in residential exposures. Also, applications of chlorpyrifos can be made professionally (not by homeowners) to ant mounds, but residential post-application contact is not anticipated from this use. Additionally, residential/recreational uses remain for aerial and ground-based fogger adult mosquitocide applications and for golf course turf applications, which could result in residential exposures.

Of the residential uses, only the roach bait products can be applied by a homeowner in a residential setting; however, a quantitative exposure/risk assessment for application of the roach bait products was not conducted because HED expects exposure to be negligible. With roach bait stations the active ingredient is completely contained within the bait station. Post-application homeowner exposure from residential ant mound treatment (applied by professionals only) was not quantitatively assessed because contact with the mound is not anticipated. Only residential exposures anticipated from the chlorpyrifos mosquitocide use and golf course use are quantitatively assessed. In addition, a residential bystander exposure has been quantitatively assessed which considers exposure from field volatilization of applied chlorpyrifos.

Estimated short-term adult and child dermal exposures, as well as child incidental oral exposure, to turf following either aerial or ground mosquito treatments do not exceed the level of concern (i.e. calculated Margins of Exposure, or MOEs, are ≥ 100). Combined child exposure estimates (dermal and incidental oral) to turf following *aerial* mosquito treatment result in risk estimates of concern; however, combined risk estimates following *ground* treatment are not of concern. Acute adult and child inhalation (spray drift) exposure following *aerial* mosquito treatment results in risk estimates that are not of concern (i.e. MOEs are ≥ 30), but risk estimates are of concern following *ground* treatment. Inhalation exposure from ground based ULV treatment was assessed by assuming that the entire active ingredient applied to a 1 acre area is airborne and available to be inhaled by a child or adult.

Adult dermal exposure risk estimates from golfing do not exceed the level of concern (i.e. MOEs are ≥ 100) using any of the transferable residue (TTR) region-specific data for the emulsifiable concentrate formulation at the 0.25 and 1.0 lb ai/A application rates.

The Agency has developed a preliminary bystander inhalation exposure assessment for chlorpyrifos using currently available inhalation toxicity and chlorpyrifos air monitoring data. EPA has assessed residential bystander exposure from field volatilization of applied chlorpyrifos based on the available *ambient* and *application site* air monitoring data. Of the 24 acute *ambient* air concentrations assessed, 4 result in risk estimates exceeding the level of concern (i.e. MOEs are < 300). No short-/intermediate-term *ambient* data assessed result in risk estimates of concern (i.e. MOEs are > 30). Of the 5 acute *application site* air concentrations assessed, 3 resulted in a risk estimate of concern (i.e. MOEs are < 300). Of the 5 short- and intermediate-term

application site air concentrations assessed, 4 resulted in risk estimates of concern (i.e. MOEs are < 30).

The bystander exposure assessment is considered preliminary. Some of the limitations identified include the assumption that an individual is exposed to a constant chlorpyrifos concentration in a stationary outdoors location for 24 hours. As part of the December 2009 SAP, the Agency presented their analysis of several models that could be used as screening tools to predict the air concentration and volatilization flux based on intrinsic properties and transport behaviors of pesticides. The Agency is currently in the process of evaluating the SAP's comments. As appropriate, the Agency may revise the modeling approach presented to the SAP may revisit the residential bystander exposure and risk assessment.

Aggregate

A quantitative aggregate (food, water and residential exposures combined) assessment was not performed for this preliminary chlorpyrifos assessment. The preliminary risk estimates for water alone exceed the level of concern and are the primary driver in this assessment. Combining food and/or residential exposures with the water exposures would not be expected to have a significant impact on the resulting risk estimates for water alone. A quantitative aggregate assessment for food, water, and residential exposures will be considered during the final chlorpyrifos risk assessment.

With regard to potential aggregate exposures for workers, the Agency is carefully considering a number of complex science issues, and extensive public comments received on OPP's proposed policy –*Revised Risk Assessment Methods for Workers, Children of Workers in Agricultural Fields and Pesticides with No Food Uses*" (EPA-HQ-OPP-2009-0889-0002).

Occupational

Short- and intermediate-term inhalation and dermal exposure and risk estimates were calculated for occupational handlers of chlorpyrifos for a variety of exposure scenarios at differing levels of personal protection (long-term exposures not expected). The assessments used surrogate data and non-chemical specific exposure studies. In total, 305 exposure scenarios which consist of unique combinations of product formulation, crop or target, application rate, and area treated were assessed.

Of the 305 exposure scenarios assessed 134 had risk estimates that did not exceed the level of concern at some level of personal protection (i.e. calculated Aggregate Risk Estimates, or ARIs, are > 1). Ninety-one (91) exposure scenarios had risk estimates not of concern when engineering controls were considered. The remaining 80 scenarios resulted in risk estimates of concern (i.e. ARIs are < 1) at all levels of personal protection and engineering controls considered.

In an effort to characterize occupational handler risk estimates calculated using both surrogate data and chemical specific biomonitoring (passive dosimetry) data, HED has presented a comparative analysis of these for applicable scenarios. Of the 4 exposure scenarios compared, 3 (mixing/loading liquids for airblast application, airblast applications, and groundboom

applications) resulted in greater risk estimates using biomonitoring data than those estimated using surrogate data (i.e., the estimated MOEs are lower). The analysis of the exposure scenario mixing/loading liquids for aerial application resulted in reduced risk estimates using biomonitoring compared to surrogate data. Because a number of issues were identified which limit the utility of the available biomonitoring data, HED has determined that these data are best suited for characterization of the estimates calculated for representative exposure scenarios using the surrogate data.

Short- and intermediate-term exposure and risk estimates were calculated for occupational handlers performing seed treatment activities in commercial settings and for occupational secondary handlers from planting chlorpyrifos-treated seeds. No chemical-specific handler exposure data were submitted in support of this use pattern.

The majority, 61 of 64, occupational handler seed treatment exposure scenarios assessed (combined dermal and inhalation) resulted in risk estimates which were not of concern (i.e. ARIs are > 1) at some level of personal protection. The remaining 3 exposure scenarios resulted in an ARI < 1 at all level of personal protection considered and, therefore, are of concern. All seed planter (secondary handler) combined short- and intermediate-term dermal and inhalation exposure scenarios assessed resulted in an ARI > 1 at some level of personal protection and, therefore, do not present risk estimates of concern.

EPA has assessed short- and intermediate term occupational post-application dermal exposure and risk for any crops which reentry into an area previously treated with chlorpyrifos is anticipated. The assessment was completed using 7 chemical-specific registrant submitted dislodgeable foliar residue (DFR) studies.

The MOEs estimated for liquid spray and granular formulation reentry are not of concern (i.e., an MOE ≥ 100) in the range of 0 to 4 days for lower to medium exposure activities and 0 to 8 days for high exposure activities, with the greater majority falling between 0 to 4 days when all exposure activities are considered. HED also estimated the MOEs for reentry into microencapsulated and total release fogger formulation treated greenhouses. These estimates range from 0 to > 35 days after treatment (the completion of the monitoring period) for all exposure activities considered.

A quantitative occupational post-application inhalation exposure assessment was not performed for chlorpyrifos. An inhalation exposure assessment was performed for occupational/commercial handlers and handlers are expected to have greater exposures than workers involved in post-application activities. The handler assessment is currently considered a worst-case assessment for post-application exposure.

Occupational/Residential Exposure to Chlorpyrifos Oxon

The Agency has considered the potential for occupational and residential exposure to chlorpyrifos oxon. Workers re-entering an environment previously treated with chlorpyrifos (occupational post-application) and the general population residing near chlorpyrifos application sites (bystanders) could potentially be exposed to the oxon as chlorpyrifos is degraded in the

environment. Dermal exposure to the oxon could occur through contact with chlorpyrifos treated surfaces and inhalation exposure through airborne oxon. However, the likelihood of exposure to the oxon is slight due to its rapid deactivation to TCP (3,5,6-trichloro-2-pyridinol). In an effort to further explore the potential for oxon exposure, EPA has researched and reviewed all available information sources. Based upon this review, EPA intends to require additional studies to address uncertainties regarding the formation of chlorpyrifos oxon in the air post-application and its formation and decay in greenhouses.

Comparison of the Chlorpyrifos Occupational Assessment (June 2000 Assessment and the 2011 Preliminary Assessment)

For comparison purpose, a range of resulting occupational handler risk estimates (MOEs) are presented for both the current preliminary (2011) chlorpyrifos assessment and the June 2000 chlorpyrifos assessment. The range represents a low, medium, and high exposure scenario. Also presented is a range of personal protection (single layer/gloves, double layer/gloves, and engineering controls). [See Table 28(dermal) and Table 29 (inhalation).]

The dermal handler risk estimates remain virtually unchanged between the 2000 and 2011 assessments since the dermal PoD is the same (NOAEL of 5 mg/kg/day from a dermal study). The 2008 SAP concurred with the selection of this PoD for assessing dermal scenarios.

The inhalation PoD in 2000 was 0.1 mg/kg/day (LOAEL/NOAEL based on 90 day inhalation studies and DNT). That same PoD is used in the current assessment except that it has been converted to an HEC (human equivalent concentration). This resulted in the reduction of the default database uncertainty factor for interspecies extrapolation from a 10x to a 3x. Thus the level of concern MOE for this assessment is 30 (compared to 100 in 2000). In addition the NOAEL was corrected to account for an 8 hour workday because worker exposure is expected to occur during the course of an average workweek (8 hours/day and 5 days/week; animals were exposed 6 hours a day in the study). The inhalation handler risk estimates have changed since the 2000 assessment. This can be mainly attributed to the use of the HEC in the preliminary assessment.

Note that the actual dermal and inhalation MOEs presented in the 2000 assessment may differ somewhat than those presented here since some of the exposure assumptions used today may vary due to refinements made since 2000. The 2011 exposure assumptions were compared to the 2000 PoD for illustrative purposes only.

2.0 HED Recommendations

2.1 Data Deficiencies

Toxicology

870.6300: Developmental Neurotoxicity (MRID 44556901). While the offspring NOAEL and LOAEL have not yet been identified for this developmental neurotoxicity study, it is recognized

that the study was well-conducted according to Agency guideline §83-6, and under GLP regulations. Remaining questions can be resolved with additional information and statistical analysis, but there are no outstanding concerns regarding the quality of the animal data. The study is currently classified as –guideline-unacceptable, but upgradeable". The study may be upgraded to –acceptable" pending submission and review of additional morphometric data for PND 66 low-dose females (parietal cortex and hippocampus measurements) (S. Makris, 3/3/00, TXR. 0014014, D254907).

Residue Chemistry

860.1500: Magnitude of the residue studies with lemon after application of Lorsban 4E and 75% WDG formulations separately to reassess the tolerance for the citrus fruit crop group; Magnitude of the residue studies to establish a tolerance for wheat hay.

860.1520: Processing studies for: cotton meal, hulls and refined oil and for soybean meal, hulls and refined oil.

Labels: Revise the corn and cotton use restrictions in the chlorpyrifos labels to eliminate feeding restrictions in treated areas. Maintain only dormant/delay dormant and trunk spray applications for tart cherries in the label of the 75% WDG end use product.

Tolerances:

The following tolerances for chlorpyrifos are necessary to address residues found in field trails:

Cotton, gin byproducts	15 ppm
Grain, aspirated fractions	22 ppm
Corn milled byproducts	0.1 ppm
Wheat milled byproducts	1.5 ppm

Revocation of the chlorpyrifos tolerance for lettuce (no registered uses; revocation pending).

Modification of the tolerance expression for chlorpyrifos in the 40 CFR 180.342 is needed to comply with the Interim Guidance for Writing Tolerance Expressions.

Occupational/Residential Exposure

The Agency intends to require additional data to address uncertainties regarding the formation of chlorpyrifos oxon in the air post-application and its formation and decay in greenhouses. In addition, several data gaps were identified in the Registration Eligibility Decision (RED) for the occupational and residential assessment of chlorpyrifos (finalized 7/31/06; IRED issued 2/2002). The only one of these requirements that has not been satisfied is the requirement for a study confirming the area treated for sod farm chlorpyrifos treatment. This requirement remains outstanding.

2.2 Tolerance Considerations

2.2.1 Enforcement Analytical Method

The methods in the PAM Volume II are adequate to analyze the residue of concern for tolerance enforcement purposes, chlorpyrifos only. The limit of detection of these methods is adequate to cover the lowest tolerance level included in the 40 CFR 180.342 for detection of chlorpyrifos only, 0.01 ppm. In addition, chlorpyrifos is completely recovered using FDA multiresidue protocols D and E (nonfatty matrices) and partially recovered using multiresidue method protocol E (fatty matrices).

2.2.2 International Harmonization

Current US permanent tolerances for chlorpyrifos are listed in 40 CFR§180.342 and are summarized in the residue chemistry chapter and in Appendix C of this document. The Codex Alimentarius Commission and Canada have established Maximum Residue Limits (MRLs) for chlorpyrifos. Mexico adopts US tolerances and/or Codex MRLs for its export purposes. US tolerances and Codex MRLs are based on the analysis of residues of chlorpyrifos. Canada MRLs are for chlorpyrifos for some commodities and for both parent chlorpyrifos and its metabolite TCP (3,5,6-trichloro-2-pyridinol; not a US residue of concern) for other commodities.

With the exception of apple commodities, harmonization with the Canada MRLs is not possible as the Canadian residue definition is for the combined residues of chlorpyrifos and TCP (in the US TCP is not considered a residue of concern for chlorpyrifos risk assessment or tolerance enforcement). Harmonization between the USA tolerances and Codex MRLs is only possible for corn, field, grain; cranberry; egg; sorghum, grain, grain; sorghum, grain, stover; and wheat, grain. In addition, two commodities of the Leafy Vegetable (CG 5) can be harmonized with the Codex, head cabbage, and Chinese cabbage (type petsai). A summary of the US and international tolerances and MRLs is included in Appendix C of this document.

2.2.3 Recommended/Reassessed Tolerances

The following tolerances would need to be established to address residues found on the following commodities in new crop field trial data received as part of the chlorpyrifos 2003 DCI:

Table 1 Recommended Tolerances for Chlorpyrifos

Commodity	Established Tolerance (ppm)	Recommended Tolerance (ppm)	Comments *Correct Commodity Definition*
Aspirated grain fractions	NA	22	
Cotton, gin by-products	NA	15	

On 5/27/09 HED established interim guidance on writing tolerance expressions for enforcement purposes. In order to add clarity to the language used to establish the coverage of the tolerance expression and measurement of the level of the residue in the RACs the text in the 40 CFR § 180.342 should read: ~~(~~a) General. (1) Tolerances are established for residues of chlorpyrifos, including its metabolites and degradates, in or on the commodities in the table below.

Compliance with the tolerance levels specified below is to be determined by measuring only chlorpyrifos." The current tolerance expression reads –chlorpyrifos *per se* (*O,O* -diethyl *O* -(3,5,6-trichloro-2-pyridyl) phosphorothioate".

2.3 Recommendations from Residue Reviews

The following recommendations were made in I. Negrón-Encarnación, 5/24/11, D388164, *Chlorpyrifos. Registration Review Action for Chlorpyrifos. Summary of Analytical Chemistry and Residue Data.*

Revise the corn and cotton use restrictions in the chlorpyrifos labels to eliminate feeding restrictions in treated areas.

The following tolerances for cotton gin byproducts and aspirated grain fractions are necessary to address residues found in field trails:

Cotton, gin byproducts ..... 15 ppm
Grain, aspirated fractions .....22 ppm

Maintain only dormant/delay dormant and trunk spray applications for tart cherries in the label of the 75% WDG end use product.

Revocation of the tolerance for lettuce is in process as uses of chlorpyrifos in this crop are not included in the label for the registered products.

Magnitude of the residue studies are needed with lemon after application of Lorsban 4E and 75% WDG formulations separately to reassess the tolerance for the citrus fruit crop group.

Magnitude of the residue studies are needed to establish a tolerance for wheat hay.

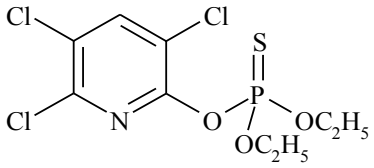
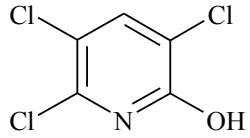
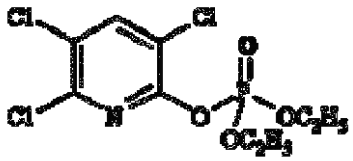
Processing studies are needed for:
Cotton meal, hulls and refined oil
Soybean meal, hulls and refined oil

Tolerances are needed to address residues of chlorpyrifos on:
Corn milled byproducts as 0.1 ppm
Wheat milled byproducts as 1.5 ppm

Modification of the tolerance expression for chlorpyrifos in the 40 CFR 180.342 is needed to comply with the Interim Guidance for Writing Tolerance Expressions.

3.0 Introduction

3.1 Chemical Identity

Table 2 Chlorpyrifos Degradate/ Residues of Concern Nomenclature.	
Chlorpyrifos	
IUPAC name	*O,O*-diethyl *O*-3,5,6-trichloro-2-pyridyl phosphorothioate
CAS name	*O,O*-diethyl *O*-(3,5,6-trichloro-2-pyridinyl) phosphorothioate
CAS registry number	2921-88-2
End-use product (EP)	Lorsban 75% WDG and Lorsban 50% WP
TCP Metabolite/Degradate (Residue of Concern for Canada) IUPAC Name 3,5,6 Trichloro-2-pyridinol	
Oxon Metabolite/Degradate Common Name Chlorpyrifos Oxon IUPAC Name O,O-diethyl. O-3,5,6-trichloro-2-pyridyl	

3.2 Physical/Chemical Characteristics

Technical chlorpyrifos is a white crystalline solid. Chlorpyrifos is stable in neutral and acidic aqueous solutions; however, stability decreases with increasing pH. Chlorpyrifos is practically insoluble in water, but is soluble in most organic solvents (i.e., acetone, xylene and methylene chloride). Chlorpyrifos is moderately volatile based on its vapor pressure of 1.87×10^{-5} mmHg at 25°C.

In the environment, hydrolysis is not expected to play a significant role in chlorpyrifos dissipation; however, under alkaline conditions laboratory studies show chlorpyrifos is susceptible to hydrolysis. Laboratory studies suggest that volatilization and photodegradation are not likely to play a significant role in the dissipation of chlorpyrifos in the environment. Nonetheless, chlorpyrifos has been detected in air samples so volatilization may play more of a role in dissipation than laboratory studies indicate. The major route of dissipation appears to be aerobic and anaerobic metabolism. Based on available data, chlorpyrifos degrades slowly in soil under both aerobic and anaerobic conditions. Degradation begins with cleavage of the phosphorus ester bond to yield 3,5,6-trichloro-2-pyridinol (TCP). Field dissipation studies show that chlorpyrifos is moderately persistent under field conditions—dissipation half-life less than 60 days. Chlorpyrifos is only slightly soluble in water but once it reaches aquatic environments

the Log K_{ow} (4.7) indicates that chlorpyrifos may bioaccumulate in fish and other aquatic organisms. A fish bioaccumulation study shows that chlorpyrifos is absorbed by fish; however, it rapidly depurates when exposure ceases.

Oxidation of chlorpyrifos to chlorpyrifos oxon could potentially occur through photolysis, aerobic metabolism, and chlorination as well as other oxidative processes. Chlorpyrifos oxon is expected to have similar fate characteristics as chlorpyrifos except chlorpyrifos oxon is more soluble in water and undergoes hydrolysis faster. The hydrolysis half-life of chlorpyrifos oxon is significantly shorter than that observed for chlorpyrifos. Chlorpyrifos oxon hydrolyses to form TCP. For chlorpyrifos, water purification (chlorination) has been shown to be a major route of chlorpyrifos oxon formation.

3.3 Anticipated Exposure Pathways

Humans may be exposed to chlorpyrifos in food and water since applications may be made directly to growing crops (food and feedstuffs) which could also result in chlorpyrifos reaching surface and ground water sources of drinking water. Registered uses that may result in residential (non occupational) exposures include aerial and ground-based fogger adult mosquitocide applications and golf course turf applications. There is also a potential for residential bystander exposure from field volatilization of applied chlorpyrifos. In occupational settings, exposure may occur while handling the pesticide prior to application, as well as during application. There is also a potential for post-application exposure for workers re-entering treated fields.

3.4 Consideration of Environmental Justice

Potential areas of environmental justice concerns, to the extent possible, were considered in this human health risk assessment, in accordance with U.S. Executive Order 12898, "Federal Actions to Address Environmental Justice in Minority Populations and Low-Income Populations," (http://www.eh.doe.gov/oepa/guidance/justice/eo12898.pdf. As a part of every pesticide risk assessment, OPP considers a large variety of consumer subgroups according to well-established procedures. In line with OPP policy, HED estimates risks to population subgroups from pesticide exposures that are based on patterns of that subgroup's food and water consumption, and activities in and around the home that involve pesticide use in a residential setting. Extensive data on food consumption patterns are compiled by the USDA under the Continuing Survey of Food Intake by Individuals (CSFII) and are used in pesticide risk assessments for all registered food uses of a pesticide. These data are analyzed and categorized by subgroups based on age, season of the year, ethnic group, and region of the country. Additionally, OPP is able to assess dietary exposure to smaller, specialized subgroups and exposure assessments are performed when conditions or circumstances warrant. Whenever appropriate, non-dietary exposures based on home use of pesticide products and associated risks for adult applicators and for toddlers, youths, and adults entering or playing on treated areas post-application are evaluated. In addition to the aforementioned exposure settings and population subgroups, the current chlorpyrifos risk assessment considered exposures to bystanders as a result of field volatilization of applied chlorpyrifos.

4.0 Hazard Characterization and Dose-Response Assessment

4.1 Toxicology Studies Available for Analysis

The toxicological database for chlorpyrifos is extensive and is adequate to support the registration review. Since the 2002 IRED/2006 RED, and in addition to many studies in the scientific literature, three new studies have been submitted to OPP: a special acute inhalation study (MRID 48139303), a comparative cholinesterase assay (MRID 48139301), and an immunotoxicity study (MRID 48139304). These submitted studies have been reviewed and found to be acceptable to support the chlorpyrifos risk assessment. The toxicity profiles and executive summaries of all submitted studies are listed in Appendix A.

The database spans multiple routes of exposure, animal species, and lifestages and consists of acute toxicity, subchronic oral, subchronic inhalation, immunotoxicity, developmental toxicity, multi generation reproduction, chronic feeding/carcinogenicity, dermal toxicity, metabolism, pharmacokinetic, acute and subchronic neurotoxicity, and developmental neurotoxicity studies. The genetic toxicity/mutagenicity database has been evaluated. In addition, special studies have been submitted including an acute neurotoxic esterase rat study, cognitive rat study, comparative cholinesterase assay where PND11 pups and adults were assessed (for both parent chlorpyrifos and its oxon metabolite) and an acute inhalation study.

In addition to the above submitted chlorpyrifos studies there are numerous literature studies available on various aspects of chlorpyrifos including inhibition of cholinesterase, neurological effects in animals and humans following gestational and post-natal exposures, pharmacokinetics, mechanism of action, as well as studies with adult human volunteers for ChE inhibition. Many of these studies were discussed at the 2008 SAP meeting and details are provided on the Science Advisory Panel website (http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm). EPA plans to finalize the science documents reviewed by the SAP in the upcoming months. However, the advice received by the Agency at the 2008 SAP meeting has been used to inform the selection of toxicological points of departure for use in this preliminary chlorpyrifos risk assessment.

4.2 Absorption, Distribution, Metabolism, & Elimination (ADME)

The metabolism and toxicokinetics (TK) of chlorpyrifos have been extensively studied in animals and humans as well as in vitro systems. Overall, rats and humans show similar patterns of metabolism for chlorpyrifos in adults.

Chlorpyrifos undergoes metabolic transformations mainly by the liver microsomal enzymes. Although, chlorpyrifos is lipophilic, its extensive metabolism into water soluble metabolites does not lead to accumulation of the parent material or its metabolites in the body tissues. The initial metabolic attack on the chlorpyrifos is its desulfuration, resulting in its bioactivation to the more toxic and potent ChE inhibitor, the oxon form. However, the oxon is unstable and is rapidly deactivated through hydrolytic cleavage by a process called dearylation releasing the 3,5,6-trichloro-2-pyridinol (TCP). Simultaneously along the desulfuration process, dearylation will be acting on both the parent chlorpyrifos as well as on the oxon metabolite leading to the release of

TCP. TCP is further conjugated to form glycine or glucuronide conjugates and eliminated into the urine. TCP is the major excreted metabolite and used as the major biomarker in pharmacokinetic, biomonitoring, and epidemiology studies.

There are several enzymes that play a role in the metabolism and toxicity of chlorpyrifos. In addition to inhibition of ChE, the oxon also binds stoichometrically to butyrlcholinesterse (BuChE; abundant in blood and other tissues). In this regard BuChE is viewed as a scavenger of the oxon formed and may prevent it from entering the brain or peripheral targets for inhibition of ChE. The cytochrome P450 family of microsomal enzymes (CYPs) is responsible for its metabolic activation and deactivation. The oxon also binds irreversibly to carboxylesterases. Carboxylesterases are distributed among different issues (liver, blood, brain) with highest abundance in the liver. The glutathione dependent enzymes play an important role in the secondary metabolism of chlorpyrifos producing water soluble metabolites that are readily excreted into the urine. Finally, another group of important enzymes in the detoxification of chlorpyrifos is the A-esterases; one such A-esterase is paraoxonase (i.e., PON1). These are calcium activated enzymes and are distributed in various tissues including the liver, brain and blood. These act on the oxon by hydrolyzing it before reaching its target AChE enzyme. Some have suggested that PON1 status is a determining factor in susceptibility to chlorpyrifos (Cole et al, 2005; Berkowitz et al, 2004; Wolff et al, 2007; Furlong et al, 2005; Brophy et al, 2001; Holland et al, et 2006; Chen et al, 2003).

The increased sensitivity of the young from acute exposure is likely attributed to a reduced capacity to detoxify chlorpyrifos in juvenile animals (Gagne and Brodeur, 1972; Benke and Murphy, 1975; Pope *et al*., 1991; Chambers and Carr, 1993; Padilla *et al*., 2000; 2002; Karanth and Pope, 2000). Specifically, in rats, A-esterase activity is virtually nonexistent in the fetus (Lassiter *et al*., 1998) and increases from birth reaching adult levels around PND21 (Mortensen *et al*., 1996; Li *et al*., 1997). Mortensen et al (1996) showed that the plasma level of CPOase[1] in PND21 pups was 1/11 that of adult animals. The animal data regarding the role of carboxylesterase in mediating OP toxicity are also quite extensive (e.g., Clement, 1984; Fonnum et al., 1985; Maxwell, 1992 a, b). Fetal rats possess very little carboxylesterase activity (Lassiter et al., 1998) with increasing activity seen as the postnatal rat matures, reaching adult values after puberty (50 days-of-age; Morgan et al., 1994; Moser et al., 1998; Karanth and Pope, 2000). There are, however, very little data in human tissues which could evaluate age-related maturation of carboxylesterase expression. The available data come from Pope et al (2005) and Ecobichan and Stephens (1973). Ecobichan and Stephens (1973) showed a steady increase in AChE and ChE levels of infants beginning at birth up to adult levels. Pope et al (2005) evaluated maturational expression of liver carboxylesterases in human liver tissues from infants (2–24 months) and adults (20–36 years). The authors report relatively small (and not statistically significant) differences in activities between children ages 2–24 months and adults (20–36 years). The Agency notes, however, that youngest age evaluated in the study was 2 months old and this individual had the lowest level of carboxylesterase.

There is a clear age-dependant sensitivity which diminishes as the pups mature; this pattern is likely reflective of the metabolic processes which rapidly mature in the rat pup. The SAP

[1] CPOase is A-esterase (PON1) activity specific to chlorpyrifos oxon

concurred with the Agency that juveniles are more sensitive than adults to ChE inhibition following acute exposures, but not necessarily for repeated exposures.

In 2008, the Agency solicited comments from the SAP on the use of information on PON1 to inform the intra-species extrapolation factor. The SAP panel agreed with EPA that PON1 status cannot be ruled out as a determinant of chlorpyrifos toxicity, and there appears to be a different susceptibility between fetuses and neonates compared to adults. The Panel did not support using such PON1 information alone to address population sensitivity, but instead suggested that PBPK modeling which accounts for all the metabolizing enzymes is a more supportive approach.

In the rat, chlorpyrifos is excreted primarily in the urine (84%) with lesser amounts excreted in the feces (5%) within 72 hours. The metabolism of chlorpyrifos is extensive, and no unchanged parent compound is found in the urine. The major urinary metabolites were 3,5,6-TCP (TCP) and glucuronide and sulfate conjugates of TCP. In humans (adult males) approximately 70% of chlorpyrifos is excreted in the urine as conjugated TCP within 5 days following acute oral exposure, and the dermal absorption is 1 to 3% in this study (Nolan *et al*. 1982, Accession No. 249203). The mean pharmacokinetic half-life for TCP in the urine is approximately 27 hours following both oral and dermal exposure.

There are some limited data that show that chlorpyrifos can be found in breast milk. Chlorpyrifos is lipophillic and has a Log Kow of 4.7, which would indicate a potential to accumulate in milk. Mattsson *et al*. (1998, 2000) provided data in rat milk which suggest that chlorpyrifos can reach milk at doses of 0.3 mg/kg/day. There is public literature that indicates that chlorpyrifos may be found in human breast milk in the U.S. (Casey 2005) and India (Srivastava *et al*., 2011). The degree to which the Indian data are relevant in the U.S. is unknown (and unlikely reflective of the general population exposed to chlorpyrifos in food/water).

Toxicokinetic (TK) studies from humans and rats show that chlorpyrifos and/or its metabolites may be available to the fetus, likely at levels similar to maternal tissues (Whyatt *et al*, 2003; Hunter al, 1999; Mattsson *et al*, 1998, 2000; Akhtar *et al*, 2006). In the 2008 draft issue paper, the Agency summarizes the studies which show that TK differences in young and adults play a key role in the age-dependant sensitivity with chlorpyrifos. Moreover, the 2008 document provides additional information in pregnant animals and humans which suggest that metabolic capacity to detoxify chlorpyrifos may be reduced during pregnancy, although the relevance of these changes is not known at low environmental levels. The Panel supported the Agency's conclusions on the role of lifestage ontogeny in potential sensitivity to chlorpyrifos and the potential that pregnant females may be more sensitive to chlorpyrifos than males (FIFRA SAP, 2008), Recent results of EPA's analyses (see BMD Appendix E) for rat data suggest that pregnant females are approximately 2-12 fold more sensitive than non-pregnant adult females, as shown in Table 3 below.

Table 3 Comparison of Cholinesterase Inhibition for Adult Pregnant Female and Non-Pregnant Rats

Endpoint	Response	Comments
Repeated Dose ChEI - male and female rats (Hoberman et al. 1998 a, b, MRID 44556901; Mattsson et al. 1998, MRID 44648101; Maurissen et al. 2000; Marty and Andrus (2010; DAS CCA MRID 48139301; 4807001)	Female rats, 11 days (CCA): $BMD_{10}/BMDL_{10}$: RBC ChEI: 0.45/0.35 brain ChEI: 1.03/0.95 mg/kg/day Female pregnant rats GD6-20; 15 days (DNT): $BMD_{10}/BMDL_{10}$: RBC ChEI: 0.06/0.03 mg/kg/day brain ChEI: 0.65/0.54 mg/kg/day	Pregnant female rats more sensitive than non-pregnant female rats for RBC and brain ChEI: RBC ChEI: 7.5-12 fold more sensitive Brain ChEI: 1.6-1.8 fold more sensitive

DNT= developmental neurotoxicity study
CCA= comparative cholinesterase study

4.3 Toxicological Effects

Cholinesterase (ChE) Inhibition. Chlorpyrifos, like other OPs, binds to and phosphorylates the enzyme, acetlycholinesterase (AChE), in both the central (brain) and peripheral nervous systems leading to accumulation of acetylcholine in critical neuronal junctions and, ultimately, to clinical signs of toxicity. This mode of action, in which ChE inhibition leads to neurotoxicity, has been well described (Mileson *et al*., 1998, Eaton *et al*., 2008; Gupta, 2011). In 2000, the Agency concluded for chlorpyrifos that inhibition of ChE provides the most sensitive dose response data in all of the animal species evaluated (rats, mice, rabbits dogs) and in humans, regardless of exposure duration and route of exposure. The available data indicate that humans are more sensitive than animals to ChE following both oral and dermal exposure (Nolan *et al*. 1982, USEPA 2000a). Numerous ChE studies are available in different lifestages and ages in rats, which were included in the 2000 risk assessment and/or discussed at the 2008 FIFRA SAP (http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm). These studies vary widely by the level and number of doses used, availability of time course information, and method of administration. The Agency has reviewed the studies submitted for registration as well as searched the public literature for studies in which adult animals and/or juvenile animals were exposed to chlorpyrifos. ChE inhibition is most commonly reported for the blood (plasma and RBC) and brain (whole or subsections). The chlorpyrifos database is unique since it includes evaluations of peripheral tissues such as the heart, diaphragm, or lung. In animals, significant inhibition of plasma and red blood cell (RBC) ChE occur at doses below those that cause brain ChE inhibition. Following inhalation exposure, inhibition of ChE in the lung was more sensitive than either RBC ChE or brain ChE inhibition.

With respect to considering the response of sensitive lifestages to ChE inhibition, the Agency has reviewed numerous repeated gestational exposure ChE studies for chlorpyrifos and other OPs. Overall, these gestational studies show that the fetus exhibits no more ChE inhibition than does the dam and in some studies fetus actually exhibits less inhibition (USEPA, 2008; USEPA,

2006). However, ChE data in fetuses from repeated dosing gestational studies may not accurately reflect potential fetal toxicity at a particular dose (USEPA 2008, draft chlorpyrifos hazard and dose-response characterization). The FIFRA SAP concurred with the Agency's conclusion with respect to interpreting ChE data from *in utero* exposures. As part of the scientific analysis presented at the 2008 SAP meeting, the Agency showed acute brain post-natal ChE studies ranging from PND1 to PND33. There is a clear age-dependant sensitivity which diminishes as the pups mature; this pattern is likely reflective of the metabolic processes which rapidly mature as the rat pup matures. The SAP concurred with the Agency that juveniles are more sensitive than adults to ChE inhibition following acute exposures, but not necessarily for repeated exposures.

The SAP also supported that pregnant animals and humans may be somewhat more sensitive to ChE inhibition from chlorpyrifos than non-pregnant adults based on a reduced capacity of key detoxification enzymes (e.g., paraoxonase, P450 isozymes) in modulating levels of chlorpyrifos in animal studies. It is unknown if the relatively small differences in enzyme levels is important at environmental exposure levels. As noted previously, pregnant rats were about 2-12 fold more sensitive than non pregnant rat females for ChE inhibition (See Table 3).

The Agency has recently reviewed an acute and repeat special non-guideline comparative cholinesterase (CCA) study, and an acute inhalation study. The CCA study was conducted to compare the relative toxicity of chlorpyrifos and chlorpyrifos oxon in both juvenile (PND11 pups) and adult rats based on ChE inhibition (Marty and Andrus 2010, MRID No.: 48139301 TXR No. 0055409). Both acute (single) dosing and a repeat 11-day exposure scenario were evaluated for chlorpyrifos and chlorpyrifos oxon. In the acute subpart, juvenile rats were dosed with chlorpyrifos via both gavage and milk. This study is considered high quality, and provides reliable measures of blood and brain ChE at the time of peak effect (6-8 hours post-dosing), use 4-6 doses and use a wide range of doses. The Agency notes that the timing of ChE measurement in this study (8 hrs for milk) is later than other studies that report the peak at between 3-6 hours but is supported by time course data collected as part of this study for 3-10 mg/kg dose levels [see EPA 2008, draft appendix C, Mode of action, inhibition of acetylcholinesterase (AChE)]. The Agency used these data to conduct benchmark dose (BMD) analysis. Following acute exposure, based on BMD analysis, PND11 pups were more sensitive than adults at 10% RBC ChE inhibition (BMD10 are 0.5 mg/kg/day and 1.9 mg/kg/day, respectively), and 10% brain ChE inhibition (BMD10 are 1.4 and 4.1 mg/kg/day, respectively) for chlorpyrifos. For acute chlorpyrifos-oxon exposure, pups were also more sensitive for RBC ChE inhibition than adults (BMD10s are 0.08 and 0.21 mg/kg/day for pups and adults, respectively). Pups were more sensitive to ChE inhibition following milk exposure than from gavage dosing based on BMD10s. Following 11 days of repeated dosing, PND11 pups were slightly more sensitive than adults to chlorpyrifos based on BMD10s for RBC ChE inhibition (0.45 mg/kg/day for adults vs 0.11-0.17 mg/kg/day for pups), but not for chlorpyrifos oxon. There was no inhibition of the brain ChE reported for the oxon at any dose up to 1 mg/kg for acute dosing and 0.5 mg/kg/day following repeat dosing in either pups or adults. The timing of measurement for the oxon was 4 hours post-dosing in this CCA. Other literature studies have reported the time of peak brain ChE inhibition was 1 hour post dosing (Betancourt and Carr 2004).

In a special acute inhalation study female rats were exposed by nose only to atmospheric concentrations of up to 53.9 mg/m^3 of particulate chlorpyrifos for six hours and allowed an additional 72 hours to recover (MRID No: 48139303 Hotchkiss *et al*. 2010, TXR # 0055409.). The peak inhibition for plasma and RBC ChE was at 2 hours post-dosing. Consistent and significant lung and plasma ChE inhibition were noted at the lowest concentration tested of 3.7 mg/m^3, which is a LOAEL. RBC and brain ChE inhibition were noted at ≥ 12.9 mg/m^3 and 53.9 mg/m^3, respectively, indicating they are less sensitive than lung and plasma ChE inhibition following acute inhalation exposures. It should be noted, however, that the lung may contain both butyrl and acetyl cholinesterase, which may partially explain the sensitivity of the lung ChE inhibition. No NOAEL was established. A BMD analysis was attempted but did not provide high confidence results due to the nature of the dose response data. The RBC ChE data had significant temporal variation and thus a reliable fit was not achieved. For the lung ChE data, no statistically reliable fit was obtained with exponential modeling using nonhomogeneous variance (suggested by BMD statistical results). However, a reliable fit was obtained with a homogeneous variance model. This analysis supports retention of a 10X LOAEL to NOAEL uncertainty factor for the single- day bystander inhalation risk assessment. Using the Agency's Reference concentration (RfC) methodology, a human equivalent concentration (HEC) was calculated to be 0.62 mg/m^3 based on the LOAEL of 3.7 mg/m^3.

Developmental Effects. There is a large body of literature on the effects of chlorpyrifos in the developing brain of laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent behavioral effects into adulthood. These data provide support for the susceptibility of the developing rodent brain to chlorpyrifos exposure. Many of these studies were reviewed by EPA in the 2000 risk assessment and for the 2008 SAP (USEPA 2000b, 2008). The SAP concurred with the Agency's conclusion that in rats, chlorpyrifos causes alterations in brain development in offspring of exposed mothers. Studies in the peer reviewed literature and results of the guideline developmental neurotoxicity (DNT) study are supportive of the possibility that chlorpyrifos exposure may affect brain development (e.g., altered synaptic development, alterations in DNA, RNA, and protein synthesis, inhibition of mitosis and mitotic figures, disruption of the structural architecture of the brain). Studies from multiple laboratories in two rodent species provide evidence that adults may exhibit persistent behavioral changes following perinatal exposures. Since several laboratories included a dose of 1 mg/kg/day, some comparisons in response may be made – these are summarized in Table 4 below. Chlorpyrifos studies in rats and/or mice have reported impaired cognition (spatial learning and working memory); changes in locomotor activity levels (exploration, rearing); and altered social interaction (aggression, maternal behavior). It is notable that the laboratory animal studies vary in experimental designs such as species, strain, gender, dosing regimens (age, routes, vehicle), and test parameters. However, in animals, the doses (1 and 5 mg/kg/day) most often used in the behavior studies are sufficient to elicit approximately ≥10% brain inhibition and ≥30% in RBC inhibition, depending on the study design and the age of the animal. The results of these studies contribute to the overall hazard characterization of chlorpyrifos but are not useful in deriving PoD for risk assessment; the SAP concurred with the Agency's proposed use of the behavioral studies.

Table 4 Summary of Tests and Outcomes in Adults (at least 5 weeks of age, males and/or females) Following Gestational and/or Postnatal Dosing of 1 mg/kg/day Chlorpyrifos.

Behavioral Domain	Device/Task	Outcomes	Species	Reference
Locomotor activity	Figure-8 maze	Decreased habituation rate	Rat	Levin et al., 2002
	Open-field	Increased activity	Mouse	Ricceri et al., 2003
	Elevated plus maze	Increased crossings Decreased crossings	Rat Mouse	Aldridgeet al., 2005c Braquenier et al., 2010
	T maze	Decreased activity	Rat	Icenogle et al., 2004 Levin et al., 2001, 2002
Learning & Memory	Radial arm maze	Increased errors	Rat	Levin et al., 2002 Aldridge et al., 2005c Johnson et al., 2009
	Morris water maze	Slower learning	Mouse	Billauer-Haimovitch et al., 2009
	Foraging in radial arm maze	Slower learning	Mouse	Haviland et al., 2010
Social Interactions (mice)	Agonistic behaviors (male)	Increased	Mouse	Ricceri et al., 2003, 2006
	Induced maternal behaviors (female)	Altered	Mouse	Ricceri et al., 2006
Anxiety/ Depression	Elevated plus maze	Increased time in open arms Decreased time in dark arms	Rat Mouse	Aldridge et al., 2005c Braquenier et al., 2010
	Light/dark box	Decreased time in light side	Mouse	Braquenier et al., 2010

Over the last 15 years, biologically plausible hypotheses for chlorpyrifos have been proposed by researchers. These include effects on signaling pathways (Slotkin, 2006), a morphogenic role of AChE effect the structure of the brain (Brimijoin and Koenigsberger, 1999 and Bigbee et al, 1999; Yang *et al*, 2008) and recently a reduction in axonal transport mediated through impaired tubulin polymerization (Prendergast *et al*, 2007; Grigoryan *et al*, 2008; Grigoryan *et al* 2009; Grigoryan and Lockridge, 2009; Jiang *et al*, 2010) Although multiple mechanisms have been proposed, a coherent mode of action with supportable key events, particularly with regard to dose-response and temporal concordance, has not yet been elucidated. The Agency continues to evaluate new studies on chlorpyrifos and if sufficient information becomes available to perform such an MOA analysis, the Agency may do so in the future. In 2008, the SAP supported the Agency's conclusions that there were insufficient data to clearly identify a specific MOA for effects in the developing nervous system. Some panel members indicated that the data cited in

Eaton *et al*. (2008) could be useful in evaluating alternative (e.g., non-cholinergic) modes of action.

Chlorpyrifos was evaluated for prenatal developmental toxicity in rats, mice and rabbits. In one rat study, developmental effects (increased postimplantation loss) were noted at 15 mg/kg/day (highest dose tested, HDT), that were also associated with maternal toxicity, while another rat study failed to observe similar developmental effects at 15 mg/kg/day. Developmental effects were also noted at higher doses in mice at 25 mg/kg/day (minor skeletal variations, delayed ossification and reduced fetal weight and length) and rabbits at 140 mg/kg/day (decreased fetal weights and crown rump lengths, and unossified xiphisternum and/or 5th sternebra). However, in both mice and rabbits, the developmental effects occurred at maternally toxic doses as indicated by reduced weight gain, and food consumption in both species, and increased mortality in mouse dams.

In the rat developmental neurotoxicity study, chlorpyrifos was associated with alterations in brain development in offspring of exposed mothers. Specifically, pups of the 1 mg/kg/day group exhibited significant decreases in measurements of the parietal cortex in female offspring at postnatal day 66. The only maternal effect at this dose was plasma and RBC ChE inhibition during the treatment period. At higher doses, pups of the 5 mg/kg/day group exhibited decreased body weight/body weight gain and food consumption in both sexes, reductions in pup viability, delays in development, decreased brain weight and morphometric alterations in the brain. However, these effects were observed in the presence of maternal toxicity as evidenced by fasciculations, hyperpnea and hyperactivity, in addition to reduced body weight gain.

Reproductive Effects. Chlorpyrifos induced reproductive toxicity in one generation of rats, but only at dose levels that induced parental toxicity. Reproductive effects in the F1 generation included reduced pup weights and increased pup mortality that corresponded to slightly but significantly reduced body weight gain in their parental F0 dams during lactation days 1-21. In addition, parental toxicity was characterized by inhibition of plasma, RBC and brain cholinesterase activities as well as histological lesions of the adrenal gland (vacuolation of cells of the zona fasciculata).

Carcinogenicity/Genotoxicity. Chlorpyrifos is not likely to be carcinogenic to humans, based on the lack of evidence of carcinogenicity in studies in rats and mice and the absence of a mutagenicity concern. Chlorpyrifos was not mutagenic in bacteria, or mammalian cells, but did cause slight genetic alterations in yeast and DNA damage to bacteria.

Immunotoxicity. There was no sign of immunotoxicity in the guideline study at the highest dose tested.

4.4 Epidemiology

4.4.1 Three Major Epidemiological Prospective Studies in Mothers and Their Children

There are three major prospective epidemiology cohort studies evaluating pre- and post-natal pesticide (chlorpyrifos and/or OPs) exposure in mother-infant pairs with birth outcomes, and

childhood neurobehavioral and neurodevelopment outcomes in neonates, infants, and young children. Two of the cohorts have also investigated the role of genetic susceptibility (PON1) in the association between pesticide exposure and adverse birth outcomes and neurodevelopmental effects. In 2008, EPA consulted the SAP on the use of these three cohort studies in mothers and their children. Details of this analysis and discussion are provided in the chlorpyrifos docket (draft Appendix D Epidemology at http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm.) EPA plans to finalize the science documents reviewed by the SAP in the upcoming months.

Funded by multiple Federal Agencies, including US EPA, the three studies originate from: (1) Columbia University, NYC, (2) Mt Sinai, School of Medicine, NYC, and (3) University of California at Berkeley (Center for Health Assessment of Mothers and Children of Salinas, CHAMACOS). The first two study populations include multi-ethnic, urban low income mother-infant pairs, and the latter reflects a farm worker/agricultural worker study population[2]. The Columbia study focuses on chlorpyrifos in cord blood and the latter two studies assess the non-specific organophosphate (OP) metabolites diethyl phosphases (DEPs) and dialkyl phosphates (DAPs) in maternal urine, and link these biomarkers of exposure with associated health outcomes in children that were exposed *in utero*.

In EPA's review of the epidemiologic evidence concerning potential neurodevelopmental effects of prenatal or early postnatal chlorpyrifos and/or OP exposure, EPA noted consistency across the studies, i.e. delays in cognitive achievement, motor control, social behavior, and intelligence measures that were reported in all three prospective cohorts (Columbia, Mt. Sinai and CHAMACOS) in children 2-3 years of age. However, EPA believes the degree to which chlorpyrifos is implicated in these outcomes varies.

More recently, in April 2011, these same researchers published results indicating that *in utero* chlorpyrifos and/or OP exposure may have persistent neurodevelopmental effects for school age children up to age 7 using the Weschsler Intelligence Scale for Children-IV. Since these studies were recently published, EPA has not conducted a full evaluation of these recent publications (of the same cohort) and integrated these data with the totality of the chlorpyrifos database, but will consider these human epidemiological studies along with the available empiricial data in a full weight of evidence analysis in the final assessment. The neurodevelopmental outcomes reported for children in these epidemiology studies are qualitatively similar to the behavioral outcomes in animal studies (following gestational and/or postnatal exposures to chlorpyrifos). Some initial aspects of these three cohort studies are as follows:

- There appears to be consistency across the three children's health cohorts in both the magnitude and direction of the association between prenatal chlorpyrifos and/or OP exposure and neurodevelopment effects measured in children at several different points in time (Rauh *et al*. 2006, 2011, Engel *et al*. 2011, Eskenazi *et al*. 2007, Bouchard *et al*. 2011). The Columbia results are associated with high chlorpyrifos cord blood levels, while the CHAMACOS and Mt. Sinai teams correlated increasing levels of maternal

[2] The Mt Sinai study included inner city, black, Hispanic and white women, while the Columbia study evaluated inner city African American and Dominican. The Berkley study included homogenous Latino women from Agricultural communities.

urinary DAPs with reported mental delays in children. Table 5 shows the results from the three cohorts on mental delays in children ages 2-3.

Table 5 Prenatal OPs and Mental Delays (Bayley Mental Development Index)

Age of Child	Berkeley (log10 DAP maternal urine, Adj Beta)	Mt. Sinai (log10 DAP maternal urine, Adj Beta)	Columbia Univ. (High v. Low chlorpyrifos cord blood , Adj Beta)
6 mo.	-1.2	--	--
1 Year	-1.3	-1.0	-0.3
2 Year	-3.5*	-2.08	-1.5
3 Year	--	--	-3.3**
*p<0.05 **p<0.1			
References: Eskenazi *et al*. 2007; Engel *et al*. 2011; Rauh *et al*. 2006			

- The Columbia cohort researchers reported that prenatal chlorpyrifos (as measured in umbilical cord blood) is associated with delays in motor development, cognitive function as well as social behavioral problems including symptoms of Attention-Deficit/Hyperactivity Disorder (ADHD) (Rauh *et al*. 2006). Recent study results indicate prenatal chlorpyrifos exposure may adversely influence intelligence measures at school age (Rauh *et al*. 2011).

  The Columbia study overlapped with residential use cancellation in 2001. For children born before cancellation, high chlorpyrifos exposure in cord plasma was significantly associated with neurodevelopmental effects. In contrast, this relationship was no longer significant for newborns born after the cancellation because the blood levels dropped. Thus, this study identifies a natural experiment and indicates the effects upon neurodevelopment in children are not observed upon cessation of the exposure. As noted by the SAP in 2008, ~~–a~~lthough the data on post-ban declines in exposure are compelling, limitations must be kept in mind when using these results in the weight of evidence. The study was not designed to assess the effect of the ban, so data are essentially cross-sectional (i.e., exposures among the same women were not measured over time)."

- Both Mt. Sinai and CHAMACOS cohorts report abnormal reflexes in neonates associated with urinary maternal DEP and DAP levels. Increases in pervasive developmental disorder were reported in both the Columbia and CHAMACOS cohorts (Rauh *et al*. 2006, Engel *et al.* 2007, Young *et al.* 2005). It was acknowledged by the SAP that there are potential confounders and issues that reduce the utility of both the Mt. Sinai and Berkeley cohorts for risk assessment. For example, both studies measured non-specific OP metabolites in urine but not chlorpyrifos. The Berkeley study has the least relevance to chlorpyrifos risk assessment because only a small percentage (10%) of the pesticides applied in Salinas Valley are chlorpyrifos therefore, chlorpyrifos would make only a small contribution to the non-specific metabolites measured in the study and study

outcomes, although this assumption has not been verified. As such, it is difficult to ascribe the effects seen to chlorpyrifos, in particular, rather than OPs in general. Nevertheless, the SAP advised that ~~"~~although Mt. Sinai and Berkely cohorts are less specific than the Columbia Study, they support the overall findings of the latter" (pg 43 SAP report)

- Although CHAMACOS and Mt. Sinai have focused on OP (i.e., DAPs) exposure and not chlorpyrifos, *per* s*e, the SAP encourage*d the Agency to consider the results of all three cohorts together with an emphasis on Columbia University for the chlorpyrifos assessment since as there are ~~"~~more similarities than discrepancies across them" (p. 43, FIFRA SAP, 2008). But the SAP also noted that it cannot be stated that chlorpyrifos is the sole contributor to the observed outcomes; exposures to all three ACh-E inhibiting insecticides may act in combination to produce the observed effects. Although the authors of the Columbia studies have attempted to isolate the effects that would be associated with chlorpyrifos, the Panel noted it is difficult to quantify the contribution of other neurotoxic compounds in such simultaneous exposures. (See follow up analysis by Whyatt and Rauh (2010) that evaluated a joint effects model and concluded chlorpyrifos and not diazinon or propoxur were associated with the outcomes).

There are several strengths of the epidemiological database associating prenatal chlorpyrifos and other OP exposure with neurodevelopment effects in neonates, infants, young children and school aged children. Specifically, the measurement of the neurodevelopmental outcomes (e.g., Brazelton index, Bayley scale, and the Weschlar Intelligence Scale) are accepted valid and reliable measurement tools in clinical and epidemiologic research. In addition, the use of biological markers of exposure [i.e., cord blood concentration of chlorpyrifos or maternal urinary DEPs and DAPs], are more accurate and reliable measures of prenatal exposure than other forms of exposure assessment such as self-report questionnaire. Notably, the exposure measurements (biomonitoring and/or personal air monitoring) were well coordinated with the exposure period of interest (third trimester for birth outcomes)[3]. The Columbia study measured chlorpyrifos in umbilical cord blood at delivery, and maternal blood measurements during pregnancy and delivery. The researchers in each of these cohorts utilized robust and appropriate statistical analysis methods to model the exposure-response association including adjustment for potentially confounding variables.

All three cohort studies have limitations that include multiple chemical exposures and exposure to other organophosphates. The exposure classification is based on maternal spot urinary samples for the Mt. Sinai and CHAMACOS studies and maternal and cord blood samples in the Columbia Study that may not necessarily represent total chlorpyrifos or OP exposure throughout pregnancy because these pesticides have short half-lives. However, the prevalence of exposure among these cohorts based upon a one-time sample indicates the total exposure may be greater than measured (exposure measurement error likely exists), and results of meconium analyses in the Columbia cohort indicate chlorpyrifos exposure occurred throughout pregnancy. Meconium is considered to be an integrative measure of exposure throughout pregnancy. In addition, the Mt. Sinai and CHAMACOS studies associate increased maternal urinary DAP levels with increased mental delays in children. DAP metabolites are non-specific metabolites that result

[3] The third trimester is a critical window of exposure for brain development.

from several OP pesticides, so it is difficult to determine which OP compound may be contributing most to the adverse findings.

While neurodevelopment deficits may be multifactor in origins, these children are from low income multi-ethnic populations and urban neighborhoods and may experience other exposures that may also influence neurodevelopmental outcomes. These may include health disparities that compound pesticide exposure such as poor diet, low access to health care, socioeconomic issues associated with low income and low education, as well as exposure to urban air pollutants. In addition, a recent follow-up publication for the Columbia cohort reported that neighborhood context and chlorpyrifos exposure were independently associated with neurodevelopment (Lovasi *et al*. 2010). Additional analyses were performed to consider neighborhood characteristics, economic deprivation, neighborhood poverty, and maternal hardship to help explain the variation in early childhood psychomotor and mental development. Adjustment for these factors did not change the chlorpyrifos and child neurodevelopment association (Lovasi *et al*. 2010).

The SAP recommended that the epidemiology and direct dosing human studies should not be considered quantitatively for PoDs, but can be used for hazard characterization. The SAP concluded that the results of the three cohort studies (Columbia University, Mt. Sinai Hospital, and the University of California at Berkeley) in concert with the animal studies indicate that ~~–m~~aternal chlorpyrifos exposure would likely be associated with adverse neurodevelopmental outcomes in humans". However, they indicated that exposure to multiple cholinesterase-inhibiting pesticides or other neurotoxicants might result in additive or interactive effects[4]. The Columbia study was considered the most epidemiologically-sound and robust because it measured chlorpyrifos in maternal and cord blood (rather than non-specific metabolites). Challenges in the interpretation of the Mt. Sinai and Berkeley studies include use of non-specific measures of pesticide exposure, based on OP and carbamate metabolites, rather than chlorpyrifos, reduce their utility in a quantitative context for the chlorpyrifos risk assessment.

The Panel recommended that the Agency conduct a full weight of evidence evaluation for the neurodevelopmental outcomes. Such an exercise requires explicit consideration of criteria such as strength, consistency, specificity related to chlorpyrifos or to its anticholinesterase effects common to OPs as a whole, dose-response, temporal concordance and biological plausibility in a framework analysis similar to that which is conducted currently for hypothesized modes of action. This allows comparative analysis across assessments of consistency of weight of evidence determinations. The weight of evidence analysis might increase confidence in this case and potentially identify additional relevant analyses to address uncertainties such as the role of other pesticides in the observed associations.

The SAP recommended that the Columbia University cohort study could be used to determine bounding values for the levels of chlorpyrifos that might cause a measurable effect. In a similar way, data from the other epidemiological studies may also be used in risk assessment. The use of a PBPK model would enable estimation of an exposure dose metric for multiple sources of

[4] Follow up analyses conducted by the Columbia Researchers (Whyatt and Rauh 2010) show that the adverse impact of chlorpyrifos on cognitive development is not due to other anticholinesterase pesticides (diazinon or propoxur exposure).

exposure, e.g., air, food, water. The panel agreed that the blood and urine data in the deliberate human dosing studies are important in interpreting the epidemiology and biomonitoring studies.

The Agency intends to carefully consider the strengths and limitations of the epidemiology studies along with the available empirical data in a full weight of evidence analysis in the final assessment.

4.4.2 Agricultural Health Study (AHS)

For chlorpyrifos, in addition to the guideline carcinogenicity studies, epidemiological data is available from an Agricultural Health Study (AHS). The Agency has reviewed the AHS report and concluded that while the AHS investigations currently published were hypothesis-generating in nature, initial strength and consistency in the findings for lung cancer and colorectal cancer are notable, and warrant further follow-up and additional research. Preliminary associations with breast and prostate cancer are weak, but also warrant monitoring the literature for additional publications on this association. There is no compelling evidence of an association with other cancer sites including pancreatic cancer, melanoma, brain, esophageal, kidney, all lymphohematopoietic cancers combined and NHL, leukemia, and multiple myeloma (C. Christensen, 6/16/11, D388167).

4.5 New Developments since the 2008 SAP

In 2008, the Agency held a SAP meeting (SAP 2008) to discuss the more recent and extensive research on various aspects of chlorpyrifos including its neurological effects in animals and humans following gestational and post-natal exposures, its pharmacokinetics, and mechanism of action. Details can be found in the Chlorpyrifos Final SAP Report at on the Scientific Advisory Panel website (http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm). Many of the key recommendations have been incorporated into this preliminary risk assessment.

Since the 2008 SAP the agency has reviewed new data and analyses, and held additional public meetings to discuss specific aspects of chlorpyrifos including:

- Evaluated new toxicity data
- Consulted the Human Studies Review Board (HSRB)
- Conducted follow up analysis on the Columbia Epidemiology Study
- Consulted the SAP in 2011 on PBPK modeling

New Toxicity Data: Since 2008, the Agency has reviewed an acute and repeat special non-guideline CCA study (Marty and Andrus 2010, MRID No.: 48139301, HED TXR No. 0055409), an acute inhalation study (2010, MRID 48139303), and immunotoxicity study. The CCA study was conducted to compare the relative toxicity of chlorpyrifos and chlorpyrifos oxon in both juvenile (PND11 pups) and adult rats based on ChE inhibition. Both acute (single) dosing and a repeat 11 day exposure scenario were evaluated for chlorpyrifos and chlorpyrifos oxon. Although the study identified both NOAELs and LOAELs for plasma, RBC and brain ChE inhibition, the Agency used these data to conduct BMD analysis (see Appendix E for BMD results). An acute rat inhalation study was evaluated that identified lung and RBC ChE

inhibition as the most sensitive effect at the lowest dose tested of 3.7 mg/m^3. A BMD analysis was attempted but did not provide high confidence results due to the nature of the dose response data. The Agency estimated a HEC of 0.62 mg/m^3 from this study for use in a preliminary assessment of bystander (field volatilization) exposures. In addition, the Agency also reviewed a guideline immunotoxicity study that did not identify adverse effects on the immune system at the highest dose tested.

HSRB: In June 2009, the Agency consulted the Human Studies Review Board (HSRB) (June 24-25, 2009; http://www.epa.gov/hsrb/jun-24-25-2009-public-meeting.htm) regarding deliberate dosing studies in adult (non-pregnant) humans that measure ChE activity and urinary and/or blood levels of chlorpyrifos and/or its metabolites. Nolan *et al* (1982; MRID 124144) was found to be scientifically and ethically conducted by HSRB and EPA also determined that the study was ethically acceptable. Kisicki *et al* (1999), (MRID 44811002) was found to be scientifically (and ethically) conducted by HSRB. However, EPA ethics review had determined that this study did not meet the Agency's ethical standards and therefore concluded that ~~,~~EPA is forbidden by 40 CFR §26.1704 to rely on the Kisicki *et al.* study, MRID 44811002, in actions taken under FIFRA or §408 of FFDCA…." (J.Carley memo dated 5/29/09; http://www.epa.gov/hsrb/files/1d6-ethics-rvw-kisicki-etal-060109.pdf). Thus, the Kisicki data have not been used in the preliminary chlorpyrifos human health risk assessment (Appendix D).

Epidemiology. The SAP recommended a number of follow up analyses for the Columbia cohort. Importantly, the panel advised that ~~,~~it would be useful to examine the results of a statistical analysis that includes all three ChE-inhibiting insecticides in the analysis model as dichotomous variables (above or below the LOD) in combination with continuous measurements for these variables." Follow up analyses conducted by the Columbia Researchers (Whyatt and Rauh 2010) show that the adverse impact of chlorpyrifos on cognitive development is not due to other anticholinesterase pesticides (diazinon or propoxur exposure), and these analyses do not reduce the chlorpyrifos effect for any of the 3-year outcomes for mental or psychomotor delays. The Columbia researchers also addressed a number of other questions raised by the SAP, and they do not affect to conclusions of their publications.

PBPK Modeling. At the 2008 SAP, the panel recommended that the Agency consider the potential for using PBPK modeling in human health risk assessment for chlorpyrifos. PBPK models have been published for chlorpyrifos (Timchalk *et al*, 2002a, 2007; Rigas *et al*, 2001; Knaak, *et al*, 2004; Georgopoulos *et al*, 2008). The model(s) developed by Dr. Charles Timchalk and co-workers at of Pacific Northwest National Laboratory has been the most extensively developed. The Timchalk model was first published in 2002 as an adult rat and human model (Timchalk *et al*., 2002a) and has been updated as more data have become available (Poet *et al*. 2003; Poet *et al*. 2004; Slikker *et al*. 2005; Timchalk *et al*. 2002b; Timchalk *et al*. 2003; Timchalk *et al*. 2005; Lowe *et al*., 2009). Timchalk *et al*. (2007) published a similar model for juvenile rats that incorporated age-dependent changes. Recently, Dow AgroSciences, Dow Chemical and Dr. Timchalk and co-workers have worked collaboratively to improve the chlorpyrifos PBPK model by considering more lifestages (6 month and 3 year olds) and evaluating population variability. The PBPK model has also been linked to a probabilistic exposure model as an approach to estimate population risk. The status of these efforts was considered by the FIFRA SAP in February 2011. At the 2011 meeting, the Panel was

supportive of the overall concepts of linking PBPK models to probabilistic exposure models and of estimating population risk; however, the Panel pointed out limitations to the current effort that precludes its use at the present time. For example, the PBPK models do not simulate pregnancy and thus do not estimate *in utero* exposure to the fetus. In addition, the model only considers oral exposure (with a particular focus on food exposure) but inhalation exposure can be an important route of exposure for chlorpyrifos for both bystanders from field volatilization and to pesticide applicators.

In addition, the Agency is aware of another PBPK modeling effort led by Dr. Dale Hattis of Clark University in collaboration with the Columbia University epidemiology team. This PBPK model may, in the future, be useful in clarifying the exposure concentrations that correspond to the chlorpyrifos levels in umbilical cord blood associated with statistically significant adverse effects on fetal growth and child neurocognitive development.

4.6 Safety factor for Infants and Children (FQPA Safety Factor)

Due to the preliminary nature of this assessment the Agency is presenting assessments reflecting both the retention of the 10X FQPA Safety Factor as in the June 2000 chlorpyrifos risk assessment (USEPA 2000), and a proposal to reduce the FQPA SF to 1X based on more recently available ChE toxicity studies and analyses. In those instances where the Agency has proposed to reduce the FQPA SF to 1x, the Agency believes data are supportive of this proposal. EPA is conducting on-going analyses of newly published literature studies on a variety of challenging scientific issues such as high to low dose extrapolation, animal to human extrapolation, evaluation of the non-cholinergic literature, and interpretation of epidemiology studies in the context of assessing human health risk to chlorpyrifos. EPA will continue to evaluate all the data/studies to determine the appropriate FQPA SF for future chlorpyrifos risk assessments.

4.6.1 Completeness of the Toxicology Database

The toxicological database for chlorpyrifos is extensive and is adequate to support the registration review (Section 4.1 above). The toxicity data base includes the standard battery of guideline studies as well as special studies conducted by the registrant. The scientific literature on chlorpyrifos includes data from many sources, in animals and humans, and some studies with atypical study designs and relatively new assessment techniques. Sources of human information include deliberate dosing studies, epidemiology studies, and metabolism studies (*in vitro* and *in vivo*). There are a variety of laboratory animal studies evaluating a broad range of doses for multiple sensitive lifestages. In addition, these studies consider different durations of exposure (acute, short-, intermediate-term and chronic) and relevant routes of exposure (oral, dermal, and inhalation), different laboratory animal species, reproductive and developmental toxicity, neurotoxicity, developmental neurotoxicity (DNT), new acute and repeat dose comparative cholinesterase assays (CCA) for both chlorpyrifos and chlorpyrifos oxon, a special acute inhalation toxicity study and a required immunotoxicity study.

4.6.2 Key Scientific Information Available to Inform the Safety Factor

The mode of action (MOA) for organophosphate pesticides, like chlorpyrifos, leading to neurotoxicity is inhibition of ChE (See Section 4.3). Concerns regarding the potential hazards to children associated with post-natal exposure to chlorpyrifos and other organophosphate pesticides is derived from data showing that the young have increased sensitivity to ChE inhibition (i.e., the young will have more inhibition than the adult when given the same administered dose). Specific to chlorpyrifos, several toxicity studies with chlorpyrifos and its oxon, including the new CCA study, show that juvenile rat pups are more sensitive to acute chlorpyrifos exposure than adult rats for ChE inhibition. The increased sensitivity of the young from acute exposure is likely attributed to a reduced capacity to detoxify chlorpyrifos in juvenile animals. There is a clear age-dependant sensitivity which diminishes as the pups mature; this pattern is likely reflective of the metabolic processes which rapidly mature in the rat pup. The SAP concurred with the Agency that juveniles are more sensitive than adults to ChE inhibition following acute exposures, but not necessarily for repeated exposures.

In addition to effects on ChE inhibition, there is concern that the young have a unique susceptibility to chlorpyrifos due to its effects on the developing brain. A number of animal toxicity studies examining the effects of chlorpyrifos in the developing brain indicate that gestational and/or early postnatal exposure to chlorpyrifos can lead to neurochemical and behavioral alterations that persist into adulthood (Gupta *et al*, 2011, USEPA 2008 draft hazard and dose response issue paper), including long-term neurobehavioral changes in motor and cognitive behaviors (Aldridge *et al*., 2005c; Levin *et al*., 2001, 2002, Ricceri *et al*., 2003, 2006 Johnson *et al* 2009). However, these findings generally occurred at doses that are often associated with ChE inhibition (> 1 mg/kg/day; the 10% BMD for brain ChE inhibition is about 1.4 mg/kg/day for acute and 0.6-0.8 mg/kg/day for repeated exposure to the PND11 pups in the CCA study) and thus at doses higher than the new oral PoDs being used in this preliminary assessment. In addition, most of the literature studies evaluating non-cholinergic mechanisms and behavioral outcomes provide insufficient information to establish a dose-response due to testing one or two treatment groups and or poor dose selection.

In addition, many *in vitro* literature studies and the guideline developmental neurotoxicity (DNT) study are supportive of the possibility that chlorpyrifos exposure may affect brain development (e.g., altered synaptic development, alterations in DNA, RNA, and protein synthesis, inhibition of mitosis and mitotic figures, and disruption of the structural architecture of the brain) (USEPA 2000b). Qualitative susceptibility between adult rats and their offspring was seen in the guideline DNT study (cholinesterase inhibition in dams at ≥0.3 mg/kg/day versus structural effects on developing brain of the PND 66 offspring at ≥ 1 mg/kg/day) (Hoberman 1998a,b, HED Review D254907). Although an apparent increased qualitative susceptibility was observed in the DNT study, the SAP panel indicated that adult brain morphometric measurements of the cortical regions displayed about 10% variability, a level expected to be within normal variability for such crude measurements. The SAP also advised that histological assessment and morphometric measurements used in the DNT have significant limitations and cannot detect changes in the network organization of the brain or possible other changes. Unbiased stereology should be used for determining cell number and tissue volume. Thus, it is

possible that the Agency's current interpretation of the PND 66 offspring morphometric data in the DNT study may be revisited pending additional information and analysis.

The mode(s) of action associated with the effects on the developing brain are still not known. However, over the last 15 years, biologically plausible hypotheses for chlorpyrifos have been proposed by researchers. These include effects on signaling pathways (Slotkin, 2006), a morphogenic role of ChE effect the structure of the brain (Brimijoin and Koenigsberger, 1999 and Bigbee *et al*, 1999; Yang *et al*, 2008) and recently a reduction in axonal transport mediated through impaired tubulin polymerization (Prendergast *et al*, 2007; Grigoryan *et al*, 2008; Grigoryan *et al* 2009; Grigoryan and Lockridge, 2009; Jiang *et al*, 2010) Although multiple mechanisms have been proposed, a coherent mode of action with supportable key events, particularly with regard to dose-response and temporal concordance, has not yet been elucidated. The Agency may consider additional studies on possible non-cholinergic modes of action in the future, including those cited by Eaton *et al*. (2008), as well as studies reported since that time. The Agency is currently updating its evaluation of the non-cholinerigic literature.

The epidemiological data (Columbia University, Mt. Sinai, and CHAMACOS) do not provide sufficiently robust dose-response information for derivation of a quantitative measure of human risk at this time. The FIFRA SAP (2008) concurred with the proposal to use these studies for qualitatively supporting the risk assessment but not for use in quantitative extrapolation. The SAP pointed out some uncertainties remain the preclude the use of the epidemiology studies in quantitative risk assessment: 1) only measuring biomarkers (3rd trimester maternal, cord blood, meconium) at 1-point in time; 2) the studies do not information as to critical window of effect; and 3) they cannot exclude possibility that effect seen due to chlorpyrifos in combination with other pesticides (additive, multiplicative effect). Similar to many other epidemiology studies, these studies have not measured air exposures, urinary or blood metabolites at or near the timing of pesticide applications. However, the FIFRA SAP said these are high quality studies and supported their use as qualitative information to support the neurodevelopmental toxicity of chlorpyrifos following gestational and/or postnatal exposures since there are ~~"~~more similarities than discrepancies across them". The Columbia study was considered the most epidemiologically-sound and robust because it measured chlorpyrifos in maternal and cord blood (rather than non-specific metabolites). Qualitative similarities between the findings in animal behavioral studies and in the epidemiology studies include impaired cognition, abnormal motor development, and altered social development in children, possibly persisting into school-age (7 years) (Rauh *et al*. 2006, 2011, Engel *et al*. 2011, Eskenazi *et al*. 2007, Bouchard *et al*. 2011).

Inhibition of ChE is the most sensitive endpoint measured in dose response studies in any animal species and in humans, regardless of route or duration of exposure. As such, ChE inhibition has been and continues to be the endpoint used for human health risk assessment for OPs, including chlorpyrifos. In the 2000 risk assessment (EPA 2000a), EPA used a weight of the evidence approach with ChE data from multiple adult laboratory animal studies and multiple species (rat, dog) as the basis of the PoD for all durations and routes of exposure. Since then, numerous new studies in juvenile animals have become available, notably acute and 11 day repeated dosing CCA studies for chlorpyrifos and its oxon. As shown in the draft EPA 2008 issue paper, and draft Appendix C (Mode of Action: Inhibition of Cholinesterase at http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm), there are extensive ChE data

in juvenile rats ranging from PND1-PND33. EPA updated the oral PoDs based on the most sensitive lifestage(s) relevant to direct oral human exposures. Rat pups younger than approximately PND10 are more physiologically similar to human fetuses *in utero*. As such, the Agency has focused its quantitative dose response efforts on rats ages PND11 and older for oral exposures using BMD modeling for 10% RBC ChE inhibition.

EPA is still evaluating the latest epidemiology (Rauh *et al*. 2011, Engel *et al*. 2011, Bouchard *et al*. 2011) and PBPK data and modeling efforts by Dow AgroSciences and Dr. Dale Hattis to be more explicit about uncertainty and variability, and to have a more accurate picture of the doses at which adverse effects might happen in humans and animals. Thus, the PoDs proposed in this preliminary assessment, and associated uncertainty/FQPA factors, could change.

4.6.3 Application of the FQPA Safety Factor for the Preliminary Risk Assessment

In this preliminary assessment, EPA is presenting the risk estimates using both the PoDs and the 10X FQPA SF retained in 2000, and oral PoDs based on new CCA study providing a sensitive endpoint and lifestage with a 1X FQPA and updated quantitative tools (BMD modeling). EPA is continuing to conduct ongoing analysis to ensure a sound scientific basis for the appropriate factor.

2000 Risk Assessment. In the June 2000 chlorpyrifos risk assessment, the FQPA safety factor was retained at 10X for the protection of infants and children to exposure resulting from chlorpyrifos (USEPA 2000a, b, c). At that time, the Agency used PoDs based on NOAELs for plasma and/or RBC ChE inhibition from adult data in laboratory animals and recommended that a 10X safety factor be retained for chlorpyrifos due to:

(1) Increased sensitivity and susceptibility was not only a high dose phenomenon since:

- Increased sensitivity to ChE inhibition following a single oral exposure to neonates was seen at substantially lower doses in PND 7 pups compared to adults (i.e., < 1.5 mg/kg/day) (Zheng *et al*. 2000); and

- A clear qualitative difference in response (i.e., susceptibility) between adult rats and their offspring was demonstrated in the developmental neurotoxicity (DNT) study (cholinesterase inhibition in dams versus structural effects on developing brain of the offspring) (Hoberman 1998a,b, HED Review D254907).

(2) New data available in the literature at the time of the 2000 risk assessment also gave rise to uncertainties such as:

- The suggestion that the inhibition of cholinesterase may not be essential for adverse effects on brain development (see EPA literature review in USEPA 2000b) [5]; and

[5] The mechanism(s) of action for the chlorpyrifos-induced changes (e.g., macromolecular synthesis, cell signaling) is/are unclear. However, given that these effects can be found after intracisternal injection of chlorpyrifos, with in

- The lack of an offspring NOAEL in the DNT based upon structural alterations in brain development as the toxicity endpoint of concern.

2011 Preliminary Risk Assessment:

As noted previously, EPA solicited comment from the SAP in 2008 on extensive research on various toxicological aspects of chlorpyrifos, including its neurological effects in animals and humans following gestational and post-natal exposures, its pharmacokinetics, and mechanism of action. Details can be found in the Chlorpyrifos Final SAP Report at (http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm). The SAP made a number of recommendations on updating the PoDs which are incorporated into the current preliminary risk assessment. Key SAP recommendations included here are: take into account all sensitive life stages; and use benchmark dose modeling instead of the NOAEL/ LOAEL approach when possible.

Since the 2008 SAP meeting, EPA has received and reviewed a new acute and repeat CCA study for both chlorpyrifos and chlorpyrifos oxon (Marty and Andrus 2010). This study is considered high quality, and provides reliable measures of blood and brain ChE at the time of peak effect (6-8 hours post-dosing), uses 4-6 doses and use a wide range of doses. BMD analyses were conducted for both RBC and brain ChE inhibition for this CCA study, in addition to many other literature and registrant studies for both acute and repeat exposure (Betancourt and Carr 2004, Zheng *et al*. 2000, Moser *et al*. 2006, Timchalk *et al*. 2006, Mattsson *et al* 1998, Hoberman 1998a,b) (see Appendix E for BMD analyses). These studies were all considered for endpoint selection. The RBC ChE inhibition BMD for acute chlorpyrifos exposure to PND11 pups administered via milk provides the lowest oral PoD in the entire database of the relevant studies[6] and thus was selected as the new acute oral PoD (Marty and Andrus 2010). The chronic PoD was based on data for pregnant rats in the DNT study (Hoberman 1998a,b), which resulted in the most sensitive PoD, and was also the basis of the 2000 cPoD, along with 4 other studies. For chlorpyrifos oxon, the CCA results were used to develop acute and chronic PoDs based on BMD analysis for RBC ChE inhibition.

Like other OPs, ChE inhibition provides the most sensitive dose-response data for chlorpyrifos. As a result, the focus of the 2011 preliminary risk assessment is on the cholinesterase inhibiting potential of chlorpyrifos. Consistent with this focus, EPA has evaluated the extensive database of ChE data for multiple lifestages and has selected the most sensitive studies which use ages relevant to human exposure. There are no residual uncertainties in the exposure database. The dietary risk assessment is conservative and is not expected to underestimate dietary exposure to chlorpyrifos and chlorpyrifos oxon. Similar to risk assessments conducted for other ChE-inhibiting pesticides where juvenile pups provide the PoDs for risk assessment, the FQPA SF is being reduced to 1X for this preliminary assessment for acute and chronic oral exposure, in addition to dermal and inhalation exposure to chlorpyrifos. The repeated inhalation PoDs are

vitro TCP treatment, and in vitro PC12 cell cultures with limited capability to activate chlorpyrifos to its ChE-inhibiting oxon, raised the issue of whether these effects can occur independent of cholinesterase inhibition.

[6] Data for pups less than PND10 were not considered relevant for direct human exposure, since this represents the human fetal stage

considered protective of sensitive lifestages (pregnant rats). For acute inhalation exposure, a 10X FQPA database uncertainty factor is retained to account for LOAEL to NOAEL extrapolation.

For chlorpyrifos oxon, the Agency proposes to reduce the FQPA SF to a 1X for acute and chronic exposure because the acute PoD is based on a sensitive lifestage (juvenile pups) in the CCA study and the chronic is based on the lowest BMDL available in the CCA study.

Table 6 Application of FQPA SF Comparison of 2000 and 2011 Risk Assessment for Chlorpyrifos

	Point of Departure (mg/kg/day)	
	2000 10X FQPA SF (a)	**Proposed 1X FQPA SF (b)**
Acute PoD	0.5 (mg/kg/day;NOAEL)	0.36 ($BMDL_{10}$)
Chronic PoD	0.03 (NOAEL)	0.03 ($BMDL_{10}$)
Incidental oral	0.5 (NOAEL)	ST: 0.1 ($BMDL_{10}$) IT: 0.03 ($BMDL_{10}$)
Dermal	ST: 5 (NOAEL) IT: 0.03 (NOAEL)	ST/IT: 5 (NOAEL) (c)
Inhalation	ST/IT: 0.1 (NOAEL)	Acute: 0.62 mg/m3 (HEC, LOAEL) (d) ST/IT: 0.0057 mg/m3 (HEC, NOAEL 24 hr residential) (e)

$BMDL_{10}$= benchmark dose lower confidence limit for 10% RBC ChE inhibition
HEC= human equivalent concentration; ST/IT= short and intermediate-term
ST= short –term; IT= intermediate- term

(a) A 10X FQPA SF is retained to the PoDs from the 2000 risk assessment because these are based on adult animal data.
(b) Except where noted, 1X FQPA SF is proposed for 2011 PoDs because they are based on the most sensitive lifestage (i.e., PND 11 and pregnant animals).
(c) For dermal exposure, a 1X FQPA SF is proposed because of the conservative nature of the PoD that is based rat data. Rats have more permeable skin than humans.
(d) For acute inhalation exposure, a 10X FQPA database uncertainty factor is applied to account for LOAEL to NOAEL extrapolation.
(e) For repeated inhalation exposure, a 1X FQPA SF is applied because the PoD is based on route-specific 90 day inhalation studies, and a LOAEL from the DNT study to protect pregnant females from RBC CHE inhibition (LOAEL of 0.3 mg/kg/day)

Table 7 Application of FQPA SF in Risk Assessment for Chlorpyrifos Oxon

	Point of Departure (mg/kg/day)	
	Proposed 10X FQPA SF	**Proposed 1X FQPA SF**
Acute PoD	--	0.05 (BMDL10) (a)
Chronic PoD	--	0.011 (BMDL10) (b)
Dermal	Not applicable	
Inhalation	Not applicable	

(a) 1X FQPA SF proposed since the aPoD is based on sensitive lifestage (juvenile animals).
(b) 1X FQPA database UF because the most sensitive PoD was selected (in 11 day CCA study using adults), that is protective of juvenile rats.

Next Steps in the FQPA SF Analysis:

Analyses are ongoing to fully examine recently proposed biologically plausible modes of action which could lead to effects on the developing brain and to consider these new data in light of the epidemiology studies in mothers and children. As such, the Agency continues to analyze and integrate the animal and human epidemiology data to ensure that a sound scientific analysis around key scientific areas such as high to low dose extrapolation, animal to human extrapolation, and interpretation of epidemiology studies in the context of assessing human health risk to chlorpyrifos. These ongoing analyses will ensure that the PoDs and UFs in this preliminary assessment are human health protective for neurodevelopmental toxicity that may arise from pre- or postnatal exposure. The Agency's final FQPA determination will be based on a full scientific weight of evidence approach that considers the best available science and integrates all key lines of evidence, from empirical animal toxicology to observational human epidemiology studies, in an integrated framework analysis and will transparently address and clearly characterize the strength of the evidence and areas of remaining uncertainty and variability. The Agency plans to conduct a full weight of evidence evaluation integrating the epidemiology studies with the experimental toxicology studies for the neurodevelopmental outcomes using the *Draft Framework for Incorporating Epidemiologic and Human Incidence Data in Human Health Risk Assessment*[7], which was reviewed favorably by the FIFRA SAP in February, 2010 (USEPA, 2010) Such a weight of evidence analysis requires explicit consideration of such criteria as strength, consistency, specificity, dose response, temporal concordance and biological plausibility.

This final determination will also consider the 2008 SAP comments and the public comments received on this preliminary risk assessment. Thus, the intra-, and inter-species UFs along with the FQPA SF could change with these additional analyses. The Agency is seeking comment on the proposed FQPA SF for the final chlorpyrifos risk assessment.

4.7 Toxicity Endpoint and Point of Departure Selections

4.7.1 Dose-Response Assessment

Table 8 summarizes the chlorpyrifos toxicity endpoints and PoDs selected from a re-evaluation of the database (including data submitted since the 2002 IRED/2006 RED). Based on the results of benchmark dose (BMD) analyses and weight of the evidence (WOE) consideration of all quality and reliable data, the most sensitive compartment (i.e., RBC, lung or brain) from the most sensitive sex in both juvenile (> PND11) and adult rats were identified and used for endpoint selection and PoD determination for the following exposure scenarios. Descriptions of the primary toxicity studies used for selecting toxicity endpoints and points of departure for various exposure scenarios are presented in Appendix A of this document. The description and results of the BMD analyses can be found in Appendix E. The SAP recommended selecting PoDs based on BMD analysis for RBC ChE inhibition for the most sensitive lifestages (i.e., pup and pregnant females). The Panel supported the continued use of route-specific data for dermal and inhalation, but advised EPA to take into account sensitive lifestages since these studies are based

[7] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0851-0004

on adult non-pregnant animals. Most of the panel believed that the PoDs based on ChE inhibition would be protective of the developing brain from low level *in utero* exposures, although there was no consensus. The Panel encouraged the Agency to address uncertainties including lack of information on an MOA for behavioral effects and *in vivo* and *in vitro* studies that indicate non-cholinergic MOA are likely to be involved in neurodevelopment and behavioral effects. The Agency is conducting ongoing analyses to ensure that the PoDs and UFs in this preliminary assessment are human health protective for neurodevelopmental toxicity that may arise from pre- or postnatal exposure.

Consistent with risk assessment on other OP and NMCs compounds, the Agency has used a benchmark response (BMR) level of 10% and has thus calculated BMD_{10}s and $BMDL_{10}$s. The BMD_{10} is the estimated dose where ChE is inhibited by 10% compared to background. The $BMDL_{10}$ is the lower confidence bound on the BMD_{10}. Extensive analyses conducted as part of the OP cumulative risk assessment (USEPA 2002) have demonstrated that 10% is a level that can be reliably measured in the majority of rat toxicity studies, and is generally at or near the limit of sensitivity for discerning a statistically significant decrease in ChE activity across the brain compartment and is a response level close to the background brain ChE level. The Agency uses the BMDL, not the BMD, for use as the PoD since the BMDL accounts for variability of the data.

The Agency has not performed BMD analysis on studies evaluating the effect of chlorpyrifos on the developing brain as these do not provide dose response data amenable to BMD modeling analysis. Specifically, these studies, in general, may include only a single dose at a particular age, do not report graded responses (i.e., all or nothing effect), and/or show non-monotonic dose response curves (e.g., response goes up then down). For these studies, the Agency simply considered the doses used.

Acute Dietary (all populations)

Two high quality studies were identified in the re-evaluation of the toxicological database; these include the new CCA rat study (MRID 48139301) and Moser *et al*. (2006) in male PND17 rats. Results of BMD analyses of these well-conducted studies revealed that male and female pup RBC ChE and male whole blood ChE inhibition were the most sensitive endpoints and appropriate as a PoD for the acute dietary (all populations) exposure scenario. A $BMDL_{10}$ of 0.36 mg/kg/day associated with RBC ChE inhibition in male and female rat pups exposed to chlorpyrifos in milk (new CCA study) was selected as a suitable PoD with support from the $BMDL_{10}$ of 0.4 mg/kg from Moser *et al.* (2006). The published studies of Zheng *et al*. (2000) and Timchalk *et al*. (2006) provide additional support for the acute PoD.

An uncertainty factor of 100X (10X to account for interspecies extrapolation and 10X for intraspecies variation) is applied to the $BMDL_{10}$ to obtain an aRfD of 0.0036 mg/kg/day. Based on the proposed FQPA safety factor of 1, the acute population adjusted dose (aPAD) is 0.0036 mg/kg/day.

Chronic Dietary (all populations)

A chronic PoD of 0.03 mg/kg/day ($BMDL_{10}$) was selected from pregnant (GD6-20) rats exposed to chlorpyrifos in the developmental neurotoxicity study (MRID 44556901, Hoberman *et al*. 1998a,b) on the basis of inhibition of RBC ChE in pregnant dams. This PoD was supported by a WOE evaluation of other studies including an oral gavage study in pregnant (GD6-LD10) rats (MRID 44648101) and the new CCA study. An uncertainty factor of 100X (10X to account for interspecies extrapolation and 10X for intraspecies variation) was applied to the $BMDL_{10}$ to obtain a cRfD of 0.0003 mg/kg/day. Based on the proposed FQPA safety factor of 1, the chronic population adjusted dose (cPAD) is 0.0003 mg/kg/day.

Incidental Oral

For short-term incidental oral exposure scenario, the results of the 11 day repeat phase of the new oral CCA study (MRID 48139301) indicated inhibition of RBC ChE in male PND11 rats as the most sensitive endpoint. A $BMDL_{10}$ of 0.1 mg/kg/day was derived from a BMD analysis of the dose- response data. For intermediate-term incidental oral exposure scenarios a $BMDL_{10}$ of 0.03 mg/kg/day was identified (see chronic dietary PoD selection above).

A total uncertainty factor of 100X is appropriate for incidental oral exposures [10X for interspecies extrapolation, 10X for intraspecies variation and a 1X FQPA safety factor].

Dermal

A short-/intermediate-term dermal PoD was selected from a 21-day dermal toxicity study (MRID 40972801) in rats based on plasma and red blood cell ChE inhibition (NOAEL = 5 mg/kg/day). The use of the 21-day dermal toxicity study is appropriate for durations up to 6 months as it is expected that steady state ChE inhibition would have been reached by approximately 21 days of dermal exposure. The Agency has previously shown (USEPA, 2001; preliminary organophosphate cumulative risk assessment) that at or near 3-4 weeks of exposure the degree of inhibition following repeated dosing with OPs does not change with increasing duration but instead remains approximately the same.

For comparison to biomonitoring data in the risk assessment, which evaluates total exposure from oral, dermal and inhalation routes (in terms of absorbed dose), the 21-day rat dermal study is used with an adjustment for 3% dermal absorption to convert the NOAEL of 5 mg/kg/day resulting from topically applied chlorpyrifos to an internal absorbed NOAEL = 0.15 mg/kg/day. The dermal absorption factor of 3% was estimated based on the ratio of the oral LOAEL of 0.3 mg/kg/day from the rat developmental neurotoxicity study (MRIDs 44556901, 44661001) to the dermal LOAEL of 10 mg/kg/day from the 21-day rat dermal study (MRID 40972801) for plasma and red blood cell cholinesterase inhibition. This absorption factor is comparable to the dermal absorption (minimum 1-3%) estimated from human data in Nolan *et al*. (1982, MRID 00249203) by back-calculating chlorpyrifos exposure based on urinary levels of TCP. Most of the absorbed dose in the worker biomonitoring study is the result of dermal exposure.

A total uncertainty factor of 100X is appropriate for dermal exposures [10X for interspecies extrapolation, 10X for intraspecies variation and a 1X FQPA safety factor].

Inhalation

An acute inhalation PoD was selected from a recently submitted special acute inhalation study (2010, MRID 48139303) based on lung and plasma ChE inhibition (LOAEL = 3.7 mg/m^3; NOAEL not established). In this special acute inhalation study, adult female rats (Crl:CD(SD)) were exposed nose only to atmospheric concentrations of 0, 3.7, 12.9, 22.1 or 53.5 mg/m^3 for six hours and allowed an additional 72 hours to recover. Using the Agency's Reference concentration (RfC) methodology, a human equivalent concentration (HEC) was calculated and used to assess acute bystander exposure and risks. The HEC for acute bystander exposure is 0.62 mg/m^3.

Short- and intermediate-term inhalation risk assessments were based on two subchronic inhalation toxicity studies (MRID Nos.40013901, 40166501, 40908401) in the rat. Using the Agency's Reference concentration (RfC) methodology, a human equivalent concentration (HEC) was calculated and used to assess both occupational and residential exposure/risks. The short- and intermediate-term inhalation HEC calculated for residential exposures was converted to a NOAEL of 0.56 mg/kg/day to allow for comparison to estimated occupational inhalation doses (which are in units mg/kg). The HECs are based on no effects on plasma or RBC ChE inhibition identified from the two rat inhalation studies. For residential bystander exposure, the HEC for acute residential bystander exposure is 0.62 mg/m^3, and is 0.0057 mg/m^3 for short- and intermediate-term exposure. Because the 90-day study was conducted 5 days per week at 6 hours/day, the short- and intermediate-term residential HEC was adjusted to represent continual (24 hr, 7 day/week) exposure. In contrast, the occupational inhalation exposure was only adjusted to account for an 8 hour workday because worker exposure is expected to occur during the course of an average workweek (8 hours/day and 5 days/week).

For acute inhalation exposures, a total uncertainty factor of 300X was applied [3X for interspecies extrapolation (reduced from 10X because RfC methodology used which takes into consideration the pharmacokinetic differences between animals and humans), 10X for intraspecies variation, and a 10X FQPA database uncertainty factor (for extrapolation from a LOAEL to a NOAEL).

For short-term and intermediate- term inhalation exposures, a total uncertainty factor of 30X was applied [3X for interspecies extrapolation (reduced from 10X because RfC methodology used), 10X for intraspecies variation and a 1X FQPA SF (because the inhalation NOAEL is considered protective of pregnant females based on effects seen in the DNT at 0.3 mg/kg/day)].

Determination of Acute and Chronic Dietary PoDs for Chlorpyrifos Oxon

There is some potential for direct exposure to the oxon metabolite of chlorpyrifos, particularly from drinking water. BMD modeling of available oxon data for acute and repeated dosing studies was conducted (Appendix E, Tables 7 and 8). The purpose of this analysis is to

determine the toxicological PoDs for the oxon (Table 9) and to assess the relative potency of chlorpyrifos and its oxon metabolite.

A $BMDL_{10}$ of 0.05 mg/kg/day associated with RBC ChE inhibition in male rat pups exposed to chlorpyrifos oxon (acute dosing CCA study using oxon) was selected for the acute dietary PoD for the oxon. An uncertainty factor of 100X (10X to account for interspecies extrapolation and 10X for intraspecies variation) is applied to the $BMDL_{10}$ to obtain an aRfD of 0.0005 mg/kg/day. Based on the FQPA safety factor of 1, the acute population adjusted dose (aPAD) is 0.0005 mg/kg/day.

The chronic dietary PoD for chlorpyrifos oxon is selected from a $BMDL_{10}$ of 0.011 mg/kg/day from an 11 day repeat dosing CCA study using oxon and is based on inhibition of RBC ChE in adult female rats. A comparison of the resulting BMD_{10}s for juvenile and for adult rats indicates that juvenile rats are no more sensitive to the oxon than are adult rats. The BMDL10 for adult rats (0.011 mg/kg/day) was selected for the PoD because it was lower than that of the juvenile rats (0.025 mg/kg/day) and would be considered protective for juveniles. Uncertainty factors of 10X to account for interspecies extrapolation and 10X for intraspecies variation is applied to the $BMDL_{10}$ to obtain an aRfD of 0.00011 mg/kg/day. Based on the FQPA safety factor of 1, the chronic population adjusted dose (cPAD) is 0.000011 mg/kg/day.

Toxicity Factor for Chlorpyrifos Oxon. The Agency developed toxicity factors to estimate the potency of chlorpyrifos oxon relative to chlorpyrifos for the aggregate assessment. While the Agency uses the BMDL, not the BMD, for use as the PoD since the BMDL accounts for variability of the data, the BMD_{10} provides a point of comparison across studies and the BMD_{10} provides the basis for determining the relative toxicity of the chlorpyrifos oxon compared to chlorpyrifos. A toxicity factor for the oxon was calculated by dividing the chlorpyrifos BMD_{10} for the endpoint associated with the most sensitive compartment from the most sensitive sex for the duration of interest by the corresponding BMD_{10} for the oxon. Table 10 summarizes the toxicity values for chlorpyrifos oxon. Acute (all populations) toxicity factors of 8.8 (males) and 11.9 (females) were calculated from BMD analysis of inhibition of male and female pup RBC ChE (acute phase of the CCA study). The chronic toxicity factor of 18.0 was derived from BMD analysis of inhibition of RBC ChE in adult female rats (adult male rats not examined) observed in the repeated phase of the CCA study. The toxicity factors may be used in aggregate assessments where exposures to chlorpyrifos and the oxon are to be combined. Adjusting for relative toxicity will allow comparison of the combined exposures to a single PoD (since PoDs are different for chlorpyrifos and oxon).

4.8 Summary of Points of Departure and Toxicity Endpoints Used in Human Risk Assessment

Table 8 Summary of Toxicological Doses and Endpoints and Points of Departure for Chlorpyrifos for Use in Preliminary Dietary, Non-Occupational (Residential), and Occupational Human Health Risk Assessments

Exposure Scenario	Point of Departure (mg/kg/day)	Study and Toxicological Effects
Acute Dietary (all populations)	$BMDL_{10}$ = 0.36 UF_A = 10x UF_H = 10x FQPA SF = 1x Acute PAD = 0.0036	Inhibition of RBC ChE in male and female rat pups. Weight of evidence from several acute oral studies: • CCA Study (MRID 48139301) in the rat – PND 11 male and female • Data on PND17 males , Moser *et al.*(2006) • Qualitative support from Timchalk *et al.* (2006) and Zheng *et al.* (2000) studies
Chronic Dietary (all populations)	$BMDL_{10}$ = 0.03 UF_A = 10x UF_H = 10x FQPA SF = 1x Chronic PAD = 0.0003	Inhibition of RBC ChE in rat dams (GD 6 – 20). Weight of evidence from studies including: • Developmental neurotoxicity study in pregnant (GD 6 - 20) rats (MRID 44556901) • Gavage study in pregnant (GD 6 – LD10) rats (MRID 44648101)
Short-Term Incidental Oral (1 – 30 days)	$BMDL_{10}$ = 0.1 UF_A = 10x UF_H = 10x FQPA SF = 1x Residential LOC for MOE=100	Inhibition of RBC ChE in PND 11 male rats. • 11 day repeat oral CCA study in the rat (MRID 48139301).
Intermediate – term Incidental Oral	$BMDL_{10}$ = 0.03 UF_A = 10x UF_H = 10x FQPA SF = 1x Residential LOC for MOE=100	See Chronic Dietary.
Dermal Short- (1 – 30 days) and Intermediate-Term (1-6 months)	NOAEL = 5 mg/kg/day [Absorbed dermal NOAEL = 0.15 (for use in comparative assessment using biomonitoring data)]	Plasma and RBC ChE inhibition. 21-day dermal study (NOAEL) and 4 day probe study (LOAEL) in adult rats (MRID 40972801).

Exposure Scenario	Point of Departure (mg/kg/day)	Study and Toxicological Effects
	UF_A = 10x UF_H = 10x FQPA SF = 1x (residential) Residential LOC for MOE=100 Occupational LOC for MOE =100	
Acute Inhalation	Inhalation LOAEL = 3.7 mg/m3 HEC = 0.62 mg/m3 (residential) UF_A = 3x UF_H = 10x FQPA UF_{DB} = 10x (LOAEL to NOAEL extrapolation (residential) Residential LOC for MOE=300	Lung ChE inhibition. • Special 6 hour acute inhalation study (MRID 48139303). (Aerosol)
Inhalation Short- (1 – 30 days) and Intermediate- (1 – 6 months)	NOAEL (calc from HEC) = 0.56 mg/kg/day (8-hr occupational) NOAELHEC = 0.0057 mg/m3 (24 hr residential) UF_A = 3x UF_H = 10x FQPA SF = 1x (residential) Residential LOC for MOE=30 Occupational LOC for MOE =30	Lack of effects in 2 rat inhalation studies at the highest dose tested: LOAEL is based on 43% plasma and 41% RBC ChE inhibition following oral doses of 0.3 mg/kg/day in the DNT study • Two 90-day inhalation studies and the rat DNT study (MRIDs 40908401; 40013901/ 40166501). (Vapor study)

Point of Departure (PoD) = A data point or an estimated point that is derived from observed dose-response data and used to mark the beginning of extrapolation to determine risk associated with lower environmentally relevant human exposures. NOAEL = no observed adverse effect level. LOAEL = lowest observed adverse effect level. UF = uncertainty factor. UF_A = extrapolation from animal to human (interspecies). UF_H = potential variation in sensitivity among members of the human population (intraspecies). UF_L = use of a LOAEL to extrapolate a NOAEL. UF_{DB} = to account for the absence of key data (i.e., lack of a critical study) or other residual uncertainties as evidenced by available data. FQPA SF = FQPA Safety Factor. PAD = population adjusted dose (a = acute, c = chronic). RfD = reference dose. RfC = reference concentration. HEC = human equivalent concentration. MOE = margin of exposure. LOC = level of concern. N/A = not applicable.

Table 9 Summary of Points of Departure, Toxicological Doses and Toxicity Endpoints for Chlorpyrifos oxon for Use in Dietary Exposure Risk Assessments

Exposure/ Scenario	Point of Departure	Uncertainty/FQPA Safety Factors	RfD, PAD, Level of Concern	Study and Toxicological Effects
Acute Dietary (General Population, including Infants and Children)	$BMDL_{10}$ = 0.05	UF_A= 10x UF_H=10x FQPA SF = 1x	Acute RfD = 0.0005 aPAD =0.0005 mg/kg/day	CCA Study (oxon), acute dosing – Inhibition of RBC ChE in male rat pups
Chronic Dietary (All Populations)	$BMDL_{10}$ = 0.011	UF_A= 10x UF_H=10x FQPA SF= 1x	Chronic RfD = 0.00011 mg/kg/day cPAD = 0.00011 mg/kg/day	CCA Study (oxon), 11 day repeat dosing – Inhibition of RBC ChE in adult female rats

Point of Departure (PoD) = A data point or an estimated point that is derived from observed dose-response data and used to mark the beginning of extrapolation to determine risk associated with lower environmentally relevant human exposures. NOAEL = no observed adverse effect level. LOAEL = lowest observed adverse effect level. UF = uncertainty factor. UF_A = extrapolation from animal to human (interspecies). UF_H = potential variation in sensitivity among members of the human population (intraspecies). UF_L = use of a LOAEL to extrapolate a NOAEL. UF_{DB} = to account for the absence of key data (i.e., lack of a critical study) or other residual uncertainties as evidenced by available data. FQPA SF = FQPA Safety Factor. PAD = population adjusted dose (a = acute, c = chronic). RfD = reference dose. RfC = reference concentration. MOE = margin of exposure. LOC = level of concern. N/A = not applicable.

Table 10 Acute and Chronic Relative Toxicity Factors for Chlorpyrifos Oxon (Compared to Chlorpyrifos)

Dietary Scenario	Toxicity Factor (based on BMD_{10} comparison)
Acute Dietary (all populations)	12 ♀ (8.8♂)
Chronic Dietary (all populations)	18

4.9 Endocrine Disruption

As required by FIFRA and FFDCA, EPA reviews numerous studies to assess potential adverse outcomes from exposure to chemicals. Collectively, these studies include acute, subchronic and chronic toxicity, including assessments of carcinogenicity, neurotoxicity, developmental,

reproductive, and general or systemic toxicity. These studies include endpoints which may be susceptible to endocrine influence, including effects on endocrine target organ histopathology, organ weights, estrus cyclicity, sexual maturation, fertility, pregnancy rates, reproductive loss, and sex ratios in offspring. For ecological hazard assessments, EPA evaluates acute tests and chronic studies that assess growth, developmental and reproductive effects in different taxonomic groups. As part of its reregistration decision, EPA reviewed these data and selected the most sensitive endpoints for relevant risk assessment scenarios from the existing hazard database. However, as required by FFDCA section 408(p), chlorpyrifos is subject to the endocrine screening part of the Endocrine Disruptor Screening Program (EDSP).
EPA has developed the EDSP to determine whether certain substances (including pesticide active and other ingredients) may have an effect in humans or wildlife similar to an effect produced by a ~~"~~naturally occurring estrogen, or other such endocrine effects as the Administrator may designate." The EDSP employs a two-tiered approach to making the statutorily required determinations. Tier 1 consists of a battery of 11 screening assays to identify the potential of a chemical substance to interact with the estrogen, androgen, or thyroid (E, A, or T) hormonal systems. Chemicals that go through Tier 1 screening and are found to have the potential to interact with E, A, or T hormonal systems will proceed to the next stage of the EDSP where EPA will determine which, if any, of the Tier 2 tests are necessary based on the available data. Tier 2 testing is designed to identify any adverse endocrine-related effects caused by the substance, and establish a dose-response relationship between the dose and the E, A, or T effect.

Under FFDCA section 408(p), the Agency must screen all pesticide chemicals. Between October 2009 and February 2010, EPA issued test orders/data call-ins for the first group of 67 chemicals, which contains 58 pesticide active ingredients and 9 inert ingredients. Chlorpyrifos was included on that list and has been issued an order to conduct the Tier 1 testing. For further information on the status of the EDSP, the policies and procedures, the list of 67 chemicals, future lists, the test guidelines and the Tier 1 screening battery, please visit our website: http://www.epa.gov/endo/.

5.0 Dietary Exposure and Risk Assessment

5.1. Residues of Concern Summary and Rationale

Plants - The qualitative nature of the residue in plants is adequately understood based on acceptable metabolism studies. The terminal residue of concern in/on plants is chlorpyrifos.

Livestock - The qualitative nature of residue in animals is adequately understood based on acceptable poultry and ruminant metabolism studies. The residue of concern in animals is chlorpyrifos.

Drinking water- The cholinesterase inhibiting metabolite, chlorpyrifos oxon, which has been characterized as having higher toxicity than chlorpyrifos, has been detected in environmental samples including drinking water, surface water, precipitation, and air. The residues of concern for drinking water are chlorpyrifos and the chlorpyrifos oxon.

Table 11 Summary of Metabolites and Degradates to be included in the Risk Assessment and Tolerance Expression

Matrix		Residues included in Risk Assessment	Residues included in Tolerance Expression
Plants	Primary Crop	Chlorpyrifos parent only	Chlorpyrifos parent only
	Rotational Crop	NA	NA
Livestock	Ruminant	Chlorpyrifos parent only	Chlorpyrifos parent only
	Poultry	Chlorpyrifos parent only	Chlorpyrifos parent only
Drinking Water		Chlorpyrifos and Chlorpyrifos oxon	NA

NA = Not Applicable

The chlorpyrifos degradate TCP is not considered a residue of concern as it does not inhibit cholinesterase (separate human health risk assessments have been performed for TCP, which has its own toxicity database).

5.2 Food Residue Profile

Currently, no petitions for the establishment of new tolerances for chlorpyrifos are pending. The previously submitted petitions 7F7248 (alfalfa, alfalfa mixed stands, grass grown for hay or pasture), 3F4188 (barley grain and forage), and 3H5662 (barley, milling fractions) were withdrawn 05/07/2009.

New crop field trial studies have been submitted for cotton gin byproducts (MRID 46651202), tart cherries (MRID 46651201), aspirated grain fractions for soybean, sorghum and wheat (MRID 46640901), and grass forage and hay as part of a data call in related to the 2002 chlorpyrifos IRED. These studies were reviewed previously and regulatory conclusions are included here for all the commodities with the exception of grass forage and hay (original petition withdrawn). In addition, petitions for a PHI reduction for sweet potato and the registration of a microencapsulated formulation were submitted but the requests were cancelled and denied, respectively.

Studies submitted to support the registration of a microencapsulated formulation of chlorpyrifos showed over tolerance residues after a foliar application of Lorsban 4E end use product to lemon with a rate of 6 lb ai/A. The maximum residue observed was 1.41 ppm while the tolerance for citrus fruit (CG 10) is 1.0 ppm. The label of the existing Lorsban 4E end use product (44.9% chlorpyrifos) allows a maximum application rate of 6.4 lb ai/A and addition of oil to the spray mixture. Under these conditions residues over tolerance may occur; therefore, HED recommends a reassessment of the tolerance for citrus fruit using lemon as the representative commodity.

The crop field trial data requested in the IRED and submitted by the petitioner is considered adequate to conclude that tolerances of 15 ppm and 22 ppm would cover any residues of

chlorpyrifos on cotton gin by products and aspirated grain fractions, respectively, and would support the current tolerance level for tart cherries under the condition that only dormant/delayed dormant and trunk spray applications are allowed on the label of the 75% WDG end use product. Acceptable storage stability data (in cherry, alfalfa forage, alfalfa hay and corn grain matrices) is available to support the storage conditions and durations of the samples of tart cherries, cotton and aspirated grain fractions used in these studies.

The dietary burden to livestock was recalculated to consider residues at tolerance level in the feedstock commodities (aspirated grain fractions and cotton gin byproducts) and to use the most current version of Table 1 of the OPPTS Test Guidelines 860.1000, released on June 2008. Based on the residues observed in the feeding study of cattle beef, dairy cow, swine and poultry at the 1x level or higher, HED concludes that the possible residues observed on livestock commodities (from animals fed with feedstock that may contain residues resulting from legal applications) are covered by the current tolerances established in the 40 CFR §180.342.

According to the revised version of Table 1of the OPPTS 860.1000, several studies are required to establish a tolerance for feed items and/or processed commodities that correspond to RACs treated with chlorpyrifos. Tolerances are required for residues of chlorpyrifos on wheat, milled byproducts; wheat, hay; corn, milled byproducts; cotton, meal, hulls and refined oil; and soybean, meal, hulls and refined oil. A magnitude of the residue study to establish a tolerance for wheat hay was required in the previous RED and has not been received. A tolerance was previously established for wheat milling fractions excluding flour as 1.5 ppm. Also, for corn milled byproducts a tolerance of 0.1 ppm was previously recommended based on concentration factors from 1.25x in grits to 2x in flour (D188151, S. Knizner, 20/Aug/1993). Tolerances for residues of chlorpyrifos on wheat milled byproducts and corn milled byproducts should be included in the 40 CFR §180.342. For cotton, processing studies are required to establish tolerances in cotton meal, cotton hulls and refined oil. Similarly, for soybean, processing studies are required to establish tolerances in soybean meal, hulls and refined oil.

[For details of the residue chemistry evaluations see I. Negrón-Encarnación, 5/24/11, D388164, *Chlorpyrifos. Registration Review Action for Chlorpyrifos. Summary of Analytical Chemistry and Residue Data*].

5.3 Water Residue Profile

EFED provided a drinking water assessment (DWA) which includes estimated drinking water concentrations (EDWCs) based on Tier II surface water and Tier I groundwater model simulations for currently registered uses of chlorpyrifos based on the most recent label data report provided by BEAD (R. Bohaty, 06/30/11, D368388 and D389480, *Revised Preliminary Registration Review Chlorpyrifos Drinking Water Assessment*). Tier II surface water EDWCs are more conservative than the Tier I groundwater EDWCs; therefore, only surface water EDWCs are discussed in the section below. This preliminary DWA also considers several sources of monitoring data including datasets from state as well as national programs. Below is a very brief summary of the DWA; see D368388/D389480 for a comprehensive characterization of the drinking water assessment.

EDWCs are provided for chlorpyrifos and chlorpyrifos oxon, a known transformation product of chlorpyrifos. EDWCs for chlorpyrifos oxon were derived from EDWCs calculated for chlorpyrifos because there are limited environmental fate data available for chlorpyrifos oxon and chlorpyrifos is expected to transform to chlorpyrifos oxon during drinking water treatment. Chlorpyrifos EDWCs were multiplied by 0.9541 (molecular weight correction factor) and 100% (maximum conversion during water purification) to generate chlorpyrifos oxon EDWCs. A 100% conversion factor for the oxidation of chlorpyrifos to chlorpyrifos oxon was used as an approximation based on bench scale laboratory data that indicate chlorpyrifos rapidly oxidizes to form chlorpyrifos-oxon almost quantitatively during typical water treatment.[8] Currently, there are no data available on the removal efficiency of chlorpyrifos prior to oxidation to chlorpyrifos oxon, or the removal efficiency of chlorpyrifos oxon. Stability studies indicate that once chlorpyrifos oxon forms during treatment little transformation is likely to occur before consumption (drinking water distribution).[5,9,10] It is possible that some drinking water treatment procedures such as granular activated carbon filtration and water softening (increased rate of chlorpyrifos-oxon hydrolysis at pH > 9) may reduce the amount of chlorpyrifos oxon in drinking water. It is unlikely that these treatment processes significantly reduce the amount of chlorpyrifos-oxon in drinking water. In addition, these treatment methods are not typical practices across the country for surface water. For these reasons, chlorpyrifos-oxon is the residue of concern for drinking water. Additional discussion of the effects of drinking water treatment on chlorpyrifos and chlorpyrifos oxon are provided in the EFED Drinking Water Assessment. Another degradation product of chlorpyrifos, 3,5,6-trichloro-2-pyridinol (TCP) is not examined in this assessment as it is no longer considered to be of toxicological concern.

Tier II modeled (surface water) chlorpyrifos-oxon EDWCs for grapes, corn/soybean and sugar beets are provided in Table 12. These water scenarios are based on both the average typical and maximum label use rates unless otherwise noted in the drinking water assessment. Grape, corn/soybean and sugar beet were singled out for this preliminary drinking water assessment as representative crops because there is a large amount (>100,000 lb) of chlorpyrifos applied to these crops per year, a substantial portion (percent crop treated/percent crop planted) of these crops are treated with chlorpyrifos, and/or the use locations are distributed throughout the United States. In addition, the reported EDWCs for grapes, corn/soybean and sugar beets are generally representative of the other chlorpyrifos use scenarios modeled when EDWCs are compared. All EDWCs for all modeled chlorpyrifos use scenarios are provided in EFED's DWA. Because chlorpyrifos is registered for use on turf (including sod farms, golf courses, road medians, and industrial areas) a percent cropped area (PCA) of 1 (considers 100% of the watershed is treated) was applied to modeling results a standard procedure in EFED. If chlorpyrifos were not used on turf, a PCA value of 0.87 (87% of the watershed is treated) would have been used based on the other crops chlorpyrifos is currently registered for use on; therefore, the EDWCs would be reduced by 13% if turf were not a registered use. This reduction is not expected to alter the conclusions of this risk assessment.

[8] Duirk, S. E.; Collette, T. W.; Degradation of Chlorpyrifos in Aqueous Chlorine Solutions: Pathways, Kinetics, and Modeling. *Environ. Sci. Technol.*, 2006, *40(2)*, 546-550.
[9] Wu, J.; Laird, D. A. Abiotic Transformation of Chlorpyrifos to Chlorpyrifos Oxon in Chlorinated Water. *Environ. Toxcol.Chem.*, **2003**, 22(2), 261-264.
[10] Tierney, D. P.; Christensen, B. R.; Culpepper, V. C. Chlorine Degradation of Six Organophosphate Insecticides and Four Oxons in Drinking Water Matrix. *Submitted by Syngenta Crop Protection, Inc.* **2001**.

Table 12 Estimated Drinking Water Concentrations of Chlorpyrifos-oxon Resulting From Chlorpyrifos Use on Grapes, Corn/Soybean and Sugar beets

Crop Scenario	Chlorpyrifos Oxon (ppb) Average Typical Rate			Chlorpyrifos Oxon (ppb) Maximum Rate		
	1-in-10 Year Peak	1-in-10 Year Annual Average	30 Year Annual Average	1-in-10 Year Peak	1-in-10 Year Annual Average	30 Year Annual Average
Grapes L.R.	2.76	0.41	0.25	107.05	14.06	9.38
Corn/soybean (a)	4.19	0.78	0.48	29.49	4.39	2.98
Sugar Beets	14.36	4.3	1.85	10.06	1.07	0.65

LR= lower rate of two grape application scenarios.

(a) Soybean was only evaluated at the maximum label rate for drinking water.

BEAD provided typical use information to EFED to help refine its assessment. In general, preliminary analysis suggest that typical single application rates correlate well with the modeled single application rates; however, in general the number of applications typically applied each year is less than the maximum allowed on the label. The results of this analysis can also be found in EFED's DWA but are not currently consider in this assessment. Typical agronomic practices also vary from those modeled. In general, the farming methods used over the last five years result in EDWCs that are lower than the most vulnerable scenarios allowed on current labels. Submission of typical use rates and agronomic practices will assist EFED in further refine its final DWA for chlorpyrifos.

There are two modeled chlorpyrifos use scenarios that result in EDWCs that are substantially higher than the majority of the modeled chlorpyrifos use scenarios. These use scenarios are for grape (high rate; 33 lb ai/A) and turf. The EDWCs reported for Grape HR (high rate; 33 lbs a.i./acre) are the result of a high application rate trunk drench/soil application which is currently permitted on labels and may not represent actual or intended use of chlorpyrifos on grape. Some recently approved labels restrict the use of chlorpyrifos on grape to 6 lbs a.i./a. The EDWCs reported for Turf FA (frequent applications) is based on 26 applications (limit of PRZM-EXAMS) and a 3 day application interval. This scenario was developed to highlight the uncertainty associated with the unrestricted use of chlorpyrifos on turf (turf labels do not currently restrict the number of chlorpyrifos applications per year or the maximum number of applications of chlorpyrifos per year) and may not may not represent actual or intended use of chlorpyrifos on turf. Because of the uncertainties with these labels, these modeled EDWCs were not used in the preliminary dietary (water) risk assessment for chlorpyrifos.

Water monitoring data from the USGS National Water-Quality Assessment Program (NAWQA), USEPA/USGS Pilot Reservoir Monitoring Program, USDA Pesticide Data Program (PDP), California Department of Pesticide Regulation (CDPR), and National Center for Water Quality Research (NCWQR) at Heidelberg College were evaluated in reference to an acute exposure to chlorpyrifos and its degradation product chlorpyrifos oxon. The monitoring data show chlorpyrifos detections at low concentrations, generally not exceeding 0.5 µg/L. For example, USGS NAWQA, which contains an extensive monitoring dataset for chlorpyrifos and chlorpyrifos oxon, reports a peak chlorpyrifos detection of 0.57 µg/L in surface water with a detection frequency of approximately 15%. CDPR and NCWQR have detected chlorpyrifos

concentrations greater than 1 ppb in surface water on several occasions. Peak concentrations of chlorpyrifos observed for CDPA and NCWQR are 3.96 and 24 ug/L, respectively. Note the data from NCWQR have not been thoroughly reviewed at this time, but are supplemental. In addition, the NCWQR data are pre-RED and subsequent mitigation. Therefore, it is unclear if NCWQR monitoring data represent current chlorpyrifos uses. EFED is in the process of acquiring more recent data from NCWQR and conducting a more thorough review of the NCWQR data.

In general, the monitoring data include sampling sites that represent a wide range of aquatic environments including small and large water bodies, rivers reservoirs, and urban and agricultural locations. The sampling sites also vary by year and there are limited sampling data available for some areas in the United States where chlorpyrifos is used. None of the monitoring programs were specifically designed to target chlorpyrifos use; therefore, peak concentrations of chlorpyrifos and chlorpyrifos oxon likely went undetected in these programs. Sampling frequencies in high chlorpyrifos use areas are not be designed to capture peak concentrations. The sample frequencies vary from bimonthly to only once per year depending on the program and the sampling site with the exceptions of NCWQR. NCWQR sample frequencies range from daily to monthly. For atrazine (90 day exposure concern) CWS monitoring sampling frequency of 7 days was chosen to be appropriate; however, a recent SAP agreed that a duration of exposure concern that is less than 7 days, such is the case for chlorpyrifos or chlorpyrifos oxon, would likely require even more frequent sampling to capture peaks. This is supported by the NCWQR data as well as PRZM-EXAMS model output time series data and underscores the need for frequent sampling in order to detect peak chlorpyrifos concentrations.

In summary, the monitoring programs analyzed in EFED's DWA do not specifically target chlorpyrifos; consequently, detections cannot be directly associated with a particular use pattern or site nor are the detections expected to represent the potential peak exposure to chlorpyrifos or chlorpyrifos oxon. Additional discussion of the monitoring data can be found in the DWA and is not further discussed in this assessment as it is not considered an appropriate estimation of the potential exposure to chlorpyrifos and chlorpyrifos oxon. The monitoring data were only analyzed in reference to an acute exposure estimation and additional analysis is needed in order to determine if the data contained in the various datasets can be used for longer term exposure durations (i.e., chronic).

DWA Uncertainties

EFED has noted several uncertainties associated with the use of chlorpyrifos. The uncertainties and assumptions are highlighted below.

- While the predominate water treatment method used to disinfect drinking water throughout the United States is chlorination, there are other treatment methods that may reduce chlorpyrifos or chlorpyrifos-oxon exposure concentrations. For facilities that utilize alternative methods, the laboratory data showing 100% conversion of chlorpyrifos to chlorpyrifos-oxon during water purification may not be applicable. Therefore, the chlorpyrifos oxon exposure values presented here may be overestimated for those facilities. Additionally, the oxon may be partially removed with certain treatment processes. In order to reduce the uncertainty associated with the EDWCs reported in this

DWA, additional data including both targeted monitoring data as well as data on the removal efficiency of chlorpyrifos and chlorpyrifos-oxon during treatment is needed. This assessment does not take into account the potential loss of mass (either chlorpyrifos or chlorpyrifos-oxon) during treatment from methods such as activated carbon, sedimentation, water softening, etc., as these treatment methods as well as the sequence of these treatment methods vary considerably across the country. Therefore, for systems that do utilize such treatment methods, the EDWCs reported in this assessment may be higher than the likely exposure concentrations in drinking water. The amount of overestimation is unknown, as currently there are no data available on the removal efficiency of either chlorpyrifos or chlorpyrifos-oxon by these various treatment methods and sequences of treatments. The exception is for water softening where laboratory data can be used to calculate the rate of hydrolysis under water softening conditions ($pH \geq 11$) for both chlorpyrifos and chlorpyrifos-oxon. Water softening, however, is not a common treatment process for surface water.

- Chlorpyrifos is registered for use on turf (including sod farms, golf courses, road medians, and industrial areas), therefore, a percent cropped area (PCA) of 1 (100% of the watershed is treated) was applied to the modeling results in order to cover the use on non-agricultural land. If chlorpyrifos was not registered on turf, the default PCA value of 0.87 (87% of the watershed is treated) would have been used. EFED is currently working on developing crop specific PCAs. For the final DWA, a turf specific PCA may be available to help further refine this assessment. This assessment is national in scope covering multiple chlorpyrifos uses; therefore, it does not take into account regional PCA values (e.g., 0.87 for Missouri, 0.82 for Ohio, 0.07 for Upper Colorado, etc.) or PCA values that represent only a single or a few crops (e.g., 0.46 for corn, 0.83 for corn and soybean, etc.).

- The monitoring programs analyzed for this drinking water assessment do not specifically target chlorpyrifos. Consequently, detections cannot be directly associated with a particular use pattern or site, nor are the detections expected to represent the potential peak chlorpyrifos or chlorpyrifos-oxon exposures. In order to reduce uncertainties and help refine the current exposure assessment, EFED is seeking to incorporate targeted monitoring data in its drinking water assessment.

- Meteorological data and crop profiles, as well as best professional judgment, were used to establish an application date for modeling; however, the selected date may not represent the intended or actual application dates. The application date used for model runs can significantly alter the EDWCs; thus, EDWCs reported could over or under predict the potential exposure. For some chlorpyrifos use scenarios several application dates were evaluated. In general, the date that provided the most conservative EDWCs and corresponded to the appropriate pest pressure are reported. A brief examination of the variation in peak EDWCs for some of the multi-run scenarios ranged from 3-23% for peak EDWCs. Scenarios examined included those that resulted in high and low EDWCs. Based on this limited examination, the application date chosen for modeling can change the peak EDWCs by as much as 23%. This is only an estimate and may vary depending

on the scenario (soil and metrological data) and may not represent all chlorpyrifos use scenarios.

- Many chlorpyrifos labels include application restrictions on a per season basis; however, for some crops there can be multiple seasons per year. For modeling purposes one season was considered to be equal to one year unless otherwise noted. If multiple crop seasons are possible per year it is conceivable that the EDWCs reported in this document may underestimate the actual exposure. In general, this assessment makes conservative assumptions regarding re-cropping and rotations. EFED evaluated a number of labels for specific information regarding application methods and timing, and noted some application rates provided on the label are on a per season basis. The yearly application rates used in this assessment are primarily based on data from BEAD's label data report. The typical use data provided by BEAD to date do not inform this uncertainty as the typical use rate information was not provided for crops that may have multiple seasons per year.

- Some of the labels do not provide maximum single or annual application rates for chlorpyrifos or application retreatment intervals. When this information is not specified on the label, a conservative application scenario was developed and modeled. For example, several labels permit trunk sprays (e.g., some orchard fruit and nut trees such as apples and almonds), at a dilution rate in lbs a.i./100 gallons of water; however, the amount of the dilution that can be applied is not stated on the label. The application rate was assumed to be lb a.i./a. It is unclear if this approach is representative of the intended or actual use scenarios. However, we did find that the average typical application rate provided by BEAD for apples was consistent with the assumed application rate for apples (trunk drench) made for modeling purposes. The extent to which actual use rates may be different is uncertain.

- Some labels restrict the amount of a specific chlorpyrifos formulation; however, the total amount of chlorpyrifos that can be applied per year is not provided. Therefore, the use of multiple chlorpyrifos-containing products is possible. This assessment does not consider the combined use of multiple chlorpyrifos containing products that contain such language, but if such use occurs the reported EDWCs in this assessment may not account for this event.

- Application rates (maximum single applications and yearly/seasonal) vary between labels. Recently approved labels better define chlorpyrifos use; however, there are still several older active labels that do not provide application restrictions or have higher maximum single and/or yearly applications rates than recently approved labels. The most conservative scenarios (highest applications rates) were modeled unless otherwise noted. In order to reduce the uncertainty associated with the EDWCs reported in this preliminary DWA, all chlorpyrifos labels should be updated to clearly state maximum yearly and single application rates, as well as minimum retreatment intervals.

- The monitoring programs analyzed in EFED's DWA do not specifically target chlorpyrifos; consequently, detections cannot be directly associated with a particular use pattern or site nor are the detections expected to represent the potential peak exposure to chlorpyrifos or chlorpyrifos oxon. In order to reduce the uncertainty associated with the interpretation of monitoring data EFED is seeking to incorporate targeted monitoring data in its DWA. Submission of such data would help refine the final risk assessment.

5.4 Dietary Risk Assessment

5.4.1 Description of Residue Data Used in Dietary Assessment

Highly refined acute and chronic dietary (food only, food and drinking water, and drinking water only) exposure and risk assessments of chlorpyrifos were conducted using the Dietary Exposure Evaluation Model DEEM-FCID™, Version 2.03. Risk estimates were determined for the general U.S. population and various population subgroups: all infants (<1 year old), children 1-2, children 3-5, children 6-12, youth 13-19, females 13-49, adults 20-49, and adults 50+ years.

Food residue data for the dietary assessment are almost entirely based upon PDP data. For crops not tested by PDP translations have been made from similar tested crops. Occasionally, older PDP data have been used where it represented the best estimate of real residues. Field trial data or tolerances have been used for a very few crops where translations from PDP data were not possible. The same data sources were used for both the acute and chronic assessments. Most input residues for the acute assessments were incorporated as residue distributions. Input residues for the chronic assessments were applied as a single point estimate (for detailed assumptions, inputs and results see D. Soderberg, 6/30/11, D388166, *Chlorpyrifos Revised Acute (Probabilistic) and Chronic Dietary Exposure and Risk Assessments for Food Only (with and without Food Handling Use included) and for Water Only for the Registration Review Action: Typical Use Rates/Water Included.*

Processing factors from cooking and processing studies were employed where available.

From PDP (and BEAD) data it appears that chlorpyrifos is either applied to a variety of crops which lack the necessary tolerances for chlorpyrifos, or possibly that residues may have occurred on several crops that are rotated in after use of chlorpyrifos on a registered crop. Residues in catfish (no tolerance) were also reported by PDP. Data on agricultural commodities without tolerances are not ordinarily included in HED assessments and were not included in this assessment. Omission of residues on these commodities may lead to underestimation of exposure in the current assessment.

Environmental Fate and Effects Division (EFED) has provided chlorpyrifos and chlorpyrifos-oxon estimated drinking water concentrations (EDWCs) for chlorpyrifos use on grapes, corn/soybean and sugar beets in order to provide a range of possible EDWCs representing the many registered chlorpyrifos uses. In general, these grape, corn/soybean and sugar beet uses represent a broad range of higher end, middle, and lower end EDWCs, respectively, modeled for all chlorpyrifos uses. These particular uses were selected as representative crops for this

preliminary drinking water assessment because there is a large amount of chlorpyrifos applied to these crops per year, a large portion of these crops are treated with chlorpyrifos, and/or the use locations are distributed throughout the United States. All estimated drinking water concentrations used in this assessment are based upon the PRZM-EXAMS model (Table 12 above). For the chronic assessment the 1-in-10 year annual means from PRZM-EXAMS were used. For acute, a distribution of the modeled EDWCs was incorporated into the assessment.

The residues of concern for chlorpyrifos in food are for the parent chlorpyrifos only. Residues of concern in drinking water may include both parent and oxon. All drinking water residues were assumed to be in the form of the oxon as scientific literature suggests rapid and complete conversion of chlorpyrifos to chlorpyrifos-oxon during drinking water disinfection and also show that the oxon is relatively stable after drinking water disinfection. For the preliminary dietary assessment, residues in food are assumed to consist of parent chlorpyrifos only, while residues in water are assumed to consist of chlorpyrifos-oxon only. Therefore food exposures are assessed to toxicological points of departure (PoDs) based upon the toxicity of the parent and water exposures are assessed to PoDs based upon the toxicity of the oxon.

5.4.2 Percent Crop Treated Used in Dietary Assessment

BEAD provided percent crop treated information for over 50 crops [Chlorpyrifos (059101) Screening Level Usage Analysis (SLUA), dated 3/10/2010, and Addendum; see Attachment 3 of D388166]. Where supplied, maximum percent crop treated estimates were used in the acute dietary risk assessment and average percent crop treated estimates were used in the chronic dietary risk assessment. 100% crop treated values were assumed for the following: bananas, figs, radishes, rutabaga roots, turnip roots and greens, garlic, shallots, Brussels sprouts, kohlrabi, collards, kale, mustard and rapeseed greens, citron, citrus hybrids, limes, pommelos, and triticale. BEAD also estimated that less than 2% (default value) of food handling establishments are treated with chlorpyrifos.

5.4.3 Acute Dietary Risk Assessment

The most highly exposed population subgroup for food only was children 1-2, at 9.0% aPAD. The exposure for the general U.S. population from food was 5.1%. Residues in peaches, peppers, apples, plums grapefruit juice, grape juice, soy milk, cranberry juice and orange juice were generally drivers of acute food exposure. (Residues on fresh peaches, plums and peppers in particular strongly tend to be on the imported crops rather than on domestically grown crops.)

For water alone using the lower end representative water scenario (sugar beet) the acute exposure for the general U. S. population ranged from 61-99% of the aPAD based upon the chlorpyrifos-oxon PoD for the maximum and typical application rates, respectively. For all infants, the most highly exposed subpopulation, the exposure ranged from 210-340% of the aPAD for the maximum and typical application rates, respectively.

For water alone using the mid-range representative scenario (corn) the acute exposure for the general U. S. population ranged from 38-240% of the aPAD based upon the chlorpyrifos-oxon PoD for the typical and maximum application rates, respectively. For all infants, the most highly

exposed subpopulation, the exposure ranged from 120-770% aPAD for the typical and maximum application rates, respectively.

For water alone using the higher end representative scenario (grape) the acute exposure for the general U.S. population ranged from 19-810% of the aPAD based upon the chlorpyrifos oxon for the typical and maximum application rates, respectively. For all infants, the most highly exposed subpopulation, the exposure ranged from 59-2700% aPAD for the typical and maximum application rates, respectively.

Table 13 Summary of Preliminary Acute Dietary Food Only Exposure and Risk (Using Parent Chlorpyrifos PoD)

Population Subgroup	Acute Food Only(99.9th percentile) **[Chlorpyrifos aPAD= 0.0036 (includes 1x FQPA Factor)]**	
	Dietary Exposure (mg/kg/day)	% aPAD
General U.S. Population	0.000182	5.1
All Infants (< 1 year old)	0.000190	5.3
Children 1-2 years old	**0.000323**	**9.0**
Children 3-5 years old	0.000275	7.6
Children 6-12 years old	0.000196	5.4
Youth 13-19 years old	0.000122	3.4
Adults 20-49 years old	0.000161	4.5
Adults 50+ years old	0.000170	4.7
Females 13-49 years old	0.000150	4.2

Table 14 Summary of Preliminary Acute Drinking Water Only Exposure and Risk (at the 99.9th Percentile Exposure; Using the Chlorpyrifos-oxon PoD)

Population Subgroup	Lower End Representative Water Scenario (a) (Sugar Beet)		Mid-Range Representative Water Scenario (a) (Corn)		Higher End Representative Water Scenario (a) (Grape)	
	Exposure (µg/kg/day) (% aPAD)		Exposure (µg/kg/day) (% aPAD)		Exposure (µg/kg/day) (% aPAD)	
	Average Typical Rate	Maximum Rate	Average Typical Rate	Maximum Rate	Average Typical Rate	Maximum Rate
General U.S. Population	0.496 (99%)	0.304 (61%)	0.192 (38%)	1.192 (240%)	0.095 (19%)	4.090 (810%)
All Infants (< 1 year old)	**1.677 (340%)**	**1.029 (210%)**	**0.608 (120%)**	**3.840 (770%)**	**0.294 (59%)**	**13.415 (2700%)**
Children 1-2 years old	0.724 (140%)	0.445 (89%)	0.271 (54%)	1.689 (340%)	0.132 (26%)	5.856 (1200%)
Children 3-5 years old	0.654 (130%)	0.404 (81%)	0.242 (48%)	1.526 (310%)	0.118 (24%)	5.259 (1100%)
Children 6-12 years old	0.452 (90%)	0.281 (56%)	0.169 (34%)	1.055 (210%)	0.082 (16%)	3.683 (740%)
Youth 13-19 years old	0.384 (77%)	0.238 (48%)	0.147 (29%)	0.912 (180%)	0.072 (14%)	3.151 (630%)
Adults 20-49 years old	0.442 (88%)	0.270 (54%)	0.164 (33%)	1.026 (210%)	0.081 (16%)	3.538 (710%)
Adults 50+ years old	0.398 (80%)	0.248 (50%)	0.138 (28%)	0.882 (180%)	0.069 (14%)	3.059 (610%)
Females 13-49 years old	0.440 (88%)	0.269 (54%)	0.162 (32%)	1.020 (200%)	0.081 (16%)	3.533 (710%)

Chlorpyrifos-oxon aPAD (includes 1x FQPA Factor) = 0.0005 mg/kg/day or 0.5 µg/kg/day

(a) Lower-end, Mid-range and Higher-end representative scenarios determined based on maximum application rate.

5.4.4 Chronic Dietary Risk Assessment

The chronic dietary exposure assessment was performed with and without food handling establishment (FHE) uses. FHE exposures are more appropriately performed as a chronic dietary

assessment. An acute assessment is more likely to overestimate the risk for FHE exposures since there are no detectable residues in the FHE studies and the percent establishments treated were below the threshold where BEAD is able to accurately quantify.

The most highly exposed population subgroup for chronic food only (excluding FHE uses) were children 1-2, at 8.4 % cPAD, using a PoD based upon the toxicity of chlorpyrifos. The exposure for the general U.S. population from food without FHE use was 3.0 %cPAD. For food with FHE use included the exposure for the general U. S. population was 3.7% cPAD, and for the most highly exposed subpopulation, children 1-2, was 11% cPAD.

For water alone exposure to chlorpyrifos oxon, the risks span a large range, depending on the representative crop assessed (sugar beets, corn, grapes) and application rate. Using the lower end representative scenario (sugar beet) risk estimates did not exceed the level of concern based on the maximum application rates, however there were some risks of concern for average typical rates assessed for infants and children. The resulting risk estimates for the general U. S. population ranged from 21-82% cPAD using a PoD based upon the toxicity of chlorpyrifos-oxon for the maximum and typical rates, respectively. For the most highly exposed subpopulation, all infants, exposure ranged from 69-270 % cPAD depending on the application rate assessed. Drinking water risk estimates for the mid-range and high end representative scenarios (corn and grapes) were not of risk concern at the typical application rates (<49% cPAD) for the highest exposed population, infants (<1 yr), but exceeded the level of concern at the maximum application rates (ranged from 280-890% cPAD) for infants (<1 yr).

The results of the chronic dietary exposure analysis are reported in column 1 of Table 15 below. As can be seen, residues do not exceed the cPAD for any population subgroup. These food exposures are based only upon field use of chlorpyrifos and do not incorporate exposure from food handling establishment (FHE) uses. Estimated potential exposures from FHE uses were assessed separately from other food exposure as a matter of convenience and are provided in column 2 of Table 15, but are additive to the other food exposures. Therefore, column 3 of Table 15 shows the total chronic food plus FHE exposure. It should be noted that there is considerable uncertainty in the exposure estimates for FHE. There appear to be three currently registered FHE uses (labels), but BEAD has been unable to estimate a percent FHE treated and has defaulted to its minimum of 2%. In addition, the expected FHE residues are based upon an FHE residue study with no detectable residues (1/2 LOD is used for FHE anticipated residues).

Table 15 Summary of Preliminary Chronic Dietary Food Only Exposure and Risk (Using Parent Chlorpyrifos PoD)

Population Subgroup	**Chronic Food Only [Chlorpyrifos cPAD= 0.0003 (includes 1x FQPA Factor)]**		**Chronic Food Handling Establishment (FHE) Only [Chlorpyrifos cPAD= 0.0003 (includes 1x FQPA Factor)]**		**Chronic Food with FHE Only [Chlorpyrifos cPAD= 0.0003 (includes 1x FQPA Factor)]**	
	Dietary Exposure (mg/kg/day)	% cPAD	Dietary Exposure (mg/kg/day)	% cPAD	Dietary Exposure (mg/kg/day)	% cPAD
General U.S. Population	0.000009	3.0	0.000002	0.7	0.000011	3.7
All Infants (< 1 year old)	0.000012	4.0	0.000004	1.3	0.000016	5.3
Children 1-2 years old	**0.000025**	**8.4**	**0.000009**	**3.0**	**0.000034**	**11**
Children 3-5 years old	0.000021	7.1	0.000006	2.1	0.000027	9.2
Children 6-12 years old	0.000013	4.3	0.000004	1.3	0.000017	5.6
Youth 13-19 years old	0.000007	2.5	0.000002	0.6	0.000009	3.1
Adults 20-49 years old	0.000007	2.3	0.000001	0.5	0.000008	2.8
Adults 50+ years old	0.000007	2.4	0.000001	0.5	0.000008	2.9
Females 13-49 years old	0.000007	2.2	0.000001	0.5	0.000008	2.7

Table 16 Summary of Preliminary Chronic Drinking Water Only Exposure and Risk (Using the Chlorpyrifos-oxon PoD)

Population Subgroup	Lower End Representative Water Scenario (a) (Sugar Beet)		Mid-Range Representative Water Scenario (a) (Corn)		Higher End Representative Water Scenario (a) (Grape)	
	Exposure (µg/kg/day) (% cPAD)		Exposure (µg/kg/day) (% cPAD)		Exposure (µg/kg/day) (% cPAD)	
	Average Typical Rate	Maximum Rate	Average Typical Rate	Maximum Rate	Average Typical Rate	Maximum Rate
General U.S. Population	0.091 (82%)	0.023 (21%)	0.016 (15%)	0.093 (84%)	0.009 (7.9%)	0.297 (270%)
All Infants (< 1 year old)	**0.297 (270%)**	**0.076 (69%)**	**0.054 (49%)**	**0.304 (280%)**	**0.028 (26%)**	**0.974 (890%)**
Children 1-2 years old	0.135 (120%)	0.034 (31%)	0.024 (22%)	0.138 (130%)	0.013 (12%)	0.441 (400%)
Children 3-5 years old	0.126 (110%)	0.032 (29%)	0.023 (21%)	0.129 (120%)	0.012 (11%)	0.513 (380%)
Children 6-12 years old	0.087 (79%)	0.022 (20%)	0.016 (14%)	0.089 (80%)	0.008 (7.5%)	0.285 (260%)
Youth 13-19 years old	0.066 (60%)	0.017 (15%)	0.012 (11%)	0.067 (61%)	0.006 (5.7%)	0.215 (200%)
Adults 20-49 years old	0.085 (77%)	0.022 (20%)	0.015 (14%)	0.087 (79%)	0.008 (7.3%)	0.277 (250%)
Adults 50+ years old	0.089 (81%)	0.023 (21%)	0.016 (15%)	0.091 (83%)	0.008 (7.7%)	0.292 (270%)
Females 13-49 years old	0.084 (77%)	0.022 (20%)	0.015 (14%)	0.086 (78%)	0.008 (7.3%)	0.276 (250%)

Chlorpyrifos-oxon cPAD (includes 1x FQPA Factor) = 0.00011mg/kg/day or 0.11 µg/kg/day

(a) Lower-end, Mid-range and Higher-end representative scenarios determined based on maximum application rates.

5.4.5 Comparison of Dietary Results for Chlorpyrifos 2000 Risk Assessment and 2011 Preliminary Risk Assessment

For comparison purposes, Table 17 and Table 18 below present the acute and chronic PoDs and resulting dietary risk estimates (for the most highly exposed subpopulations only: young children and/or infants) for the June 2000 chlorpyrifos risk assessment and for the current 2011 preliminary assessment.

The acute and chronic PoDs and resulting dietary risk estimates (for the most highly exposed subpopulations only: young children and/or infants) are compared for the June 2000 chlorpyrifos risk assessment and for the current 2011 preliminary assessment.

In 2000 the acute and chronic dietary PoDs were based on NOAELs (plasma and/or RBC ChEI) from oral studies using adult laboratory animals (including pregnant females). The same PoD, based on toxicity of parent chlorpyrifos, was selected for both food and water. A 10x FQPA factor was retained.

For the 2011 preliminary assessment, the acute and chronic PoDs for *food* exposures were based on the toxicity of parent chlorpyrifos (BMDs for RBC ChEI) to juvenile and pregnant animals, respectively. The acute and chronic PoDs for *water* exposures were based on the toxicity of the chlorpyrifos oxon (BMDs for RBC ChEI) from studies where juvenile and adult animals were directly dosed with the oxon. A 1x FQPA factor is proposed.

The acute dietary (food only) risk estimates for the most highly exposed subpopulation were 82% of the aPAD (2000) and 9% of the aPAD (2011).

In 2000 the acute EDWC was not included in the dietary analysis (water residues not incorporated directly into DEEM analysis) and a % aPAD result was not calculated. Instead a Drinking Water Level of Concern (DWLOC) method was used. An estimated ≤18% aPAD value for 2000 water was estimated herein for comparison purposes only and reflects the exposure amount allowed for water in the 'risk cup' after food exposures are subtracted. In the 2011 preliminary water assessment, a range of representative scenarios was assessed (higher end, mid-range, and lower end). The resulting acute drinking water risk estimates (for infants) ranged from 59% to 340% aPAD for average typical application rates and from 210% to 2700% aPAD for the maximum application rates.

The chronic dietary (food only) risk estimates for the most highly exposed subpopulation were 51% of the cPAD (2000) and 11% of the cPAD (2011).

As in the 2000 acute water assessment, the 2000 chronic water assessment used a DWLOC approach. A ≤ 49% cPAD value was estimated for 2000 water. In the 2011 preliminary water assessment, a range of representative scenarios was assessed (higher end, mid-range, and lower end). The resulting chronic drinking water risk estimates (for infants) ranged from 26% to 270% cPAD for average typical application rates and from 69% to 890% cPAD for the maximum application rates.

It is important to note that, aside from differences in the PoDs and FQPA factors, there have been changes in the dietary input assumptions since 2000. For example, updated food monitoring data and percent crop treated data were used in the 2011 preliminary assessment. For water, in 2000 EDWCs were based on parent chlorpyrifos and were derived from the SCI-GROW model for groundwater and monitoring data for surface water. It is now believed that the existing water monitoring data are not representative of the potential exposure in drinking water and is not recommended for use in quantitative risk assessment. Groundwater EDWCs are expected to be low relative to surface water based on environmental fate characteristics of chlorpyrifos. Therefore, the SCI-GROW modeling results used in 2000 likely underestimate the potential exposure. The 2011 preliminary risk assessment has used a range of surface water EDWCs derived using PRZM-EXAMS modeling. In 2000 the residue of concern in drinking water was assumed to be parent chlorpyrifos. Empirical data indicate rapid conversion of chlorpyrifos to chlorpyrifos oxon during typical drinking water treatment; therefore, this preliminary assessment considers the oxon as the residue of concern in treated drinking water and assumes 100% conversion of chlorpyrifos to oxon. The chlorpyrifos oxon is more toxic than parent chlorpyrifos.

Table 17 Comparison of Chlorpyrifos Acute PoDs and Risk Estimates for 2000 Assessment and 2011 Preliminary Assessment

Acute Dietary Risks			
For highest exposed sub-population	**In 2000** Food and Water PoD (CPY): 0.5 mg/kg/day; total UF=1000 (FQPA=10x)	**In 2011** Food PoD (CPY): 0.36 mg/kg/day; total UF 100 (FQPA=1x) Water PoD (Oxon): 0.05 mg/kg/day; UF 100 (FQPA=1)	
	% aPAD	**% aPAD**	
		Average Typical Application Rate	Maximum Application Rate
Food	82	9.0 (a)	
Drinking Water			
Lower		340	210
Mid-range		120	770
Higher	Estimated ≤18*	59	2700
Aggregate	DWLOC method; not of concern	Not assessed in preliminary assessment	

* not calculated in 2000 (DWLOC method used); this estimated value represents the difference between the aPAD and food exposures, i.e. what was left in the risk cup for water after taking into account food exposures.
(a) Food estimates are highly refined and thus the average typical and maximum application rate scenarios are not applicable.

Table 18 Comparison of Chlorpyrifos Chronic PoDs and Risk Estimates for 2000 Assessment and 2011 Preliminary Assessment

Chronic Dietary Risks (includes FHE uses)			
For highest exposed sub-population	**In 2000** Food and Water PoD (CPY): 0.03 mg/kg/day; total UF=1000 (FQPA=10x)	**In 2011** Food PoD (CPY): 0.03 mg/kg/day; total UF 100 (FQPA=1x) Water PoD (Oxon): 0.011 mg/kg; UF 100 (FQPA=1x)	
	% cPAD	**% cPAD**	
		Average Typical Application Rate	Maximum Application Rate
Food	51	11 (a)	
Drinking Water			
Lower		270	69
Mid-range		49	280
Higher	Estimated ≤49%*	26	890
Aggregate	DWLOC method; not of concern	Not assessed in preliminary assessment	

* not calculated in 2000 (DWLOC method used); this estimated value represents the difference between the cPAD and food exposures, i.e. what was left in the risk cup for water after taking into account food exposures.
(a) Food estimates are highly refined and thus the average typical and maximum application rate scenarios are not applicable.

6.0 Residential (Non-Occupational) Exposure/Risk Characterization

6.1 Residential Handler Exposure

Based upon review of all chlorpyrifos registered uses, only the roach bait products can be applied by a homeowner in a residential setting; however, exposure/risk from application of the roach bait products was not quantitatively assessed because HED expects handler exposure to be negligible. The roach bait product is designed such that the active ingredient is contained within the bait station, therefore, limiting contact with the active ingredient in the product.

6.2 Residential Post-Application Exposure

Chlorpyrifos can be used in areas frequented by the general population including ant mounds on residential properties, golf courses and as an aerial and ground-based (thermal aerosol and fog machine) ULV mosquitocide applied by a public agency made in the vicinity of residential areas. As a result, individuals can be exposed by entering these areas if they have been previously treated. Short-term dermal (adults and children) and incidental oral (children only) exposures to turf following aerial and ground based ULV mosquito treatments have been assessed. Short- and intermediate-term dermal exposure/risk to adults resulting from playing golf has also been assessed. The assumptions and factors used in these risk calculations are consistent with current HED policy for completing residential exposure assessments (i.e., *Draft SOPs for Residential Exposure Assessment*). In addition to these factors, HED has used turf transferable residue (TTR) data from a chemical-specific turf study (MRID 44829601). Post-application exposure from residential ant mound treatment (applied by professional only) was not quantitatively assessed because contact with the mound is not anticipated.

A quantitative residential post-application acute inhalation exposure (spray drift) assessment was also conducted for ground and aerial ULV mosquitocide application. The assessment of residential post-application inhalation was conducted under the assumption that people may be present in the residential setting during the actual ULV application (ground and aerial). This inhalation scenario is anticipated to be an acute event. In contrast, dermal and incidental oral exposures from ULV applications (due to the subsequent settling of airborne residues on turf) are anticipated to be short-term in duration because the potential for exposure extends beyond the application event. For these reasons, residential acute inhalation estimates from ULV application have been presented (Table 21and Table 22) but not aggregated with the other routes of exposure assessed. While the assessment of post-application inhalation from the mosquitocide use has been included under the residential post-application section, it may be more accurately characterized as a spray drift exposure.

Chemical-specific data for mosquito uses are not available. Therefore the equations and assumptions for these scenarios were taken from the *Draft SOPs for Residential Exposure Assessment*. In addition to the use of the Residential SOPs, the unique nature of the mosquito control uses requires additional information to determine the deposition rate of chlorpyrifos (i.e., the amount deposited on residential turf). Deposition rates for ground-based foggers were derived from non-chemical specific studies (Moore *et al,* 1993*;* Tietze *et al*, 1994). In order to calculate deposition and breathing level air concentration from aerial ULV applications, HED

used *AgDRIFT* (V 2.01) which is the model that was developed as a result of the efforts of the *Spray Drift Task Force (SDTF).* Inhalation exposure from ground based ULV treatment was assessed by assuming that the entire active ingredient applied to a 1 acre area is airborne and available to be inhaled by a child or adult.

Risks were calculated using the Margin of Exposure (MOE) approach, which is a ratio of the body burden to the toxicological endpoint of concern. Exposures were calculated by considering the potential sources of exposure then calculating dermal, inhalation and non-dietary ingestion exposures. Short-term dermal (adults and children) and incidental oral (children only) exposures to turf following aerial and ground-based ULV mosquito treatments and adults golfing on treated turf were calculated. In addition, acute inhalation exposures to adults and children were estimated from aerial and ground ULV mosquitocide applications. Detailed assumptions and equations used to estimate exposure and risks can be found in W. Britton, 6/27/11, D388165, *Chlorpyrifos: Occupational and Residential Exposure Assessment*.

Estimated short-term adult and child dermal and child incidental oral exposure to turf following aerial and ground mosquito treatments do not exceed the level of concern (i.e. MOEs are ≥ 100). Combined child exposure estimates (dermal and incidental oral) to turf following aerial mosquito treatment result in risk estimates of concern; however, combined risk estimates following ground treatment are not of concern. Acute adult and child inhalation (spray drift) exposure following aerial mosquito treatment results in risk estimates that are not of concern (i.e. MOEs are ≥ 30), but risk estimates are of concern following ground treatment.

Adult dermal exposure from golfing does not exceed the level of concern (i.e. MOEs are ≥ 100) using any of the transferable residue (TTR) region-specific data for the emulsifiable concentrate formulation at the 1.0 lb ai/A, or 0.25 application rates.

Table 19 Adult and Child Short-term Risks (MOEs) from Residential Post-application Exposure to Turf Following Aerial ULV Mosquito Treatments At 300 Foot Spray Release Height (LOC is an MOE = 100)

Adult	
Dermal	430
Children 3 to < 6	
Dermal	260
Incidental Oral	130
Combined Exposure	88

Table 20 Adult and Child Short-term Risks (MOEs) from Residential Post-application Exposure to Turf Following Ground-based ULV Mosquito Treatments At 300 Foot Spray Release Height (LOC is an MOE = 100)	
Adult	
Dermal	2,200
Children 3 to < 6	
Dermal	1,300
Incidental Oral	670
Combined Exposure	440

Table 21 Adult and Child Acute Risk (MOEs) from Residential Post-application Inhalation Exposure Following Aerial ULV Mosquito Treatments At 300 Foot Spray Release Height (LOC is an MOE = 300)	
Adult and Children 3 to < 6	
Inhalation	1,600

Table 22 Adult and Child Acute Risk (MOEs) from Residential Post-application Inhalation Exposure Following Ground ULV Mosquito Treatments (LOC is an MOE = 300)	
Adult and Children 3 to < 6	
Inhalation	17

Table 23 Adult Estimated Short- and Intermediate-term Risk (MOEs) from Post-application Golfing Exposure to Chlorpyrifos Treated Golf Course Turf (MRID 44829601)		
Application Rate - 1.0 lb ai/A		
State	**Emulsifiable Concn.**	**Granular**
CA	830	960
IN	1,200	NA
MS	710	NA
Application Rate – 0.25 lb ai/A		
State	**Emulsifiable Concn.**	**Granular**
CA	3,300	3,800
IN	4,600	NA
MS	2,800	NA

HED has relied upon the Draft SOPs for Residential Exposure Assessment for all residential scenarios assessed. The data used in the chlorpyrifos residential post-application exposure assessment represent the best exposure data and approaches that are currently available. To the extent possible, HED has used chlorpyrifos-specific data such as the TTR data used for assessment of exposure to treated golf course turf. Chemical-specific data for aerial and ground based ULV mosquito uses are not available. For ground based ULV application, HED used data

from studies conducted to measure off site deposition from these applications. Data similar to that for ground applications were not available to determine aerial deposition. In order to calculate chlorpyrifos deposition on turf and air concentration at breathing level from aerial ULV applications, HED used the *AgDRIFT* (V 2.01) model. Once the deposition input was identified, HED used the high-end equations and assumptions from the *Draft SOPs for Residential Exposure Assessment* to assess dermal exposure to turf and inhalation exposure from mosquito applications. Although the SOPs were initially developed for direct turf applications, the models are used in this assessment to determine if there is a potential concern using a conservative, screening level approach.

HED believes that the values presented in this assessment represent the highest quality results that could be produced based on the currently available post-application exposure data. The quality of individual inputs should be considered when interpreting the risk results. It is difficult to ascertain where, on a distribution, the calculated values fall because the distributional data for exposure, residue dissipation and many other parameters are unrefined. HED does believe, however, that the risks represent conservative estimates of exposure because maximum application rates are used to define residue levels upon which the calculations are based. Additionally, estimates are thought to be conservative even when measures of central tendency (e.g., most transfer coefficients are thought to be central tendency) are used because values that would be considered to be in the lower percentile aspect of any input parameter have not been used in the calculations.

6.3 Residential Bystander Post-application Inhalation Exposure

Recently, the Agency has begun exploring the development of an approach for assessing inhalation exposure resulting from the field volatilization of conventional pesticides. The Agency has sought expert advice and input on these issues from its Federal Insecticide, Fungicide, and Rodenticide Act Scientific Advisory Panel in December 2009. More information on pesticide volatilization can be found on the Agency's website at http://www.epa.gov/opp00001/about/intheworks/volatilization.htm.

The Agency has developed a preliminary bystander volatilization inhalation exposure assessment for chlorpyrifos using currently available inhalation toxicity and air monitoring data. There are 15 available chlorpyrifos air monitoring studies (brief study summaries available in W. Britton, 6/27/11, D388165, *Chlorpyrifos: Occupational and Residential Exposure Assessment*). These include:

- 2 application site studies done in Tulare and Lompoc Counties, CA by the California Air Resource Board (CARB), and
- 13 ambient air studies
    - 2 conducted in North Central and Yakima Valley, OR by the University of Washington Department of Environmental and Occupational Health Sciences; and
    - 11 conducted by Pesticide Action Network North America (PANNA), two in Cowiche and Tieton, WA, and nine in Lindsay, CA.

Application site air monitoring refers to the collection of air samples around the edges of a treated field during and after a pesticide application. Samples are generally collected for short intervals (e.g., < 8 hours), for at least the first day or two after application with subsequent samples increasing in duration. In this type of study, it is typically known when an application occurred, the equipment used for the application, and the application rate. Application site monitoring data represents an exposure to vapors at or near the field edge resulting from an application.

Ambient air monitoring typically is focused on characterizing the airborne pesticide levels within a localized airshed or community structure of some definition (e.g., city, township, or municipality). This type of monitoring effort also can be focused on capturing chronic background levels or other temporal characteristics of interest such as focusing on seasonal pesticide use patterns. Typically, samples are taken for 24 consecutive hours and collected at the same site over an extended period of time (e.g., several weeks or months). In contrast to application site air monitoring, information on the precise timing and location of pesticide applications are rarely collected in ambient air monitoring studies. However, this does not mean that an application did not occur near an ambient sampler during the monitoring period

HED has assessed residential bystander exposure to chlorpyrifos based on the available ambient and application site air monitoring data Table 24. The chlorpyrifos bystander volatilization inhalation exposure assessment includes acute and short-/intermediate-term exposure scenarios. The acute scenario compares the maximum air concentration detected in the monitoring studies to the acute HEC. The short-/intermediate-term scenario compares the arithmetic mean chlorpyrifos air concentration from several monitoring studies to the short -term HEC.

EPA has assessed residential bystander exposure from field volatilization of applied chlorpyrifos based on the available *ambient* and *application site* air monitoring data. Of the 24 acute *ambient* air concentrations assessed, 4 result in risk estimates exceeding the level of concern (i.e. MOEs are < 300). No short-/intermediate-term *ambient* data assessed result in risk estimates of concern (i.e. MOEs are > 30). Of the 5 acute *application site* air concentrations assessed, 3 resulted in a risk estimate of concern (i.e. MOEs are < 300). Of the 5 short- and intermediate-term *application site* air concentrations assessed, 4 resulted in risk estimates of concern (i.e. MOEs are < 30).

Table 24 Chlorpyrifos Preliminary Volatilization MOE Analysis for Residential Bystanders

Study	Year of Study	Sampler/Site Location	Maximum Air Concentration (ng/m^3)	Arithmetic Mean Air Concentration (ng/m^3)	Acute MOEs[a] (LOC is an MOE = 300)	Short- / Int.-term MOEs[b] (LOC is an MOE = 30)
Ambient Air Data						
Washington DOH	2008	North Central District Ambient	21	7	29,000	850
		North Central District Receptor	607	33	1,000	180

Table 24 Chlorpyrifos Preliminary Volatilization MOE Analysis for Residential Bystanders

Study	Year	Sampler/Site Location	Maximum	Arithmetic	Acute	Short- /
		Yakima Valley Ambient	30	9	21,000	620
		Yakima Valley Receptor	243	30	2,500	190
Lompoc County, CA (CARB)	2003	Central	8.3	1.5	19,000	3,800
		Northwest	8.4	0.84	19,000	6,800
		Southwest	6.8	0.78	24,000	7,400
		West	17	2.3	9,400	2,500
Tulare, CA (CARB)	1996	Air Resource Board	39	10	16,000	590
		Jefferson Elementary School	432	94	1,400	61
		Kaweah School	412	70	1,500	82
		Sunnyside Union Elementary School	815	52	760	110
		University of CA, Lindcove Field Station	168	39	3,700	150
Cowiche, WA (PANNA)	2006		462	155	350	37
Tieton, WA (PANNA)	2005		475	182	340	32
Lindsay, CA (PANNA)	2004	Blue House	137	54	1,200	110
Lindsay, CA (PANNA)	2004	Green House	720	120	220	50
Lindsay, CA (PANNA)	2004	Orange House	1,340	190	120	30
Lindsay, CA (PANNA)	2004	Purple House	180	48	900	120
Lindsay, CA (PANNA)	2004	Red House	90	43	1,800	130
Lindsay, CA (PANNA)	2005	Blue House	421	107	380	54
Lindsay, CA (PANNA)	2005	Green House	1,119	177	140	32
Lindsay, CA (PANNA)	2005	Orange House	561	188	290	31
Lindsay, CA (PANNA)	2005	Purple House	515	123	310	47
Application Site Data						
Washington DOH	2008	North Central District Perimeter Site	1145	153	540	37
		Yakima Valley Perimeter Site	1,002	294	620	20
Tulare, CA (CARB)	1996	North	27,700	7,706	22	1
		East	14,700	5,974	42	1
		South	25,400	5,664	24	1

a. Acute MOE = Acute HEC (62,000 ng/m^3) / Study maximum air concentration (ng/m^3).
b. Short-term MOE = Short-term HEC (5,700 ng/m^3) / Study arithmetic mean air concentration (ng/m^3).

Characterization of Bystander Risk Assessment/Uncertainties

Some of the limitations and considerations that have been identified that should be considered in the interpretation of these results include:

- Most of the data utilized in this preliminary assessment are 24-hour air samples. When these data are used, an assumption is made that an individual is exposed to the same air concentration for 24-hours every day. However, this is not always the case as real world time-activity data indicate that many parts of the population move from site to site on a daily basis (e.g., go to work and back).

- This assessment is only representative of outdoor concentrations (i.e., the exposure and risk estimates assume an individual is outdoors all the time). It does not take into account potential effects of air conditioning systems and similar air filtration systems which could potentially reduce air concentrations indoors. The Agency believes that indoor concentrations will be at worst equivalent to outdoor concentrations and may potentially be lower.

- All of the data used for this analysis have been generated in California and Washington; however, chlorpyrifos is used in many regions throughout the country. Therefore, the results based on the limited available air monitoring data were used to represent the rest of the country due to a lack of adequate information for any other region. It is unclear what potential impacts this extrapolation might have on the risk assessment. Factors such as meteorology and cultural practices may impact the overall amounts of chlorpyrifos that volatilize from a treated field as well as the rate at which it volatilizes.

- As part of the December 2009 SAP, the Agency presented their analysis of several models that could be used as screening tools to predict the air concentration and volatilization flux based on intrinsic properties and transport behaviors of pesticides. These models would allow the Agency to better represent the potential volatilization of semi-volatile pesticides across various regions of the country and thus would provide refinement to this assessment over using straight air monitoring data. The SAP provided a number of comments regarding the Agency's model analysis, including the recommendation to evaluate some additional models. The Agency is currently in the process of evaluating the SAP's comments. As appropriate, the Agency will revise the modeling approach presented to the SAP for determining the rate of volatilization (flux) for semi-volatile pesticides and for estimating air concentrations of applied pesticides in the atmosphere under varying environmental conditions. After any policies or procedures are put into place, the Agency may revisit the residential bystander exposure and risk assessment.

6.4 Spray Drift

Spray drift is a potential source of exposure to residents nearby to spraying operations. This is particularly the case with aerial application, but, to a lesser extent, could also be a potential source of exposure from the ground application method employed for chlorpyrifos. The Agency has been working with the Spray Drift Task Force, EPA Regional Offices and State Lead Agencies for pesticide regulation and other parties to develop the best spray drift management practices (see the Agency's Spray Drift website for more information at http://www.epa.gov/opp00001/factsheets/spraydrift.htm). On a chemical by chemical basis, the Agency evaluates the need for interim mitigation measures for aerial applications for placement on product labels/labeling. The Agency has completed its evaluation of the new database submitted by the Spray Drift Task Force, a membership of U.S. pesticide registrants, and is developing a policy on how to appropriately apply the data and the AgDRIFT computer model to its risk assessments for pesticides applied by air, orchard airblast and ground hydraulic methods. After the policy is in place, the Agency may seek further refinements in spray drift management practices to reduce off-target drift with specific products with significant risks associated with drift.

A quantitative residential post-application (acute) inhalation exposure (spray drift) assessment was conducted for ground and aerial ULV mosquitocide applied by a public agency made in the vicinity of residential areas (Section 6.2 above). Inhalation exposure from ground based ULV treatment was assessed by assuming that all of the active ingredient applied to a 1 acre area is airborne and available to be inhaled by a child or adult. HED used *AgDRIFT* (V 2.01), which is the model that was developed as a result of the efforts of the *Spray Drift Task Force (SDTF)*, to determine residue deposition and the airborne concentration of chlorpyrifos anticipated from aerial product application.

7.0 Aggregate Exposure/Risk Characterization

In accordance with the FQPA, HED must consider and aggregate chlorpyrifos exposures and risks from three major sources: food, drinking water, and residential exposures. In an aggregate assessment, exposures from relevant sources are added together and compared to quantitative estimates of hazard (e.g., a NOAEL or PAD), or the risks themselves can be aggregated. When aggregating exposures and risks from various sources, HED considers both the route and duration of exposure.

A quantitative aggregate (food, water and residential exposures combined) assessment was not performed for this preliminary chlorpyrifos assessment. The preliminary risk estimates for water alone exceed the level of concern and are the primary driver in this assessment. Combining food and/or residential exposures with the water exposures would not be expected to have a significant impact on the resulting risk estimates for water alone. A quantitative aggregate assessment for food, water, and residential exposures will be considered during the final chlorpyrifos risk assessment.

8.0 Cumulative Exposure/Risk Characterization

Section 408(b)(2)(D)(v) of the Federal Food, Drug, and Cosmetic Act (FFDCA), as amended by the Food Quality Protection Act (1996) stipulates that when determining the safety of a pesticide chemical, EPA shall base its assessment of the risk posed by the chemical on, among other things, available information concerning the cumulative effects to human health that may result from dietary, residential, or other non-occupational exposure to other substances that have a common mechanism of toxicity. The reason for consideration of other substances is due to the possibility that low-level exposures to multiple chemical substances that cause a common toxic effect by a common mechanism could lead to the same adverse health effect as would a higher level of exposure to any of the other substances individually. A person exposed to a pesticide at a level that is considered safe may in fact experience harm if that person is also exposed to other substances that cause a common toxic effect by a mechanism common with that of the subject pesticide, even if the individual exposure levels to the other substances are also considered safe.

Chlorpyrifos is a member of the organophosphate (OP) class of pesticides. Other members of this class of pesticides are numerous and include azinphos methyl, diazinon, chlorpyrifos-methyl, dichlorvos, dicrotophos, dimethoate, disulfoton, methamidophos, methidathion, monocrotophos, naled, oxydemeton-methyl, phorate, phosmet, pirimiphos-methyl, and trichlorfon to name a few. EPA considers organophosphates to express toxicity through a common biochemical interaction with cholinesterase which may lead to a myriad of cholinergic effects and, consequently the organophosphate pesticides should be considered as a group when performing cumulative risk assessments. HED published the final guidance that it now uses for identifying substances that have a common mechanism of toxicity (FR 64(24) 5796-5799, February 5, 1999) –Proposed Guidance of Cumulative Risk Assessment for Chemicals that have a Common Mechanism of Toxicity" was made available for public comment in the Federal Register (65 FR 40644, June 30, 2000). The Agency presented this approach to the FIFRA Scientific Advisory Panel in late September, 2000. The SAP reviewed revised methods used to conduct a preliminary cumulative risk assessment for organophosphate pesticides in 2002 (US EPA, 2002), found at http://www.epa.gov/scipoly/sap/2002/index.htm.

The Agency has completed a cumulative risk assessment for OPs, (US EPA, 2001), a revised cumulative risk assessment for OPs, (US EPA, 2002), and an updated OP cumulative risk assessment (US EPA, August 2006) which can be found on the Agency's web site http://www.epa.gov/pesticides/cumulative/rra-op/. The cumulative effects of exposure to multiple OPs, including chlorpyrifos, are evaluated in those documents. OPP is in the process of evaluating the most current methods and data for suitability of use in the next version of the OP cumulative risk assessment.

9.0 Occupational Exposure/Risk Characterization

9.1 Short-/Intermediate-Term Handler Risk

Chlorpyrifos is an organophosphate insecticide currently registered for the control of various insects. Targeted pests include aphids, cockroaches, cutworms, fleas, grasshoppers, ticks, etc. Chlorpyrifos is manufactured as granular, microencapsulated, soluble concentrate/liquids, water dispersible granular in water soluble packets (WSP) and wettable powder packaged in WSP formulations, as well as impregnated paints, cattle ear tags, insect bait stations and total release foggers. Registered use sites include the following uses: food crops, including fruit and nut trees, many types of fruits and vegetables, and grain crops; and non-food crops such as forage, golf course turf, industrial sites, greenhouse and nursery production, sod farms, and wood products. Public health uses include aerial and ground-based fogger treatments to control mosquitoes. There are a wide range of application rates. HED has conducted a review of all active product labels. Table 25 and Table 26 below summarize all agricultural and non-agricultural use sites identified for chlorpyrifos under this review, respectively. Maximum application rate (lb ai/A) and application equipment for each site are also identified. Various chlorpyrifos product formulations are represented by the application rates and methods presented in the tables.

Table 25 Summary of Maximum Application Rates for Chlorpyrifos Agricultural Uses

Crop or Target	Maximum Application Rate (lb ai/A)	Application Equipment
Alfalfa	1.0	Aerial, Chemigation, Groundboom, Tractor Drawn Spreader
Asparagus	1.0	Aerial, Groundboom
	1.5	Tractor Drawn Spreader
Beets (Table and Sugar Grown for Seed)	1.9	Aerial
Brassica Vegetable (Bok Choy, Broccoli, Broccoli Raab, Brussel Sprout, Cabbage, Cauliflower, Chinese Broccoli, Collards, Kale, and Kohlrabi)	1.0	Aerial
	2.3	Groundboom, Tractor Drawn Spreader
Carrot (Grown for Seed)	0.94	Aerial, Groundboom
Citrus Fruit	6.0 (AZ and CA), 3.5 (States other than AZ and CA)	Aerial, Airblast, Groundboom
	1.0	Tractor Drawn Spreader
	4.0	Backpack Sprayer, Handgun, Low Pressure Handwand
Clover (Grown for Seed)	1.0	Aerial, Groundboom
Corn (Field, Grown for Seed and Sweet)	1.5	Aerial, Chemigation
	3.0	Groundboom
	1.3	Tractor Drawn Spreader
Cotton	1.0	Aerial, Chemigation, Groundboom
Cranberry	1.5	Aerial, Chemigation, Groundboom
Fig (CA only)	2.0	Groundboom
Grapes	2.0	Airblast
	1.0	Backpack Sprayer, Low Pressure Handwand
Legume Vegetables (Succulent or Dried, Except for Soybeans)	1.0	Groundboom
Mint (Peppermint and Spearmint)	2.0	Chemigation, Groundboom
Onion (Dry Bulb)	1.0	Groundboom, Tractor Drawn Spreader, Handgun
Peanut	2.0	Aerial, Groundboom, Tractor Drawn Spreader
Peppers	1.0	Groundboom

Table 25 Summary of Maximum Application Rates for Chlorpyrifos Agricultural Uses

Crop or Target	Maximum Application Rate (lb ai/A)	Application Equipment
Pineapple (Non-bearing)	1.9	Groundboom, Airblast
Radish	2.8	Aerial, Groundboom, Tractor Drawn Speader, Handgun
Radish (Grown for Seed)	0.94	Aerial, Groundboom
Rutabaga	2.3	Aerial, Groundboom, Tractor Drawn Spreader
Sorghum	1.0	Aerial, Chemigation, Groundboom
	3.3	Tractor Drawn Spreader
Soybean	1.0	Aerial, Chemigation, Groundboom
Strawberry	1.0	Aerial
	2.0	Groundboom
Sugarbeet	1.0	Aerial, Chemigation, Groundboom
	2.0	Tractor Drawn Spreader
Sugarbeet (Grown for Seed)	1.9	Aerial, Chemigation, Groundboom
Sunflower	2.0	Aerial, Groundboom
Sweet Potato	2.0	Groundboom, Tractor Drawn Spreader
Tobacco	2.0	Groundboom, Tractor Drawn Spreader
Tree Nuts (Almonds, Filberts, Pecans, Walnuts)	4.0	Groundboom, Handgun
	2.0	Aerial, Airblast
	0.03 (lb ai/gallon)	Low Pressure Handwand, Backpack Sprayer
Tree Fruit (Apples, Cherry, Nectarine, Peach, Pear, Plum, Prune, Sour Cherry)	2.0	Aerial, Airblast
	0.03 (lb ai/gallon)	Backpack Sprayer, Drench/Dip, Handgun, Low Pressure Handwand
Turnip	2.3	Groundboom, Tractor Drawn Spreader
Wheat	1.0	Aerial, Chemigation, Groundboom
	2.0	Groundboom

Table 26 Summary of Maximum Application Rates for Chlorpyrifos Non-Agricultural Uses

Crop or Target	Maximum Application Rate (lb ai/A)	Application Equipment
Ants (Fire Ant Mound, Carpenter)	0.080 (lb ai/gallon)	Backpack Sprayer, Handgun, Low Pressure Handwand
Cattle Ear Tags	0.0033 (lb ai/ ear tag)	Hand
Christmas Trees (Nurseries and Plantations, Stumps)	1.0	Airblast
	0.94	Aerial (Helicopter)
	0.03 (lb ai/gallon)	Backpack Sprayer, Handgun, Low Pressure Handwand
Golf Course Turf	1.0	Belly Grinder, Low Pressure Handwand, Push Type Spreader, Tractor Drawn Spreader, Turfgun
Grass Seed (Perennial Crops)	1.0	Groundboom
Greenhouse and Nursery Production (Bedding Plants, Containerized Ornamentals, Cut Flowers, Flowering Hanging Baskets, Pine Seedling Transplant, Potted Flowers, Ornamentals, Trees and Shrubs)	4.0	Aerial, Groundboom
	1.1	Belly Grinder, Push Type Spreader, Tractor Drawn Spreader
	1.0	Airblast
	0.16 (lb ai/gallon)	Backpack Sprayer, Handgun, Low Pressure Handwand
	0.02 (lb ai/gallon)	Soil Drench
	0.01 (lb ai/can)	Total Release Fogger
Mosquitocide (Outdoor Residential, Recreational, or Other Non-Cropland Areas)	0.010	Wide Area Aerial and Ground

Table 26 Summary of Maximum Application Rates for Chlorpyrifos Non-Agricultural Uses

Crop or Target	Maximum Application Rate (lb ai/A)	Application Equipment
Non-crop Areas (Commercial Indoor/Outdoor Industrial Sites, Commercial Livestock Holding and Housing, Dumpsters/ Trash Areas, Food Processing Plants, Grown for Seed, Industrial Plant Site Perimeter Treatments, Manufacturing Sites, Power Utilities, Railroad Box Cars, Railroad Equipment, Road Medians, Ship Holds, Sod Farms, Telecommunications, Warehouse Sites)	1.0	Aerial, Belly Grinder, Groundboom, Push Type Spreader, Tractor Drawn Spreader
	0.11 (lb ai/gallon)	Backpack Sprayer
	0.080 (lb ai/gallon)	Handgun, Low Pressure Handwand, Paint Brush/Roller
	0.044 (lb ai/1,000 sq ft)	Shaker Container
	0.018 (lb ai/1,000 sq ft)	Open Pour Bag
Ornamentals (Cut Flowers, Industrial Buildings/Plant Sites Perimeter Treatments and Road Medians, Evergreens, Field Grown Nursery Stock, Flowers, Greenhouses, Non-bearing Fruit Trees Shrubs, Nurseries, Trees, Vines, Woody)	6.0	Belly Grinder, Push Type Spreader, Tractor Drawn Spreader
	4.0	Groundboom
	2.0	Aerial, Airblast
	0.16 (lb ai/gallon)	Backpack Sprayer, Handgun, High Pressure Handwand, Low Pressure Handwand
	0.020 (lb ai/gallon)	Drench/Dip
Roach Control Bait Stations	0.00040 (lb ai/gallon)	Hand
Sewer Manhole Walls	0.080 (lb ai/gallon)	Backpack Sprayer, Low Pressure Handwand, Paint Brush/Roller
Total Release Fogger (Greenhouse Bedding Plants, Cut Flowers, Flowering Hanging Baskets, Potted Flowers, Ornamentals)	0.010 (lb ai/container)	Hand

Table 26 Summary of Maximum Application Rates for Chlorpyrifos Non-Agricultural Uses

Crop or Target	Maximum Application Rate (lb ai/A)	Application Equipment
Trees (Cottonwood and Poplar Trees Grown for Pulp, Conifer, Deciduous, Grown in Nurseries and Greenhouses)	2.0	Airblast, Handgun, Low Pressure Handwand, Backpack Sprayer
	1.9	Aerial
Turfgrass (Sod or Seed)	4.0	Aerial, Groundboom
	1.0	Tractor Drawn Spreader
Wood Products (Fence Posts, Industrial Sites, Landscape Timbers, Logs, Manufacturing, Pallets, Processed Wood, Right of Way, Railroad Ties, Utility Poles, Wooden Containers)	6.0	Belly Grinder, Push Type Spreader
	0.17 (lb ai/gallon)	Low Pressure Handwand
	0.080 (lb ai/gallon)	Backpack Sprayer, Handgun, Paint Brush/Roller

Under current policy, both short- term (up to 30 days) and intermediate-term (30 days up to 6 months) assessments are completed for occupational scenarios in essentially all cases, because these kinds of exposures are likely and acceptable use/usage data are not available to justify disregarding intermediate-term scenarios. Long-term exposure (essentially every working day over a year) is not anticipated based upon the use profile of chlorpyrifos.

Short- (1 to 30 days) and intermediate-term (1 to 6 months) inhalation and dermal exposure and risks were calculated for occupational handlers of chlorpyrifos for a variety of exposure scenarios at differing levels of personal protection. Occupational handler exposure estimates used three major unit exposure data sources, PHED (Pesticide Handlers Exposure Database), the Outdoor Residential Exposure Task Force (ORETF), and recently completed exposure scenario monographs as conducted and submitted by the Agricultural Handlers Exposure Task Force (AHETF). In addition to those surrogate data, two non-chemical specific exposure studies were used (MRID 44793301 and MRID 45250702).

In addition to the aforementioned studies, five chemical specific (chlorpyrifos) handler exposure (biomonitoring and passive dosimetry) studies were previously submitted in support of chlorpyrifos reregistration (MRID 42974501, Shurdut, B.A. *et al*. 1993; MRID 43138102; Honeycutt, R.C. & Day, E.W. Jr. 1994; MRID 44483501 R. F. Bischoff 1998; MRID 44739302, Knuteson *et al*. 1999; and MRID 43027901 Contardi *et al*. 1993). These studies have been reviewed and considered for use by the HED. Based on HED's review of the five chemical specific studies, a number of issues were identified which limit the utility of the available data. HED has determined that these data are most useful as a tool for comparison to the estimates generated for representative exposure scenarios using the surrogate data. That comparison is presented separately in this document. Citations and a full description of the study summaries and issues are presented in W. Britton, 6/27/11, D388165, *Chlorpyrifos: Occupational and Residential Exposure Assessment*.

Because the same adverse effect (i.e., ChEI) was seen following dermal, incidental oral and inhalation exposures, MOEs estimated for these routes of exposure can be combined. However, because the LOCs for dermal/incidental oral and inhalation routes of exposure are not the same (an MOE of 100 defines dermal/incidental oral while inhalation is defined by an MOE of 30) an aggregate risk index (ARI) was required to combine or aggregate estimated MOEs. EPA identifies as a level of concern ARIs that fail to reach or exceed the level of 1. ARIs below 1 result in a risk estimate of concern.

Of the 305 exposure scenarios assessed 134 had risk estimates that did not exceed the level of concern at some level of personal protection (i.e. ARIs are > 1). Ninety-one (91) exposure scenarios had risk estimates not of concern when engineering controls were considered. The remaining 80 scenarios resulted in risk estimates of concern (i.e. ARIs are < 1) at all levels of personal protection and engineering controls considered.

Characterization of Occupational Handler Risk Assessment/Uncertainties

The occupational handler exposure and risk assessment for chlorpyrifos is based upon an array of calculations completed for all identified exposure scenarios using the short- and intermediate-term endpoints. HED completes both short- and intermediate-term assessments for occupational scenarios in essentially all cases, because exposures of these durations are likely and acceptable use/usage data are not available to justify deleting intermediate-term scenarios. HED identified 49 different exposure scenarios which are defined based on the equipment used to make applications or the type of formulation used. Within each of these categories, different application rates and acres treated values were considered to evaluate the broad range of applications that may occur with each kind of equipment (e.g., a groundboom may be used for turf or agriculture). Finally, it should be noted that each calculation was completed at different levels of personal protection to allow for a more informed risk management decision. Even given the scope of the calculations that have already been completed, it is possible that some uses of chlorpyrifos that have not been quantitatively addressed in this document due to lack of exposure data or other information.

The data used in the chlorpyrifos occupational handler risk assessment represent the best data and approaches that are currently available. While some of the data which have been used may not be of optimal quality, they represent the best available data for the scenario in question. In many cases, the Pesticide Handlers Exposure Database (PHED) was used to develop the unit exposure values. PHED data quality varies widely from scenarios that meet guideline requirements for studies to others where a limited number of poor quality data points are available. The results for each scenario should be reviewed in the context of the quality of these data. PHED unit exposure values represent a central tendency of the data (i.e., geometric mean, median or arithmetic mean depending upon the distribution of the data). As such, the values based on the recent studies also are measures of central tendency (e.g., the geometric means were selected from each study for assessment purposes in most cases). HED used recently developed data from AHETF to assess the exposure scenarios mixing/loading liquid formulations and application of liquid sprays via open cab groundboom. HED has reviewed the data for the two studies and has confirmed that it meets study design benchmarks outlined in the AHETF Governing Document (AHETF, 2007) and is considered the most reliable data for assessing exposure and risk for these exposure scenarios. The efforts undertaken by AHETF represent a well-designed, concerted process to collect reliable, internally-consistent, and current exposure data in a way that takes advantage of and incorporates a more robust statistical design, better analytical methods, and improved data handling techniques. For the purpose of the assessment of the two exposure scenarios, HED has used the arithmetic mean unit exposure for short- and intermediate-term exposure durations as recommended in the study reviews (D373605). The AHETF scenarios were recently posted and are publically available on the EPA website as of 4/8/2011 (http://www.epa.gov/pesticides/science/handler-exposure-data.html). This new data were included in the presentation of the most current data used to assess exposure and risk for occupational pesticide handlers.

Along with the unit exposure values used in the assessment, other inputs include application rates and daily acres treated values. The application rates selected for occupational handler risk assessment represent the maximum amounts that are allowed by the label for all uses. The

application rates that were selected for use in the risk assessment were defined based on a review of all current labels. The other key input for completing handler risk assessments used for defining how much chemical can be used in a day is how much can be treated in a day which is generally expressed as the number of acres treated per day. The values used for this parameter represent the HED's most current thinking.

In addition to the key sources of information considered above, there are many underlying factors that may impact the overall results of a risk assessment. For example, the protection factors used for adding additional levels of dermal and respiratory protection may impact the overall risk picture. The factors used in this assessment by HED have been in use for many years. There are exposures monitoring issues which must be considered. For example, in many cases the data included in PHED are based on the use of cotton gloves for hand exposure monitoring which is thought by many to have the potential to overestimate exposure because they potentially retain residue more than a bare hand would over the course of a work day. Such intangible elements of the risk assessment reflect many of the hidden uncertainties associated with exposure data.

In summary, HED believes that the risk values presented in this occupational assessment represent the highest quality results that could be produced given the exposure, use, and toxicology data that are available. Risk managers and other interested parties should consider the quality of individual inputs when interpreting the results and make decisions accordingly. It is difficult to ascertain at what point on a distribution the values which have been calculated fall because the distributional data for exposure, application rates, acres treated and many other parameters are unrefined. HED does believe, however, that the risks represent conservative estimates of exposure because maximum application rates are coupled with high acreage estimates to define risk estimates that likely fall in the upper percentiles of the actual exposure distributions. Additionally, risk estimates are thought to be conservative even when measures of central tendency are combined because values that would be considered to be in the lower percentile aspect of any input parameter have not been used in the calculations.

Occupational Handler Comparison Using Biomonitoring Data

Occupational handler exposure estimates were based on surrogate data from AHETF, ORETF and PHED, as well as two non-chemical specific studies. In addition to these data, five chemical specific handler exposure studies submitted in the past in support of chlorpyrifos re-registration were reviewed and considered for use by the HED. Risk estimates have been calculated using absorbed doses (mg/kg) measured from the biomonitoring studies determined to be acceptable for quantitative risk assessment purposes. In order to characterize occupational risk estimates calculated using surrogate, passive dosimetry exposure data, HED has presented a comparative analysis of these data and biomonitoring data available for applicable exposure scenarios. Comparative estimates using chemical specific handler exposure studies are limited to the level of clothing and personal protection worn by the participants when the studies were conducted. Comparative short- and intermediate term occupational handler exposure/risk using biomonitoring and surrogate exposure data are presented in Appendix B which accompanies document D388165.

All five chemical specific exposure studies (MRID 42974501, MRID 43138102, MRID 44739302 and MRID 43027901 and MRID 44483501) were reviewed for ethical conduct. All but one (MRID 44483501) were determined to be ethically relevant to the standards of the time that the studies were conducted. In addition, all studies were reviewed as to their relevance in the current market in regard to product formulation. Based on this analysis, one study measured the mixing/loading of a wettable powder formulation which is no longer supported by the product registrant. Despite this limitation, the application of the mixed wettable powder formulation was considered for use because the formulation was mixed into a liquid form and is, therefore an acceptable surrogate. HED considered use of the four remaining handler exposure studies and weighed the strengths and weaknesses of each. A number of issues limit the utility of the studies, including: the sufficiency of sample size, PPE worn by study participants, sufficient risk mitigation is not provided (e.g., additional PPE or engineering controls), and the studies do not encompass all handler uses of the chemical.

The number of monitored workers across the four studies range from 15 to 1. HED has typically relied on the criteria set forth by the Pesticide Assessment Guideline which recommend 15 exposure measurements as a minimum for each exposure scenario. Only 1 of the 4 studies meets this criterion. For example, a mixer/loader/applicator greenhouse study (MRID 43027901) attempted to monitor 6 different greenhouse handler scenarios and, as a result was only able to collect 5 measurements for 2 exposure scenarios, 3 for another, and only 1 for each of the remaining 3 scenarios.

Workers participating in the four reviewed studies wore a wide range of clothing and personal protective equipment. All workers wore an inner dosimeter which included t-shirt and briefs used to measure the penetration of chlorpyrifos through the outer dosimeter. The outer dosimeter was typically represented either by coveralls, or long sleeved shirt and long pants. For example, in the mixing/loading for aerial applications study (MRID 44739302), participants wore chemical-resistant gloves, apron and knee high boots. In another study, mixing/loading/applying for ornamentals in greenhouses, the workers wore chemical-resistant gloves, socks, rubber boots, and protective eyewear. In this same study, participants who conducted overhead applications also wore neoprene rain paints and a rain jacket over the coveralls, a half face respirator equipped with two organic vapour cartridges and pre-filters, and a face shield. The study authors attempted to account for the additional PPE through means of assigned penetration factors. It is possible that the additional PPE, despite correction, affect passive and biomonitoring results by reducing worker exposure to chlorpyrifos that would have otherwise deposited on the inner or outer dosimeters in their absence and likewise, result in an assessment which potentially underestimates human health exposure/risk for the exposure scenario. This is of particular concern in the greenhouse study because the personal protective equipment worn by study participants making overhead applications exceeds current labelled requirements.

Because the four available (ethically acceptable) studies were conducted with study participants wearing a specific combination of clothing and PPE, the utility of the data are limited to assessment of occupational handler exposure/risk which is represented by that level of clothing or personal protection. For example, in the mixing/loading for airblast application study (MRID 43138102) workers wore inner dosimeters (t-shirts and briefs), short sleeved shirt, long pants and

outer dosimeters (coveralls), chemical resistant gloves and half-face respirator. Therefore, data from the study would be limited to the assessment of occupational handlers mixing/loading liquids with double layer clothing, gloves and PF10 respirator. Furthermore, since the available exposure studies are specific to particular handler activities and formulations (e.g., mixing/loading for liquid formulations) the utility of the data are limited to these parameters.

In view of the issues outlined, HED has determined that of the four available studies reviewed, three should be considered for quantitative risk assessment purposes (MRIDs 42974501, 43138102, and MRID 44739302). The mixing/loading liquids for aerial applications study (MRID 44739302) was used to present a comparative biomonitoring estimate for the mixing/loading liquids for aerial applications exposure scenario at double layer clothing, gloves and no respirator level of personal protection. The mixing/loading liquids for and airblast application study (MRID 43138102) was used to present a comparative estimate for the mixing/loading liquids for airblast application and airblast application exposure scenario at single layer clothing, gloves and PF10 respirator level of personal protection. Study MRID 42974501 measured exposure from the mixing/loading and application of a wettable powder formulation and mixing/loading of a liquid formulation for ground boom application. Because the wettable powder formulation is no longer supported, only the subsequent liquid application by ground boom exposure data was used to present a comparative estimate at single layer, gloves and PF10 respirator level of personal protection.

Appendix B, which accompany document D388165 present the comparative short- and intermediate-term occupational handler exposure/risk estimates, respectively. Risk estimates compare the level of personal protection measured in biomonitoring exposure studies and the corresponding level estimated using surrogate exposure data.

In an effort to characterize occupational handler risk estimates calculated using both surrogate data and passive dosimetry (chemical specific handler) exposure data, HED has presented a comparative analysis of these for applicable exposure scenarios. Comparative risk estimates were calculated using absorbed doses measured from chemical specific handler studies determined to be acceptable for quantitative risk assessment purposes. The comparison of handler risk estimates was limited based on the level of clothing and personal protection worn by the participants when the biomonitoring studies were conducted.

Of the 4 exposure scenarios compared, 3 (mixing/loading liquids for airblast application, airblast applications, and groundboom applications) result in biomonitoring estimates of greater risk potential than those estimated using surrogate data (i.e., the estimated MOEs are lower). The analysis of the exposure scenario, mixing/loading liquids for aerial application, results in reduced risk potential (a 3.8X reduction in MOE estimate). Because a number of issues were identified which limit the utility of the available biomonitoring data, HED has determined that these data are best suited for characterization of the estimates calculated for representative exposure scenarios using the surrogate data.

Commercial Seed Treatment

Occupational handlers may experience short- and intermediate-term (dermal and inhalation) exposure to chlorpyrifos while performing seed treatment activities in commercial settings. In addition, occupational secondary handlers may experience short- and intermediate-term exposure while planting chlorpyrifos-treated seeds. No chemical-specific handler exposure data were submitted in support of this use pattern. In order to assess commercial seed treatment and seed planting activities, unit exposure data were taken from HED ExpoSAC Policy 14: SOPs for Seed Treatment. The amount of active ingredient handled depends on the application rate (lb ai/lb seed) and the pounds of seed treated in a day (or the pounds of seed that can be planted in a day), all of which vary depending upon the seed type. Values for the amount of seed treated and planted per day were obtained from HED ExpoSAC Policy 15.

Commercial seed treatment exposure and risk estimates were calculated using the formulas and MOE approach used for other occupational handler scenarios. It should be noted that for commercial seed treatment, the application rate is presented in units of lbs ai/lb seed and daily amount handled is presented in units of lbs seed/day.

The majority, 61 of 64, occupational handler seed treatment exposure scenarios assessed (combined dermal and inhalation) resulted in risk estimates which were not of concern (i.e. ARIs are > 1) at some level of personal protection. The remaining 3 exposure scenarios resulted in an ARI < 1 at all level of personal protection considered and, therefore, are of concern. All seed planter (secondary handler) combined short- and intermediate-term dermal and inhalation exposure scenarios assessed resulted in an ARI > 1 at some level of personal protection and, therefore, do not present risk estimates of concern.

Complete results for short- and intermediate-term commercial seed treatment and secondary handler exposure is presented in Appendix C which accompanies document D388165.

9.2 Short-/Intermediate-Term Post-Application Risk

9.2.1 Dermal Post-Application Risk

HED has assessed short- and intermediate term occupational post-application dermal exposure and risk for any crops which reentry into an area previously treated with chlorpyrifos is anticipated. The assessment was completed using 7 chemical-specific registrant submitted DFR studies. The studies, which encompass the use of five different formulations and twelve different crops, have been extrapolated to other groups based on the nature of the crop and application method and used to calculate risks for post-application workers in every region of the county. The results of the post-application exposure and risk assessment are summarized in Table 27 below. A full presentation of post-application exposure and risk including estimates calculated for low, medium and high contact activities and resulting REIs reference Appendix E which accompanies D388165.

The MOEs estimated for liquid spray and granular formulation reentry are not of concern (i.e., an MOE ≥ 100) in the range of 0 to 4 days for lower to medium exposure activities and 0 to 8 days

for high exposure activities, with the greater majority falling between 0 to 4 days when all exposure activities are considered. HED also estimated the MOEs for reentry into microencapsulated and total release fogger formulation treated greenhouses. These estimates range from 0 to > 35 days after treatment (the completion of the monitoring period) for all exposure activities considered.

Table 27 Results of the post-application exposure and risk assessment

Crop Group	Crop	App. Rate (lbs ai/A)	DFR Data Source	DFR Study Location	Growing Region	Estimated REI Range (days) (Dermal LOC = 100)
Berry: Low	Strawberry	1.0	MRID 42974501 (cauliflower WP)	AZ	3, 5, 10 and 12	0 - 2
	Cranberry	1.5			1, 5, 10 and 12	0 - 4
Field and Row Crops: Low to Medium	Clover (Grown for Seed)	1.9	MRID 44748102 (sugar beet EC)	MN	5	1
				OR	11	1
	Perennial Grass Seed Crops	1.0	MRID 44748102 (sugar beet EC)	MN	5	1
				OR	11	1
	Alfalfa	1.0	MRID 44748102 (cotton EC)	TX	5 and 7	1
	Cotton	1.0	MRID 44748102 (cotton EC)	CA	10	0 – 3
				MS	4	0 - 1
				TX	6 and 8	0 - 1
	Peppermint/ Spearmint	2.0	MRID 44748102 (sugar beet EC)	MN	5	0 - 1
				OR	11	0 - 1
	Wheat	1.0	MRID 44748102 (sugar beet EC)	CA	8	1
				MN	5 and 7	1
	Soybean	1.0	MRID 44748102 (cotton EC)	MS	4 and 5	0 - 1
	Sugar Beet	1.0	MRID 44748102 (sugar beet EC)	CA	10	0 - 1
				MN	5	0 - 1
				OR	11	0 - 1
Field and Row Crops: Tall	Corn: Sweet	1.5	MRID 44748102 (sweet corn EC)	IL	1 and 5	0 - 3
				MN	1 and 5	0 - 3
				OR	11	0 - 2
	Corn: Sweet	1.0	MRID 44748102 (sweet corn EC)	IL	1 and 5	0 - 3
				MN	1 and 5	0 - 3
				OR	11	0 - 2
	Corn: Field, Including Grown for Seed	1.5	MRID 44748102 (sweet corn EC)	IL	5	0 - 3
				MN	5	0 - 3

Crop Group	Crop	App. Rate (lbs ai/A)	DFR Data Source	DFR Study Location	Growing Region	Estimated REI Range (days) (Dermal LOC = 100)
	Corn: Field, Including Grown for Seed	1.0	MRID 44748102 (sweet corn EC)	IL	5	0 - 3
				MN	5	0 - 3
	Sorghum	1.0	MRID 44748102 (sweet corn EC)	IL	5, 6, 7 and 8	0 - 1
				MN	5, 6, 7 and 8	0 - 1
	Sunflowers	1.5	MRID 44748102 (sweet corn EC)	IL	5 and 7	1
				MN	5 and 7	1
Tree Fruit: Deciduous	Apple (Dormant and Delayed Dormant)	2.0	MRID 44748101 (apple WP)	CA	10	0 - 1
				WA	11	0 - 2
				NY	1, 2, 5	0 - 2
	Cherry (Sweet) (Dormant and Delayed Dormant)	2.0	MRID 44748101 (apple WP)	CA	10	0 - 1
				WA	11	0 - 2
				NY	5	0 - 2
	Cherry (Sour) (Dormant and Delayed Dormant)	2.0	MRID 44748101 (apple WP)	NY	1 and 5	0 - 2
	Peaches (Dormant and Delayed Dormant)	2.0	MRID 44748101 (apple WP)	CA	10	0 - 1
				NY	2	0 - 2
	Pears (Dormant and Delayed Dormant)	2.0	MRID 44748101 (apple WP)	CA	10	0 - 1
				WA	11	0 - 2
	Nectarines, Plums, Prunes (Dormant and Delayed Dormant)	2.0	MRID 44748101 (apple WP)	CA	10	0 - 1
	Apples	1.5	MRID 44748101 (apple WP)	CA	10	0 - 1
				WA	11	0 - 4
				NY	1, 2, 5	0 - 4
	Cherries (Sour)	1.5	MRID 44748101 (apple WP)	NY	1 and 5	
	Peaches (Post-harvest)	3.0	MRID 44748101 (apple WP)	CA	10	0 - 1
				NY	2	0 - 2
	Pears (Post-harvest)	2.0	MRID 44748101 (apple WP)	CA	10	0 - 1
				WA	11	0 - 2
Tree Fruit: Evergreen	Conifer Trees and Christmas Tree Plantations	1.0	MRID 43062701 (citrus EC)	CA	Any	0 - 1
	Citrus	6.0 (CA and AZ)	MRID 43062701 (citrus EC)	CA	3 and 10	0 - 2

Crop Group	Crop	App. Rate (lbs ai/A)	DFR Data Source	DFR Study Location	Growing Region	Estimated REI Range (days) (Dermal LOC = 100)
		3.5	MRID 43062701 (citrus EC)	CA	3 and 10	0 - 1
Forestry	Cottonwood/ Poplar Trees Grown for Pulp (Dormant)	2.0	MRID 44748101 (apple WP)	WA	11	2
				NY	1 and 7	2
	Deciduous Trees (Plantations and Seed Orchards)	1.0	MRID 44748101 (apple WP)	CA		1
				WA		1
				NY		1
Tree Nuts	Almonds	2.0	MRID 44748101 (almond WP)	CA (arid)	10	1
	Almonds (Dormant and Delayed Dormant)	2.0	MRID 44748101 (almond WP)	CA (arid)	10	1
	Filberts	2.0	MRID 44748101 (pecan EC)	GA	12	0
	Pecans		MRID 44748101 (pecan EC)	GA	2	0
				LA	6	0
				TX	8	0
	Walnuts		MRID 44748101 (pecan EC)	TX	10	0
	Filberts (Dormant and Delayed Dormant)	2.0	MRID 44748101 (pecan EC)	GA	12	0
	Walnuts (Dormant and Delayed Dormant)		MRID 44748101 (pecan EC)	TX	10	0
Ornamentals/ Nurseries (Outdoor Only)	Deciduous Trees in Nurseries and Orchards Except Apples (Dormant and Delayed Dormant) Non-bearing Apple Trees	2.0	MRID 44748101 (apple WP)	CA	10	0
				WA	11	1
				NY	1, 2, 5	1
Ornamentals/ Nurseries (Outdoor Only)	Non-bearing Citrus, Tree Nut and Cherry	4.0	MRID 43062701 (citrus EC)	CA	2, 3, 6, 8, 10, 12	0
	Non-bearing Peach and Nectarine Trees	3.0	MRID 44748101 (apple WP)	CA	10	1
				NY	2	1

Crop Group	Crop	App. Rate (lbs ai/A)	DFR Data Source	DFR Study Location	Growing Region	Estimated REI Range (days) (Dermal LOC = 100)
	Conifers in Nurseries	1.0	MRID 43062701 (citrus EC)	CA	Any	0
Field and Row Crops: Low to Medium (Outdoor Only)	Ornamentals	2.0	MRID 44748102 (sugar beet EC)	CA	Any	0 - 5
				MN	Any	0 - 6
				OR	Any	0 - 2
Vegetable: Root and Tuber	Carrot (Grown for Seed)	0.94	MRID 44748102 (sugar beet EC)	CA	10	0 – 1
				MN	3 and 5	0 – 1
	Radish (Grown for Seed)	0.94	MRID 44748102 (sugar beet EC)	MN	3 and 5	0 – 1
Vegetable: Fruiting	Pepper	1.0	MRID 44748102 (cotton EC)	CA	9 and 10	0 - 2
				MS	2 and 3	0 - 1
				TX	8	0 - 1
Vegetable: Head and Stem Brassica	Broccoli, Brussel Sprout and Cauliflower	1.0	MRID 42974501 (cauliflower WP)	AZ	10	0 - 8
	Cabbage		MRID 42974501 (cauliflower WP)	AZ	1, 2 and 5	
Vegetable: Leafy	Bok Choy	1.0	MRID 42974501 (cauliflower WP)	AZ	3 and 10	0 - 4
	Collards, Kale, Kohlrabi	1.0	MRID 42974501 (cauliflower WP)	AZ	2	
Stalk and Stem: Vegetable	Asparagus	1.0	MRID 44748102 (sugar beet EC)	CA	10	0 – 1
				MN	5	0 - 1
				OR	11	0 - 1
	Non-bearing Pineapple	1.9	MRID 44748102 (cotton EC)	MS	13	0 - 1
Vine/ Trellis	Grapes (Dormant and Delayed Dormant) Grapes (Post-harvest and Prior to Budbreak)	2.0	MRID 43062701 (citrus EC)	CA	10	0
Turf	Turf for Sod and Seed	4.0	MRID 448296-01 (turf EC and WP)	CA	10	1
				IN	5	1
				MS	2 and 6	1
	Turf for Golf Course	1.0	MRID 448296-01 (turf EC and WP)	CA	10	0
				IN	5	0

Crop Group	Crop	App. Rate (lbs ai/A)	DFR Data Source	DFR Study Location	Growing Region	Estimated REI Range (days) (Dermal LOC = 100)
				MS	2 and 6	0
Field and Row Crops: Low to Medium	Soybeans	1.0	MRID 44748102 (sweet corn G)	IL	4 and 5	0
	Sugar Beet	2.0	MRID 44748102 (sweet corn G)	IL	5	0
				OR	10 and 11	0 - 1
	Peanuts	4.0	MRID 44748102 (sweet corn G)	IL	2 and 6	0 - 1
Field and Row Crops: Tall	Corn: Sweet	1.0	MRID 44748102 (sweet corn G)	IL	1 and 5	0
				OR	11	0
	Corn: Field and Grown for Seed	1.0	MRID 44748102 (sweet corn G)	IL	5	0
Nursery	Woody Ornamentals (In Container and Field Grown)	6.0	MRID 44748102 (sweet corn G)	IL	Any	0
				OR		
Turf	Turf for Sod or Seed	1.0	MRID 448296-01 (turf G and fertilizer)	CA	Any	0
	Golf Course					0
Greenhouse (Microencap. Formulations)	Commercial Ornamentals, Greenhouse Production: Bedding Plants, Cut Flowers, Flowering Hanging Baskets, Potted Flowers, Ornamentals, Trees and Shrubs	1.4	MRID 46722702 (smooth ornamentals ME)	MO	Any	0 to > 35
Greenhouse (Total Release Fogger and. Liquid Concentrate Formulations)	Ornamentals	2	MRID 46722701 (hairy ornamentals ME)	MO	Any	18 to > 30
	Commercial Ornamentals, Greenhouse Production: Bedding Plants, Cut Flowers, Flowering Hanging Baskets, Potted Flowers, Ornamentals, Trees and Shrubs	0.01 lb ai/ fogger/ 3,000 sq ft 0.15 lb ai/A	MRID 46722701 (hairy ornamentals ME)	MO	Any	2 - 22

Characterization of Occupational Post-Application Risk Assessment

The occupational post-application exposure and risk assessment for chlorpyrifos is based upon an array of calculations completed for 15 different crop groups. These unique crop groupings are defined essentially based on the nature of the crop where a work activity would take place. Within each of these groupings, ranges of transfer coefficients were considered to reflect differences in exposures that would be associated with the variety of cultural practices required to produce the crop/product. Transfer coefficients are used "generically" to allow for estimation of exposure for any pesticide active ingredient using estimates for exposure time and the concentration of residue the workers will contact that is specific to the pesticide of interest. The Agency has adopted a method of clustering groups, crop growth stages, and post-application activities into groups that are expected to result in comparable exposure. Chlorpyrifos post-application exposures were estimated over subsequent days after application to reflect residue dissipation over time in the environment and to allow for a more informed risk management decision.

The exposure data used in the chlorpyrifos post-application exposure and risk assessment represent the best data and approaches that are currently available. The latest HED transfer coefficients have been used to complete the assessment, as referenced from the Science Advisory Council for Exposure (ExpoSAC) Policy Number 3.2: Agricultural Transfer Coefficients (5/5/11). Most of the transfer coefficient values in Policy 3.2 are based on the work of the Agricultural Reentry Task Force (ARTF). The choice of post-application activities studied by the ARTF, as well as the subsequent assignment of transfer coefficients derived from these studies to non-monitored post-application activities was developed with input from both the ARTF and HED staff Agency and reviewed by the FIFRA SAP in 2008. It is possible that there are exposure scenarios not addressed by HED either due to the lack of appropriate exposure data or because the transfer coefficient model is not appropriate or little or no foliar contact is associated with a specific activity. Furthermore, unlike the vast majority of crop-activity combinations listed under the Transfer Coefficient Table in Policy 3.2, some common agricultural activities do not follow the standard "foliar-based" transfer coefficient methodology. This should not be interpreted to mean that there is no potential exposure from these activities but rather that "foliar-based" transfer coefficients are not applicable to evaluate worker exposure. For example, the crop-activity combinations of mechanical windrowing, mechanical sweeping and dormant hand pruning for the "nut tree" crop grouping are standard cultural practices; however, do not follow the standard transfer coefficient methodology.

HED completed the assessment of occupational post-application exposure and risk to chlorpyrifos using 7 chemical-specific DFR studies submitted by the registrant in support of the re-registration of chlorpyrifos. The studies, which encompass the use of five different formulations and twelve different crops, have been extrapolated to other groups based on the nature of the crop and application method and used to calculate risks for post-application workers in every region of the county. It is standard practice for the Agency to use these kinds of studies in this manner. Furthermore, it is possible that the use of the 7 chemical specific DFR studies to represent all crops and regions within the country could lead to results that do not reflect actual use practices and conditions in some parts of the country. Furthermore, the extrapolation of DFR data from one crop may not represent precisely the dissipation of another.

For example, DFR data which measured the dissipation of chlorpyrifos from cotton after application of an emulsifiable concentrate were used to represent dissipation of chlorpyrifos from soybeans which like cotton, are classified in the low/medium field row crop grouping. HED assumes that residue dissipation monitored in available studies approximates residues from like crops, but the extent that these residues might be an under- or over-estimate is unknown. Finally, DFR data for several crops were conducted in multiple states reflective of the regions of the country where the crops are typically grown and chlorpyrifos is used. HED has presented all state-specific DFR data for each crop under the assumption that these data accurately reflect dissipation anticipated in the different regions of the country (e.g., the subtropical Southeastern U.S. and the semi-arid climate of the Central Valley of California). HED has considered available use and usage information in development of the occupational post-application assessment and has refined the use of available region-specific DFR data to those areas of the U.S. where chlorpyrifos usage occurs.

In summary, the Agency believes that the risk values presented in this post-application assessment represent the highest quality results that could be produced given the exposure, use, and toxicology data that are available. Risk managers and other interested parties should consider the quality of individual inputs when interpreting the results and make decisions accordingly. It is difficult to determine where on a distribution the values which have been calculated fall because the distributional data for exposure, residue dissipation and many other parameters are unrefined. The Agency does believe, however, that the risks represent conservative estimates of exposure because maximum application rates are used to define residue levels upon which the risk calculations are based and most maximum application rates exceed what is assumed to be typical.

9.2.2 Inhalation Post-application Risk

There are multiple potential sources of post-application inhalation exposure to individuals performing post-application activities in previously treated fields. These potential sources include volatilization of pesticides and resuspension of dusts and/or particulates that contain pesticides. The Agency sought expert advice and input on issues related to volatilization of pesticides from its Federal Insecticide, Fungicide, and Rodenticide Act Scientific Advisory Panel (SAP) in December 2009, and received the SAP's final report on March 2, 2010 (http://www.epa.gov/scipoly/SAP/meetings/2009/120109meeting.html). The Agency is in the process of evaluating the SAP report as well as available post-application inhalation exposure data generated by the Agricultural Reentry Task Force and may, as appropriate, develop policies and procedures, to identify the need for and, subsequently, the way to incorporate occupational post-application inhalation exposure into the Agency's risk assessments.

However, based on the Agency's current practices, a quantitative post-application inhalation exposure assessment is not typically performed for a chemical when it is characterized by low acute inhalation toxicity (Toxicity Category III and IV) and low vapor pressure. Chlorpyrifos does not fit into these categories as it is classified as Toxicity Category II for inhalation toxicity and has a moderate vapor pressure of 1.9×10^{-5} mm Hg at 25º C. The inhalation exposure potential from occupational/commercial post-application activities may be elevated based upon these criteria. A quantitative occupational post-application inhalation exposure assessment was

not performed for chlorpyrifos; however, an inhalation exposure assessment was performed for occupational/commercial handlers. It is expected that while many of these handler inhalation exposure estimates are of concern to HED, exposure and risk from occupational post-application inhalation would be of no greater concern than occupational handler inhalation estimates.

Chlorpyrifos can be used in indoor facilities as well as agricultural/commercial outdoor uses. Indoor use sites for chlorpyrifos include greenhouse use, indoor commercial uses (e.g., warehouses, indoor industrial sites) and commercial seed treatment facilities. HED has not assessed post-application inhalation exposure for greenhouses due to requirements for high air exchange rates and ventilation regulations. The Worker Protection Standard for Agricultural Pesticides contains requirements for protecting workers from inhalation exposures during and after greenhouse applications through the use of ventilation requirements [40 CFR 170.110, (3) (Restrictions associated with pesticide applications)]. Furthermore, HED assumes that commercial applicators do not typically return to the treated areas after an indoor use site pesticide application and thus an occupational post-application inhalation exposure assessment was not performed for commercial applicators. Seed treatment assessments provide quantitative inhalation exposure assessments for seed treaters and secondary handlers (i.e. planters). It is expected that these exposure estimates would be protective of most post-application inhalation exposure scenarios.

9.2.3 ORE Evaluation of Chlorpyrifos-oxon

HED has considered the exposure potential for occupational and residential exposure to chlorpyrifos-oxon. Workers re-entering an environment previously treated with chlorpyrifos (occupational post-application) and the general population residing near chlorpyrifos application sites (bystanders) could potentially be exposed to the oxon as chlorpyrifos is metabolized in the environment. Dermal exposure to the oxon could occur through contact with chlorpyrifos treated surfaces and inhalation exposure through airborne oxon. However, the likelihood of exposure to the oxon is slight due to its rapid deactivation to TCP in the environment. In an effort to further explore the potential for oxon exposure, HED has researched and reviewed all available information sources. Chlorpyrifos and chlorpyrifos-oxon were measured in several air monitoring studies. A search of open literature resulted in 4 metabolism studies which measured whole fruit and leaf surface residue of chlorpyrifos and chlorpyrifos-oxon. [See W. Britton, 6/27/11, D388165, *Chlorpyrifos: Occupational and Residential Exposure Assessment,* Section 4 for a full discussion of study results and conclusions.]

The Agency has considered the potential for occupational and residential exposure to chlorpyrifos-oxon. Workers re-entering an environment previously treated with chlorpyrifos (occupational post-application) and the general population residing near chlorpyrifos application sites (bystanders) could potentially be exposed to the oxon as chlorpyrifos is degraded in the environment. Dermal exposure to the oxon could occur through contact with chlorpyrifos treated surfaces and inhalation exposure through airborne oxon. However, the likelihood of exposure to the oxon is slight due to its rapid deactivation to TCP (3,5,6-trichloro-2-pyridinol). In an effort to further explore the potential for oxon exposure, EPA has researched and reviewed all available information sources. Based upon this review, EPA intends to require additional studies to

address uncertainties regarding the formation of chlorpyrifos-oxon in the air post-application and its formation and decay in greenhouses.

Dermal exposure to the oxon on foliar surfaces from reentry into an outdoor environment previously treated with chlorpyrifos is not anticipated and, therefore, has not been assessed. However, HED is concerned, based on study results, that the formation of the oxon may be greater and its deactivation slower in greenhouses when compared to the outdoor environment and that an assessment may be needed for exposure to the oxon in greenhouse settings. In order address these uncertainties and more accurately address the risk potential for exposure from occupational reentry into greenhouses treated with chlorpyrifos, HED requires a study to measure chlorpyrifos and oxon residues on leaf surfaces following treatment with a liquid formulation of chlorpyrifos in greenhouses.

9.2.4 Comparison of the Chlorpyrifos 2000 Risk Assessment and 2011 Preliminary Risk Assessment

Table 28 and Table below present a range of resulting occupational handler risk estimates (MOEs) for both the current preliminary (2011) chlorpyrifos assessment and the June 2000 chlorpyrifos assessment for comparison purposes. The range represents a low, medium, and high exposure scenario. Also presented is a range of personal protection (single layer/gloves, double layer/gloves, and engineering controls). Table 28 shows the short-term and intermediate –term dermal risk estimates and Table 29 shows the short-term and intermediate –term inhalation risk estimates.

The dermal handler risk estimates remain unchanged between the 2000 and 2011 assessments since the dermal PoD is the same (NOAEL of 5 mg/kg/day from a dermal study). The 2008 SAP concurred with the selection of this PoD for assessing dermal scenarios.

The inhalation PoD in 2000 was 0.1 mg/kg/day (NOAEL based on inhalation studies). That same PoD is used in the current assessment except that it has been converted to an HEC (human equivalent concentration). This resulted in the reduction of the default database uncertainty factor for interspecies extrapolation from a 10x to a 3x. Thus the level of concern MOE for this assessment is 30 (compared to 100 in 2000). In addition the NOAEL was corrected to account for an 8 hour workday because worker exposure is expected to occur during the course of an average workweek (8 hours/day and 5 days/week; animals were exposed 6 hours a day in the study). The inhalation handler risk estimates have changed since the 2000 assessment. This can be mainly attributed to the use of the HEC in the preliminary assessment.

Note that the actual dermal and inhalation MOEs presented in the 2000 assessment may differ somewhat than those presented here since some of the exposure assumptions used today may vary due to refinements made since 2000. The 2011 exposure assumptions were compared to the 2000 PoD for illustrative purposes only.

Table 28 Comparative Analysis of Occupational Handler Exposure Estimates Considering 2000/2011 Dermal PoDs Using Low, Medium and High Level Representative Scenarios

Level	Exposure Scenario	Target	App. Rate[a] (lb ai/A)	Level of Personal Protection – Risk Estimates (MOE)		
				Single Layer[1], Gloves	Double Layer[2], Gloves	Engineering Control
Risk Estimates with 2011 Assessment Dermal PoD (5 mg/kg/day)						
Low	Groundboom Applications	Non-Crop Areas (Industrial Plant Sites, Road Medians, and Sod Farms), Turfgrass	0.25	1,100	1,400	3,400
Medium	Mixing/Loading Liquids for Groundboom Application	Asparagus, Brussel Sprouts, Sugarbeet	1.0	120	150	510
High	Airblast Applications	Citrus (CA and AZ)	6.0	6	11	77
Risk Estimates with 2000 Assessment Dermal PoD (5 mg/kg/day)						
Low	Groundboom Applications	Non-Crop Areas (Industrial Plant Sites, Road Medians, and Sod Farms), Turfgrass	0.25	1,100	1,400	3,400
Medium	Mixing/ Loading Liquids for Groundboom Application	Asparagus, Brussel Sprouts, Sugarbeet	1.0	120	150	510
High	Airblast Applications	Citrus (CA and AZ)	6.0	6	11	77

1. Single layer (long-sleeve shirt, long pants, shoes, socks), chemical resistant gloves
2. Double layer (single layer clothing with the addition of coverall), chemical resistant gloves

Table 29 Comparative Analysis of Occupational Handler Exposure Estimates Considering 2000/2011 Inhalation PoDs Using Low, Medium and High Level Representative Scenarios

Level	Exposure Scenario	Target	App. Rate[a] (lb ai/A)	Level of Personal Protection – Risk Estimates (MOE)			
				No Respirator	PF5[1] Respirator	PF10[2] Respirator	Engineering Control
Risk Estimates with 2011 Assessment Inhalation PoD (0.56 mg/kg/day) – LOC is an MOE = 30							
Low	Groundboom Applications	Non-Crop Areas (Industrial Plant Sites, Road Medians, and Sod Farms), Turfgrass	0.25	5,800	29,000	58,000	46,000
Medium	Mixing/ Loading Liquids for Groundboom Application	Asparagus, Brussel Sprouts, Sugarbeet	1.0	2,200	11,000	22,000	5,900
High	Airblast Applications	Citrus (CA and AZ)	6.0	36	180	360	1,800
Risk Estimates with 2000 Assessment Inhalation PoD (0.1 mg/kg/day) – LOC is an MOE =100							
Low	Groundboom Applications	Non-Crop Areas (Industrial Plant Sites, Road Medians, and Sod Farms), Turfgrass	0.25	1,100	5,100	10,000	8,100
Medium	Mixing/ Loading Liquids for Groundboom Application	Asparagus, Brussel Sprouts, Sugarbeet	1.0	400	2,000	4,000	1,100
High	Airblast Applications	Citrus (CA and AZ)	6.0	6	32	65	320

1. Single layer (long-sleeve shirt, long pants, shoes, socks), chemical resistant gloves
2. Double layer (single layer clothing with the addition of coverall), chemical resistant gloves

10.0 Incident Report

One component of the Agency's registration review program is consideration of human observational information including incident data, medical case reports, general medical information, biomonitoring data, and epidemiology studies. In conjunction with a human health risk assessment based on other data sources, such human incident and other human data can assist the Agency in better defining and characterizing the risk of pesticides/pesticide products. Based on the frequency and the effects noted in the Agency's earlier scoping or Tier I incident assessment (Hawkins M. and Cordova J., 10/15/2008), the Agency determined that chlorpyrifos human incident data are an important source of information to consider in its updated chlorpyrifos risk assessment.

HED has prepared a chlorpyrifos incident report review (S. Recore *et al*., 6/27/11, D388406, *Chlorpyrifos: Tier II Incident Report*). The review considers a variety of types and sources of human observational information including human incident data, medical data/case report information, and epidemiological information in an effort to inform the re-evaluation of chlorpyrifos in this phase of registration review. The human incident databases that were reviewed are:

- the OPP Incident Data System (IDS);
- the National Pesticide Information Center (NPIC);
- NIOSH's Sentinel Event Notification System for Occupational Risks (SENSOR);
- the California Pesticide Illness Surveillance Program Incident Data (CA PISP).

Together, these databases indicate that the number of incidents associated with chlorpyrifos declined post-2002, correlating well with the phase out/cancellation of the almost all chlorpyrifos residential products in December 2001. In addition, the Agency's findings are consistent with other incident cases investigations of American Association of Poison Control Centers **(**AAPCC) data which have reported a decrease in the number of chlorpyrifos incidents that is temporally associated with the phase out/cancellation of most residential chlorpyrifos products.

While the chlorpyrifos incidents are reported to have declined substantially (95%) among residential users from 2002 to 2010, it is unclear if occupational incidents have also decreased. Specifically, chlorpyrifos occupational incidents, reported in CA PISP and SENSOR databases, appear to be constant over time, despite risk mitigation implemented including reduced application rates and seasonal maximum limits, increased retreatment intervals, increased PPE and/or use of engineering controls which were required as well as increased reentry intervals (REIs) for a number of crops. However, a number of these incidents appear to be due to accidents and misuse. Overall, the NIOSH SENSOR database indicated that the largest number of incidents are exposures due to actual application of chlorpyrifos, but California PISP data suggests that drift of chlorpyrifos to adjacent fields appears to be the largest contributor to occupational exposure. OPP will continue to monitor these incidents and remain alert for any changes in trend or patterns.

In addition to the incident/poisoning data and medical case reports, epidemiological research can be an important source for human observational data and can potentially assist in identifying, characterizing, and (ideally) quantifying linkages between human exposures and resulting health effects. For chlorpyrifos, epidemiological data is available from both the Agricultural Health Study (AHS) and from a variety of university-based research groups. While the AHS investigations currently published were hypothesis-generating in nature, initial strength and consistency in the findings for lung cancer and colorectal cancer are notable, and warrant further follow-up and additional research. Preliminary associations with breast and prostate cancer are weak, but also warrant monitoring the literature for additional publications on this association. There is no compelling evidence of an association with other cancer sites including pancreatic cancer, melanoma, brain, esophageal, kidney, all lymphohematopoietic cancers combined and NHL, leukemia, and multiple myeloma (C. Christensen, 6/16/11, D388167).

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Appendix A. Toxicology Profile and Executive Summaries

A.1 Toxicology Data Requirements

The requirements (40 CFR 158.340) chlorpyrifos are in the table below. Use of the new guideline numbers does not imply that the new (1998) guideline protocols were used.

Study	Technical	
	Required	Satisfied
870.1100 Acute Oral Toxicity	yes	yes
870.1200 Acute Dermal Toxicity	yes	yes
870.1300 Acute Inhalation Toxicity	yes	yes
870.2400 Primary Eye Irritation	yes	yes
870.2500 Primary Dermal Irritation	yes	yes
870.2600 Dermal Sensitization	yes	yes
870.3100 Oral Sub-chronic (rodent)	yes	yes
870.3150 Oral Sub-chronic (non-rodent)	yes	yes
870.3200 21-Day Dermal	yes	yes+
870.3250 90-Day Dermal	CR	--
870.3465 90-Day Inhalation	CR	yes
870.3700a Developmental Toxicity (rodent)	yes	yes
870.3700b Developmental Toxicity (non-rodent)	yes	yes
870.3800 Reproduction	yes	yes
870.4100a Chronic Toxicity (rodent)	yes	yes
870.4100b Chronic Toxicity (non-rodent)	yes	yes
870.4200a Oncogenicity (rat)	yes	yes
870.4200b Oncogenicity (mouse)	yes	yes
870.4300 Chronic/Oncogenicity	yes	yes
870.5100 Mutagenicity—Gene Mutation - bacterial	yes	yes
870.5300 Mutagenicity—Gene Mutation - mammalian	yes	yes
870.5375 Mutagenicity—Structural Chromosomal Aberrations	yes	yes
870.5395 Mutagenicity—Other Genotoxic Effects (MN Assay)	yes	yes
870.6100a Acute Delayed Neurotoxicity (hen)	yes	yes
870.6100b 90-Day Neurotoxicity (hen)	yes	yes
870.6200a Acute Neurotoxicity Screening Battery (rat)	yes	yes
870.6200b 90-Day Neurotoxicity Screening Battery (rat)	yes	yes
870.6300 Developmental Neurotoxicity	yes	no, but upgradeable*
870.7485 General Metabolism	yes	yes
870.7600 Dermal Penetration	CR	yes
870.7800 Immunotoxicity	yes	yes
Special Studies Comparative Cholinesterase Assay	yes	yes

CR: Conditionally Required, *Performed, But Guideline-Unacceptable Rating, Upgradeable if additional morphometric data is submitted
+Satisfied Guideline 82-2, but not 870.3200 since N<10/sex

A.2 Toxicity Profiles

Table A.2.1 Acute Toxicity Profile - Test Substance

Guideline No.	Study Type	MRID(s)	Results	Toxicity Category
870.1100	Acute Oral LD50 - rat	44209101	223 mg/kg M&F	II
870.1200	Acute Dermal LD50 - rat Acute Dermal LD50 - rabbit	Accession No. 112115 44209102	202 mg/kg >5000 mg/kg	II, IV
870.1300	Acute Inhalation LC50; rat Supplementary	00146507 and Accession No. 257590	LC50 > 0.2 mg/L (200 mg/m3) (nominal concentration)	II
870.2400	Eye Irritation - rabbit	44209103	slight irritation resolved within 24 hours	IV
870.2500	Dermal Irritation - rabbit	44209104	mild irritant; (irritation resolved within 7 days)	IV
870.2600	Dermal Sensitization - guinea pig	44209105	non-sensitizing	NA

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.3100	90-Day Oral Toxicity (Rat)	MRID #: 40436406 Acceptable/guideline 0, 0.025, 0.5, or 10 mg/kg/day (0, 0.5, 10 or 200 ppm)	95.5% a.i. chlorpyrifos NOAEL ChEI: none for plasma ChEI due to reductions in male plasma enzymes at 0.025 mg/kg/day LOAEL ChEI: 0.025 mg/kg/day (significant 22%↓ in plasma ChE activity that was dose-related) NOAEL (systemic): 0.5 mg/kg/day LOAEL (systemic): 10 mg/kg/day Effects: decreased weight gain and slight decreases in packed cell volume, red cells and hemoglobin Note: Female ChEI data is unreliable due to a possible reporting error. RBC and brain ChE activity were not measured.
870.3100	90-Day Oral Toxicity (Rat)	MRID #: 40952801 Acceptable/guideline 0, 0.1, 1, 5 or 15 mg/kg/day	95.7 - 98.5% a.i. chlorpyrifos NOAEL: 0.1 mg/kg/day (plasma and RBC ChEI) LOAEL: 1 mg/kg/day (significant plasma and RBC ChEI in both sexes) Effects: increased organ weights (brain and heart), and reduced weight gain at 15 mg/kg/day and increased adrenal gland vacuolation and significant brain ChEI in both sexes 5 and 15 mg/kg/day.
870.3150	Sub-chronic Oral (capsule) in Beagle Dogs	MRID #: 42172801 Acceptable/guideline 0, 0.01, 0.22, or 5 mg/kg/day	95.8% a.i. chlorpyrifos NOAEL: 0.01 mg/kg/day LOAEL: 0.22 mg/kg/day (significant 33-67% ↓ plasma and 24-46% ↓ RBC ChEI) Effects: Brain ChEI (46% ↓) occurred at 5 mg/kg/day. Comments: At 0.01 mg/kg/day, plasma ChEI noted in females (significant 20-24% at week 6, and non-significant 24% at week 12) and males (15% at week 13) that was not considered of sufficient magnitude and consistency to be biologically and toxicologically meaningful.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.3200	21-Day Dermal Toxicity Study in Rats	MRID # 40972801 Satisfies Guideline 82-2, but has less than 10 animals/sex for 870.3200 0, 0.1, 0.5, 1 or 5 mg/kg/day (21 day study) 0, 1, 10, 100 or 500 mg/kg/day (4-day dermal probe study)	100% pure chlorpyrifos NOAEL: 5 mg/kg/day (plasma and RBC ChEI) LOAEL: 10 mg/kg/day (45% plasma and 16% RBC ChEI following 4 days of exposure) NOAEL (systemic): 5 mg/kg/day LOAEL (systemic): Not Identified Effects: Slight erythema in 2/4 females at 1 and 10 mg/kg/day, respectively. 4-day Dermal Probe Study as well
870.3465	90-Day, Sub-chronic Inhalation in Rats (nose only)	MRID # 40013901 & 40166501 Acceptable/guideline 0, 5.2, 10.3 or 20.6 ppb (0, 72, 143 or 287 µg/m^3) (maximum dose equivalent to 0.044-0.082 mg/kg/day)	100% pure chlorpyrifos NOAEL: not identified (ChEI and systemic) LOAEL: not identified at highest attainable vapor concentration (>20.6 ppb or >0.082 mg/kg/day or >287 µg/m^3) (ChEI and systemic)
870.3465	90-Day, Sub-chronic Inhalation in Rats (nose only)	MRID # 40908401 Acceptable/guideline 0, 5, 10 or 20 ppb (0, 70, 143 or 287 µg/m^3) (equivalent to 0, 0.024, 0.048 or 0.097 mg/kg/day, respectively)	95% a.i. chlorpyrifos NOAEL: not identified (ChEI and systemic) LOAEL: not identified at highest attainable vapor concentration (>20 ppb or 0.097 mg/kg/day) (ChEI and systemic)
870.3700a	Developmental Study in CD rats (gavage)	MRID# 40436407 Acceptable/guideline 0, 0.5, 2.5 or 15 mg/kg/day (gestation day 6-15)	96.1% a.i. chlorpyrifos Maternal NOAEL: none observed for plasma ChEI; 2.5 mg/kg/day for systemic Maternal LOAEL: 0.5 mg/kg/day (decreased plasma ChEI); 15 mg/kg/day (systemic) based on decreased food consumption (only the first few days of dosing) and body weight during dosing. Developmental NOAEL: 2.5 mg/kg/day Developmental LOAEL: 15 mg/kg/day (HDT) based on an increase in post-implantation loss. Comments: RBC and brain ChE were not measured.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.3700a	Developmental Study in F344 rats (gavage)	MRID# 00130400 Acceptable/guideline 0, 0.1, 3, or 15 mg/kg/day (gestation day 6-15)	96.6% a.i. chlorpyrifos Maternal NOAEL: 0.1 mg/kg/day (plasma and RBC ChEI) Maternal LOAEL: 3 mg/kg/day (90.3% plasma and 74.3% RBC ChEI) Developmental NOAEL: 15 mg/kg/day (HDT) Developmental LOAEL: Not Identified
870.3700a	Developmental Study in CF-1 Mice (gavage)	MRID# 00095268 Unacceptable/Non-guideline 0, 0.1, 1, 10, or 25 mg/kg/day (gestation day 6-15)	96.8% a.i. chlorpyrifos Maternal NOAEL: 0.1 mg/kg/day (plasma and RBC ChEI); 10 mg/kg/day (systemic toxicity) Maternal LOAEL: 1 mg/kg/day (plasma and RBC ChEI); 25 mg/kg/day (systemic toxicity) based on decreased body weight, food and water consumption, and increased mortality. Developmental NOAEL: 1 mg/kg/day (plasma and RBC ChEI); 10 mg/kg/day for systemic toxicity Developmental LOAEL: 10 mg/kg/day (plasma and RBC ChEI); 25 mg/kg/day (systemic toxicity) based on minor skull variations, delayed ossification of skull bones and sternebrae and reduced fetal body length. Comments: Brain ChE not measured.
870.3700b	Developmental Study in New Zealand rabbits (gavage)	MRID# 40436408 Acceptable/guideline 0, 1, 9, 81, or 140 mg/kg/day (gestation day 7-19)	96.1% a.i. chlorpyrifos Maternal NOAEL: none observed for plasma ChEI; 81 for systemic toxicity Maternal LOAEL: 1 mg/kg/day (plasma ChEI); 140 for systemic toxicity based on reduced food consumption, body weight loss, and apparent post-implantation loss. Developmental NOAEL (systemic): 81 mg/kg/day Developmental LOAEL (systemic): 140 mg/kg/day based on slightly decreased fetal weights and crown-rump lengths, and an increased incidence of unossified xiphisternum and/or 5th sternebra.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.3800	2-Generation Reproduction Toxicity in SD Rats	MRID# 41930301 Acceptable/guideline 0, 0.1, 1, or 5 for 10 mg/kg/day (F0) or 12 (F1) weeks prior to mating, through lactation and weaning	97.8-98.5% a.i. chlorpyrifos Parental NOAEL: 0.1 mg/kg/day Parental LOAEL: 1 mg/kg/day (significant 43-59% plasma, and 65-69% RBC ChEI at 1 mg/kg/day; and 48-49% brain ChEI and histological lesions of the adrenal gland at 5 mg/kg/day). Reproductive NOAEL: 1 mg/kg/day Reproductive LOAEL: 5 mg/kg/day (HDT) based on reduced pup weight and increased pup mortality in F1 generation only.
870.3800	3-Generation Reproduction Toxicity in SD Rats	MRID # 00029064, 00064934 Acceptable/guideline 0, 0.03, 0.1,or 0.3 mg/kg/day for first generation, and 0.1, 0.3 or 1 mg/kg/day for second and third generation	Parental NOAEL: 0.1 mg/kg/day Parental LOAEL: 0.3 mg/kg/day (plasma and RBC ChEI) Reproductive NOAEL: 1 mg/kg/day (HDT) Reproductive LOAEL: Not Identified
870.3800	Reproduction Study in Rats	MRID# 00130401 Acceptable in combination with studies 00029064 & 00064934 0, 0.5, 0.8, 1.2 mg/kg/day in Sprague-Dawley Rats	NOAEL Neonatal Survival: 1.2 mg/kg/day (Primary purpose of the study) NOAEL Reproduction: 1.2 mg/kg/day NOAEL General Toxicity: 0.8 mg/kg/day LOAEL General Toxicity: 1.2 mg/kg/day based on decreased weight gain in males
870.4100a	Chronic feeding study in CD-1 mice (2 yrs)	MRID # 00054352 & 00142902 (Accession No. 242059) Acceptable/guideline 0, 0.5, 5 or 15 ppm (highest dose tested is 2.25 mg/kg/day)	99.6% a.i. chlorpyrifos LOAEL: 2.25 mg/kg/day (90%↓plasma, and 50%↓ RBC ChE activity relative to controls after 1 week) NOAEL(systemic) = 2.25 mg/kg/day LOAEL (systemic): Not Determined Effects: no systemic effects observed at highest dose tested (HDT). No treatment-related tumors. ChE only measured at 15 ppm (2.25 mg/kg/day) after 1 and 4 weeks.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.4100b	Chronic feeding study in beagle dogs (2 yrs)	MRID # 00064933 & 00146519 Acceptable/guideline 0, 0.01, 0.03, 0.1, 1 or 3 mg/kg/day	97.2-98.8% a.i. chlorpyrifos NOAEL: 0.01, 0.03, & 1 mg/kg/day for plasma, RBC and brain ChEI, respectively LOAEL (plasma ChEI): 0.03 mg/kg/day (mostly significant mean of 23-29% ↓ at 1 year and 10-24% ↓ at 2 years) LOAEL (RBC ChEI): cannot be established due to data quality issues LOAEL (brain ChEI): 3 mg/kg/day (19.4-20.8% ↓ at 2 yr) NOAEL (systemic): 1 mg/kg/day LOAEL (systemic): 3 mg/kg/day Effects: increased absolute and relative liver weights that could be an adaptive response
870.4200a	Carcinogenicity /chronic feeding study in F344 rats (2 yrs)	MRID # 42172802 Acceptable/guideline Males: 0, 0.0132, 0.33 or 6.99 mg/kg/day Females: 0, 0.0146, 0.365 or 7.78 mg/kg/day (0, 0.2, 5 or 100 ppm)	96.1% a.i. chlorpyrifos NOAEL:0.0132 mg/kg/day LOAEL: 0.33 mg/kg/day (significant 15-51% plasma ChEI in both sexes, 19-31% RBC ChEI at 104 weeks vs. controls and 11-17% RBC ChEI vs. vehicle controls) NOAEL (systemic):0.33 mg/kg/day LOAEL (systemic): 6.99 mg/kg/day Effects: decreased body weights in males and females, and cataracts, and diffuse retinal atrophy in females. No evidence of carcinogenicity.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.4200a	Carcinogenicity /chronic feeding study in F344 rats (2 yrs)	MRID # 40952802 Acceptable/guideline 0, 0.05, 0.1, 1 or 10 mg/kg/day	Lorsban 98.5% pure NOAEL: 0.1 mg/kg/day (plasma and brain ChEI) LOAEL: 1 mg/kg/day (significant 39-86% plasma, 14-34% RBC and 5-9% brain ChEI) NOAEL (systemic): 1 mg/kg/day LOAEL (systemic): 10 mg/kg/day Effects: decreased body weight gain, red blood cells, hemoglobin, cholesterol, protein, and globulin, and increased platelets and specific gravity, increased adrenal gland weight, and fatty vacuolation of the zona fasciculata. No evidence of carcinogenicity.
870.4200b	Carcinogenicity/ chronic feeding study in CD-1 mice (78 weeks)	MRID # 42534201 Acceptable/guideline Males: 0, 0.89, 8.84, 45.2 mg/kg/day Females: 0, 0.938, 9.79, or 48.1 mg/kg/day (0, 5, 50 or 250 ppm)	95.5% a.i. chlorpyrifos NOAEL: none for ChEI LOAEL: 0.89 males; 0.938 females mg/kg/day (significant 45-51% plasma ChEI in both sexes) NOAEL (systemic): 8.84 males, 9.79 females mg/kg/day (50 ppm) LOAEL (systemic): 48.1 females, 45.2 males mg/kg/day (HDT; 250 ppm) Effects: decreased body weight gain and food consumption in males, decreased water consumption in females, increased incidences of keratitis and hepatocyte fatty vacuolation, and increased incidence of gross clinical findings (ocular opacity and hair loss) in both sexes. Brain cholinesterase was inhibited at the high dose in both sexes. No evidence of carcinogenicity. Brain ChEI at high dose. Note: The validity of the RBC ChE assay is questionable.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.5100	Gene Mutation Bacterial Cell (Ames Reversion)	MRID# 00157058 and 40436411 Acceptable/guideline Tested in Salmonella strains TA 98, TA 100, TA 1535, TA 1537 and TA 1538 in the presence and absence of S-9 at concentrations of 30, 100, 300, 1000, 3000 and 10000 µg/plate	Negative for reverse mutations Positive controls caused appropriate mutagenic responses.
870.5300	Gene Mutation Mammalian Cell (CHO Cells/HGPRT)	MRID# 00152683 and 40436410 Acceptable/guideline Tested for gene mutation potential at 0, 10, 20, 25, 30, 40 & 50 µM in mammalian cells	Negative for reverse mutations Cytotoxic at 10 µM and above without metabolic activation and no toxicity with activation. Precipitate formed at 30 µM and higher concentrations with or without activation.
870.5375	In vitro Cytogenetics	MRID# 40436409, 44533401 Acceptable/guideline Concentrations assayed were as follows with non-activation in the 10 hour assay at 1.56, 3.12, 5.2, 10.4, 15.6, 31.2, 52, 104 & 156 µg/ml and in the 19-20 hour assay at 0.975, 1.47, 2.93, 4.89, 9.75, 14.7, 29.3, 48.9, 97.5 & 147 µg/ml. Concentrations tested with activation in the two 10 hour assays were 1, 1.5, 3, 5, 10, 15, 30, 50 & 100 µg/ml and 2.95, 4.95, 9.85, 14.8, 29.6, 49.4, 98.5 & 296 µg/ml, plus concentrations for the 19-20 hour assay were 9.75, 14.7, 29.3, 48.9, 97.5, 147 & 293 µg/ml.	Negative for chromosome aberrations Cytotoxicity was shown in both non-activated as well as in activated assays. Positive controls mitomycin C (for non-activation) and cyclophosphamide (for activation) caused the appropriate mutagenic responses.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.5395	Micronucleus Assay in Mammalian Erythrocytes	MRID# 00152684 Acceptable/guideline Tested at levels of 0, 7, 22, 70 mg/kg by gavage in corn oil in the mouse	Not clastogenic
870.5500	DNA Repair Assay in Bacteria	Accession# 256040 Acceptable/guideline	Increased damage to DNA was detected
870.5550	Unscheduled DNA Synthesis in Rat Hepatocytes	MRID# 00157057 Acceptable/guideline Tested with concentrations from 10E-06 M to 10E-04 M in isolated rat hepatocytes	Negative for induction of UDS The high dose was cytotoxic and also formed a precipitate.
870.5575	Mitotic Gene Conversion in Yeast	Accession# 256040 Acceptable/guideline	Increased recombination frequency detected
870.6100a	Acute Delayed Neurotoxicity Study in Hens	MRID# 00097144 and 40510601 Acceptable/guideline 0, 50, 100 or 110 mg/kg	96.8% a.i. chlorpyrifos NOAEL: 110 mg/kg (HDT) LOAEL: Not Determined Not neurotoxic
870.6200a	Acute Neurotoxicity Study in Rats	MRID 42669101 and 42943101 Acceptable/guideline 0, 10, 50 or 100 mg/kg	98.2% a.i. chlorpyrifos NOAEL (systemic): 10 mg/kg LOAEL (systemic): 50 mg/kg Effects: Decreased body weight, and motor activity and increased incidence of adverse clinical signs

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.6200b	13-Week Rat Neurotoxicity Study in Rats	MRID 42929801 Acceptable/guideline 0, 0.1, 1, 5, or 15 mg/kg/day	98.2% a.i. chlorpyrifos NOAEL (systemic): ≥15 mg/kg/day LOAEL (systemic): none established Effects: Decreased motor activity and an increased incidence of urine incontinence in females. Note: This study did not measure cholinesterase activity.
870.6300	Developmental Neurotoxicity in Rats	MRID: 44556901 Guideline-Unacceptable, But Upgradeable 0, 0.3, 1, or 5 mg/kg/day (gestation day 6 through lactation day 11)	99.8% a.i. chlorpyrifos Maternal NOAEL: none observed for plasma or RBC ChEI Maternal LOAEL: ≤0.3 mg/kg/day (43%↓ plasma and 41%↓% RBC ChE activity relative to controls) Note: Submission of further morphometric data may upgrade the study.
	Cholinesterase and Metabolite Determination Study in Rats (Companion Study of the Developmental Neurotoxicity Study)	MRID# 44648101 Acceptable/Non-Guideline 0, 0.3, 1, or 5 mg/kg/day (gestation day 6 through lactation day 11)	99.8% a.i. chlorpyrifos Maternal Effects: Dams in the 0.3 mg/kg/day group exhibited a 33%↓ plasma and 26↓% RBC ChE activity relative to controls Developmental Effects: Pups in the 5 mg/kg/day group exhibited an 85%↓ plasma, 92↓% RBC, 82%↓ heart and 60%↓ brain ChE activity relative to controls Note: This is a pharmacokinetic study, and therefore, NOAELs and LOAELs were not identified.
870.7485	Acute Pharmacokinetic Study in Rats	MRID 44648102 Acceptable/Non-Guideline 0.5, 1, 5, 10, 50, 100 mg/kg	89.4-99.8% a.i. chlorpyrifos NOAEL: 0.5 mg/kg LOAEL: 1 mg/kg (28-40% plasma ChEI at the peak time of inhibition, 3-6 hours post exposure) Other: significant brain ChEI at doses ≥10 mg/kg Note: red blood cell ChE measurements were not collected.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.7485	Metabolism and Pharmacokinetics In Fischer 344 Rats	MRID# 40458901 Acceptable/guideline 0.5 or 25 mg/kg of ^{14}C labeled chlorpyrifos or 15 daily doses of 0.5 mg/kg unlabeled chlorpyrifos followed by one dose of 0.5 mg/kg of ^{14}C labeled chlorpyrifos.	During 72 hours, more than 84% of the radioactivity was recovered in the urine, about 5% was found in the feces and less than 0.2% was found in the tissues and carcass. The metabolism of chlorpyrifos was extensive, and no unchanged parent compound was found in the urine. The major urinary metabolites were TCP, as well as glucuronide and sulfate conjugates of TCP.
870.7485	Metabolism and Pharmacokinetics In Fischer 344 Rats	MRID# 44648102 Acceptable, Non-guideline 0.5, 1, 5, 10, 50, or 100 mg/kg and followed vs time Four male rats were given a single gavage dose of labeled chlorpyrifos at a concentration of 5 or 100 mg/kg and were sacrificed three hours later.	Peak chlorpyrifos blood concentrations occurred within three hours of treatment. Plasma ChE activity decreased in a time- and dose-dependent manner. The plasma ChE activities of rats treated with 0.5, 1, 5 or 10 mg/kg were maximally decreased 3-6 hours after treatment, with both the decrease and recovery of activity being dose-dependent. In the 1 mg/kg dose group, plasma ChE activity was significantly inhibited approximately 28% and 40% relative to controls at 3 and 6 hours post exposure, respectively. By 12 hours post-exposure, plasma ChE activity was still significantly inhibited about 16% for the 1 mg/kg group. The decrease in plasma ChE activity of rats treated with 50 or 100 mg/kg began within 10 minutes of treatment. By 12 hours after treatment, plasma ChE activity in both groups were approximately 11% of the control group and had not shown signs of recovery. Brain cholinesterase activity was not affected as dramatically by test material treatment as plasma activity with only the 10, 50, and 100 mg/kg dose groups showing significant effects. The brain cholinesterase activity of rats treated with 10 mg/kg test material began to decline within three hours of treatment and was significantly decreased by six hours after treatment. The brain cholinesterase activity in the 50 or 100 mg/kg dose groups decreased significantly within one hour of treatments; and by 12 hours, it was approximately 30% and 20%, respectively, of control. In none of the affected groups did brain cholinesterase show signs of recovery.

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
870.7600	Dermal Penetration (Human)	Accession No. 249203 Single doses of 0.5 mg/kg (N=1) and 5.0 mg/kg (N=5) to male humans	Based on the urinary excretion of the 3,5,6-TCP metabolite, the minimum absorption was approximately 1-3% dermally. The proportion of the administered dose metabolized to this pyridinol is unknown, these estimates are considered minimum values (i.e. absorption could be higher).
870.7800	Immunotoxicity in Female Crl:CD(SD) Rats	MRID 48139304 Acceptable/guideline 0, 0.416, 2.13, or 10.7 mg/kg/day	NOAEL is 10 mg/kg/day LOAEL not established
Special Study	Special Acute Neurotoxic Esterase (NTE) Rat Study	MRID 44273901 Acceptable/Non-guideline 0, 1, 5, 10, 50 or 100 mg/kg	98.1% a.i. chlorpyrifos NOAEL: 1 mg/kg [plasma ChE, and RBC and heart acetyl cholinesterase (AChE)] LOAEL: 5 mg/kg (45% plasma ChEI; 17% RBC AChEI; and 19% heart AChEI). Effects: NTE was not inhibited at any dose. Note: cholinesterase measurements were made 24 hours post exposure.
Special Study	Cognitive Rat Study	MRID 44020901 Acceptable/Non-Guideline 0, 1, 3, or 10 mg/kg/day for 5 days/week for 4 weeks	98.1% a.i. chlorpyrifos NOAEL: none observed (plasma and RBC ChE), LOAEL: 1 mg/kg/day (68% plasma ChEI; 56% RBC ChEI and 8% brain ChEI). NOAEL (systemic): 1 mg/kg/day (miosis) LOAEL (systemic): 3 mg/kg/day (miosis)

Table A.2.2 Chlorpyrifos Sub-chronic, Chronic and Special Studies Toxicity Profile

Guideline No.	Study Type	MRID# / Classification /Doses	Results
Special Study	Comparative Cholinesterase Assay	MRID 48139301 Acceptable/Non-Guideline	Repeat Dosing Data Chlorpyrifos (CPY) & Oxon (CPO) NOAEL/LOAEL in mg/kg/day (% Inhibition) Plasma ChE: Pups & Adult CPY 0.1/0.5 (46%) & 0.1/0.5 (46%) CPO 0.01/0.5 (62%) & 0.01/0.5 (76%) RBC ChE: Pups & Adult CPY 0.1/0.5 (18%) & 0.1/0.5 (20%) CPO 0.01/0.5 (84%) & 0.01/0.5 (87%) Brain ChE: Pups & Adult CPY 0.5/1 (19%) & 0.5/1 (9%) CPO Not inhibited & Not inhibited
Special Study	Acute Inhalation Study	MRID 48139303 Acceptable/Non-Guideline	NOAEL Not Identified LOAEL 3.7 mg/m^3 based on lung cholinesterase activity

A.3 Hazard Identification and Endpoint Selection

A.3.1 Acute Population Adjusted Dose (aPAD) - Females age 13-49

No endpoint selected for this category.

A.3.2 Acute Population Adjusted Dose (aPAD) - General Population

Study Selected: Comparative Cholinesterase Assay (CCA)
MRID No.: 48139301
Executive Summary: See Appendix A.4.9
Dose and Endpoint for Risk Assessment: $BMDL_{10}$=0.36 mg/kg
Comments about Study/Endpoint/Uncertainty Factors: The CCA study is currently thought to be the most appropriate endpoint for chlorpyrifos. The UF_A=10 and the UF_H=10, with a proposed FQPASF =1. The acute PAD=0.0036 mg/kg/day.

A.3.3 Chronic PAD

Study Selected: DNT Gavage Study in Pregnant Rats
MRID No.: 44648101 and 44556901
Executive Summary: See Appendix A.4.7, Guideline 870.6300
Dose and Endpoint for Risk Assessment: $BMDL_{10}$=0.03 mg/kg/day
Comments about Study/Endpoint/Uncertainty Factors: The ChE data is currently thought to be the most appropriate endpoint for chlorpyrifos. The UF_A=10 and the UF_H=10, with a proposed FQPA SF=1. The chronic PAD=0.0003 mg/kg/day.

A.3.4 Incidental Oral Exposure (Short- and Intermediate-Term)

Short Term Exposure

Study Selected: Repeat Oral CCA Study in Rat
MRID No.: 48139301
Executive Summary: See Appendix A.4.9 Special Studies
Dose and Endpoint for Risk Assessment: $BMDL_{10}$=0.1 mg/kg/day
Comments about Study/Endpoint/Uncertainty Factors: The CCA study is currently thought to be the most appropriate endpoint for chlorpyrifos. The UF_A=10 and the UF_H=10, with a proposed FQPA SF=1.

Intermediate Term Exposure
See Chronic Dietary Endpoint

A.3.5 Dermal Exposure (Short-, Intermediate- and Long-Term)

Study Selected: 21-Day Dermal Study
MRID No.: 40972801
Executive Summary: See Appendix A.4.1, Guideline 870.3200

Dose and Endpoint for Risk Assessment: NOAEL = 5 mg/kg/day based on plasma and RBC ChE inhibition seen at LOAEL = 10 mg/kg/day.
Comments about Study/Endpoint/Uncertainty Factors: A repeated dose dermal study is the most appropriate study for this exposure and ChE inhibition is the most appropriate assay. The UF_A=10, the UF_H=10, the proposed FQPA SF =1 (residential exposures).

A.3.6 Inhalation Exposure (Acute, Short- and, Intermediate-Term)

Acute Exposure

Study Selected: Acute Inhalation Study
MRID No.: 48139303
Executive Summary: See Appendix A.4.9, Special Study
Dose and Endpoint for Risk Assessment: LOAEL = 3.7 mg/m^3 and the HEC = 0.62 mg/m3 (RfC=0.00021 mg/m^3) based on ChE inhibition. A NOAEL was not identified.
Comments about Study/Endpoint/Uncertainty Factors: This study is appropriate since it is the correct exposure of inhalation and ChE activity is appropriate as an endpoint for chlorpyrifos. For residential: The UF_A=3, UF_H=10, FQPA UF_{DB} =10 (extrapolation of LOAEL to NOAEL).

Short and Intermediate Term Exposure

Study Selected: 90-Day Inhalation Studies
MRID No.: 40908401, 40013901, 40166501, 44556901
Executive Summary: See Appendix A.4.1, Guideline 870.3465
Dose and Endpoint for Risk Assessment: NOAEL (calculated from an HEC) = 0.56 mg/kg/day (for occupational) and HEC = 0.0057 mg/m3) based on ChE inhibition seen at LOAEL = 0.3 mg/kg/day in the DNT study.
Comments about Study/Endpoint/Uncertainty Factors: This study is appropriate since it is the correct exposure of inhalation and ChE activity is appropriate as an endpoint for chlorpyrifos. The HEC=0.0057 mg/m^3. For residential: UF_A=3, UF_H=10, FQPA SF=1. For occupational: UF_A=3, UF_H=10.

A.4 Executive Summaries

A.4.1 Sub-chronic Toxicity

870.3100 90-Day Oral Toxicity – Rat

In a sub-chronic oral toxicity study in rats (MRID 40436406), chlorpyrifos (95.5% a.i.) was fed to 20 rats/sex/dose at dose levels of 0, 0.5, 10 or 200 ppm (equivalent to 0, 0.025, 0.5 or 10 mg/kg/day) for 13 weeks.

There were no treatment related effects on mortality, clinical signs, histopathology or organ weights. A significant decrease in body weight gain was observed in high dose males during the first half of the study, and in high dose females during the first three weeks. However, body weight in exposed animals was similar to controls by week 13. Food consumption in the high-

dose animals was also significantly increased during the time of increase body weight gain. Hematological effects were observed in both high-dose males and females, characterized by significantly reduced packed cell volume (PCV), hemoglobin (HB) and erythrocyte (RBC) group means relative to controls, which is suggestive of anemia. However, these parameters were within the normal range. Urinalysis revealed that males in the high dose group had a significantly reduced urine volume, increased urine pH, a higher specific gravity and a higher protein grading, which appear to be treatment-related.

No biologically or significant or treatment-related differences were noted for clinical chemistry parameters, with the exception of cholinesterase (ChE) inhibition. Significant and dose-related plasma ChE inhibition of 22, 37 and 72% was observed in the 0.5, 10 and 200 ppm male groups, respectively. In females, plasma cholinesterase was also significantly inhibited at 91 and 57% for the 10 and 200 ppm groups, respectively, but was not inhibited in the low dose group (10% increase). However, the registrant acknowledged the possibility that the cholinesterase data for the 10 and 200 ppm female groups were accidently switched. Red blood cell and brain cholinesterase activity were not evaluated in this study.

The LOAEL for plasma cholinesterase inhibition is 0.5 ppm (0.025 mg/kg/day) for males, which is the lowest dose tested. No NOAEL was observed for cholinesterase inhibition. The systemic NOAEL and LOAEL are 10 and 200 ppm, respectively (0.5 and 10 mg/kg/day, respectively) based on decreased body weight gains and possible anemia. The study is classified as acceptable/guideline.

Chlorpyrifos was administered (0.1, 1, 5, and 15 mg/kg/day) in the diet for 90 days to CDF Fischer 344 rats (MRID 40952801). Body weight and body-weight gain were decreased in the high dose males (15 mg/kg) at the beginning (first 4 weeks) and near the end (day 70 on) of the study. Plasma and RBC cholinesterase activities were decreased in both sexes at the interim time point at 1, 5, and 15 mg/kg (dose-related) and in females at the 0.1 mg/kg dose level. At termination, brain cholinesterase was decreased (dose-related) at the 5 and 15 mg/kg dose levels in both sexes; plasma cholinesterase activity was decreased at 1, 5 and 15 mg/kg in both sexes; and erythrocyte cholinesterase activity was decreased in both sexes at 5 and 15 mg/kg and in females also at 1 mg/kg. The only other treatment-related effect was increased vacuolation in the adrenal gland in males of the 5 and 15 mg/kg dose groups.

The NOAEL can be set at 0.1 mg/kg, the LOAEL at 1 mg/kg, based on decreased plasma and RBC cholinesterase activities. The study is classified as acceptable/guideline.

870.3150 90-Day Oral Toxicity – Dog

In a sub-chronic oral toxicity study in dogs (MRID 42172801), chlorpyrifos (95.8% a.i.) was administered by gelatin capsule to 4 beagle dogs/sex/dose at dose levels of 0, 0.01, 0.22, or 5 mg/kg/day each day for 13 weeks.

There were no treatment related effects on mortality, clinical signs, body weight, food consumption, ophthalmological examination, urinalysis, or organ weights. Although some statistically significant differences were noted in some hematological parameters, these findings were not considered biologically significant, or treatment related. No biologically significant

differences were noted for clinical chemistry parameters, with the exception of cholinesterase (ChE) inhibition. Significant and dose-related plasma and red blood cell ChE inhibition were observed in both sexes throughout the study. Plasma ChE was significantly inhibited in males (33-63%) and females (42-67%) exposed to 0.22 mg/kg/day and in males (69-85%) and females (64-87%) exposed to 5 mg/kg/day. Red blood cell ChE was also significantly inhibited in males (32-46%) and females (24-38%) exposed to 0.22 mg/kg/day during weeks 6 and 12 and in males (38-85%) and females (29-86%) exposed to 5 mg/kg/day during weeks 1, 6 and 12. Brain ChE activity was significantly reduced 46% at 5 mg/kg/day in both males and females. Although possible treatment-related gross and microscopic pathology changes were observed in the high dose animals, these findings were not observed in the 2-year dog study, and only occurred in one male and one female. These include a thickened muscular wall of the duodenum and an area of papillomatous hyperplasia (pyloric).

The NOAEL and LOAEL for plasma and red blood cell cholinesterase inhibition are 0.01 and 0.22 mg/kg/day, respectively. The study is classified as guideline/acceptable.

870.3200 21/28-Day Dermal Toxicity – Rat

In a 21-day dermal toxicity study (MRID 40972801), 5 Fischer 344 rats/sex/dose were dermally exposed to 0, 0.1, 0.5, 1 or 5 mg/kg/day chlorpyrifos (100% a.i.) in corn oil on a 12 cm^2 area of the back once per day, 6 hours/application, 5 days/week for a total of 15 applications in 21 days. In a 4-day dermal probe study used to select the doses, 4 female Fischer 344 rats/dose were similarly treated via dermal application at dose levels of 0, 1, 10, 100 or 500 mg/kg/day chlorpyrifos in corn oil for four consecutive days.

In the 21-day study, there were no signs of treatment-related systemic or dermal toxicity at doses up to 5 mg/kg/day, including effects on cholinesterase inhibition, body weight, food consumption, ophthalmological examination, hematology, or clinical chemistry. In the 4-day probe study, 2 of 4 females in the 1 and 10 mg/kg/day groups developed slight erythema. Dose-related plasma (45, 92 and 98%↓) and red blood cell (16, 49 and 75%↓) cholinesterase inhibition were observed in the 10, 100 and 500 mg/kg/day groups. However, statistical analyses were not conducted. The cholinesterase activities of the 1 mg/kg/day females were slightly decreased, but within the historical control range. No other treatment-related effects were noted in the dermal probe study.

The NOAEL and LOAEL for plasma and red blood cell cholinesterase inhibition are 5 and 10 mg/kg/day, respectively, based on the results of both the 21-day and 4-day dermal probe studies. Satisfies the guideline requirement (82-2), but not guideline 870.3200, which requires 10 animals/sex/dose for dermal toxicity testing.

870.3465 90-Day Inhalation – Rat

In a sub-chronic nose-only inhalation study (MRID 40908401), Fischer 344 rats (10/ sex/ concentration) were exposed nose only to Chlorpyrifos (95% a.i.) at vapor concentrations of 0, 5, 10, or 20.6 ppb (0, 72, 143 or 287 $\mu g/m^3$, respectively) 6 hours/day, 5 days/week for 13 weeks. These concentrations resulted in estimated maximum exposures of 0, 0.024, 0.048 and 0.097

mg/kg/day, respectively based on the EPA default ventilation rate of 0.00715 m^3/hr for rats (average of males and females), and average study specific body weights of 0.189 and 0.127 kg for male and female controls, respectively. The study author stated that the saturation or near saturation level was 20 ppb.

There were no treatment-related effects on mortality, body weight, clinical signs, ophthalmoscopy, hematology, gross pathology or histopathology. In females, food consumption was slightly depressed throughout the study in all dose groups without correlation to the dose level, although this observation was not considered of toxicological significance due to only slight decreases in corresponding body weights. There were some sporadic differences in clinical chemistry parameters, although these were not considered treatment-related due to a lack of dose-response and inconsistency between interim and terminal values. Sporadic differences in organ weights also were not considered treatment-related and appeared to be attributed to the increase mean body weights.

Significant plasma cholinesterase (ChE) inhibition was observed in the high dose males (23%) and females (25%) at the terminal sacrifice. Significant plasma ChE inhibition was also noted in females of the 5 and 10 ppb groups (26 and 40%, respectively), although a dose-response relationship was not apparent. Interim (8 week) measurements were similar or slightly greater than controls. Red blood cell (RBC) (interim and terminal) and brain (terminal) ChE activities were not significantly inhibited at any dose level. It should be noted that the chlorpyrifos concentrations in the exposure chambers at 13 weeks were approximately 12, 16 and 24 ppb, which exceeds the 5, 10 and 20 ppb average exposure levels and this may partially explain the terminal results, while the 8 week concentrations were closer to the average levels. The plasma ChE inhibition was not considered of toxicological significance because of the minimal inhibition (23-25%) at the high dose, lack of dose-response, and an absence of inhibition in the 8 week interval.

No LOAEL was identified in this study. Therefore, the NOAEL for systemic effects and plasma cholinesterase inhibition exceeds 20 ppb or 0.097 mg/kg/day. This study is classified as acceptable/guideline.

In a sub-chronic, nose-only inhalation study (MRID 40013901 & 40166501), Fischer 344 rats (10/sex/concentration) were exposed nose only to Chlorpyrifos at vapor concentrations of 0, 5.2, 10.3, or 20.6 ppb (0, 72, 143 or 287 μg/m^3, respectively) 6 hours/day, 5 days/week for 13 weeks. Cholinesterase activity was measured at study termination. The maximum dose to rats in the 20.6 ppb group was estimated to be 0.044-0.082 mg/kg/day based on average study specific body weights of 0.15 and 0.282 kg for female and male control animals, respectively and the EPA default rat ventilation rate of 0.00715 m^3/hr (average for males and females).

There were no treatment-related effects on body weight, clinical signs, urinalysis, hematology, clinical chemistry, organ weights, gross pathologic or histopathologic evaluations, or plasma, red blood cell or brain cholinesterase activities. Although female rats of all treatment groups had a slight (<4%) but significant decrease in red blood cell count, and males of all treatment groups had slightly elevated (approximately 13%) serum urea nitrogen, these observations were not

considered treatment-related due to a lack of dose-response, and all values were within the historical control range.

No LOAEL was identified in this study. Therefore, the NOAEL for systemic toxicity and cholinesterase inhibition exceeds 20 ppb or 0.082 mg/kg/day. The studies are classified as acceptable/guideline.

A.4.2 Prenatal Developmental Toxicity

870.3700a Prenatal Developmental Toxicity Study – Rat

Chlorpyrifos was dosed via gavage at 0, 0.5, 2.5 and 15 mg/kg/day in CD rats during gestation day 6-15 (MRID 40436407).

Based on ChE inhibition, the maternal NOAEL is < 0.5 mg/kg/day (LDT) with the maternal systemic toxicity NOAEL =15 mg/kg/day. The maternal systemic toxicity LOAEL =15 mg/kg/day (decrease in food consumption only first few days of dosing) and decrease in body weight gain during dosing. The developmental toxicity NOAEL = 2.5 mg/kg with the LOAEL =15 mg/kg/day (increase in post implantation loss). This study is classified as acceptable/guideline.

In a developmental toxicity study (MRID 00130400) Chlorpyrifos, 96.6% a.i., was administered to Fischer 344 rats by gavage at dose levels of 0 (corn oil vehicle only), 0.1, 3.0, or 15 mg/kg/day from days 6 through 15 of gestation. There were 31 rats each in the control and the 0.1 mg/kg/day (LDT) groups and 32 in the 3.0 mg/kg/day (MDT) group and 33 in the 15 mg/kg/day (HDT) group.

Maternal toxicity - There were no deaths in any group. Food and water consumption were not altered by compound exposure and liver weights (the only organ for which weights were obtained) was not altered in dosed groups compared to controls. Mean group maternal body weight gain was not affected in the LDT or MDT compared to controls but was reduced 26% during the period of dosing (gestation days 6-15) in the HDT. This effect appeared to transient though, as the HDT group had weight gain similar to the other groups in the post-treatment period (gestation days 16-21).Clinical signs of toxicity were evident at the HDT only. Excessive salivation, perineal urine stains, peri-ocular porphyrin deposits, vaginal bleeding, and tremors were noted throughout the dosing period in the HDT. Most cesarean section parameters were not altered by compound exposure. The only parameter that was altered was the pre-implantation loss. Pre-implantation loss in the controls was 5.3% while it was 9.4, 7.2 and 17% in the LDT, MDT and HDT groups respectively. Inhibition of cholinesterase activity was seen in the MDT and HDT groups. Plasma cholinesterase activity was decreased from 44.73 µ/ml in the controls to 4.28 and 1.56 µ/ml in the MDT and HDT respectively (these represent inhibitions of 90.3% and 96.5%, respectively). Erythrocyte cholinesterase activity was reduced from 11.98 µ/ml in the controls to 3.07 and 2.51 µ/ml in the MDT and HDT respectively (this represents inhibitions of 74.3% and 79%). The cholinesterase values for the LDT were similar to controls with plasma being 42.28 and erythrocyte being 11.85 µ/ml.

The systemic maternal LOAEL is 15 mg/kg/day, based on clinical signs such as salivation, and tremors. The systemic maternal NOAEL is 3.0 mg/kg/day. The maternal cholinesterase LOAEL is 3.0 mg/kg/day based on statistically significant decreases in erythrocyte and plasma cholinesterase activity. The maternal cholinesterase NOAEL is 0.1 mg/kg/day.

External examinations, visceral examinations and skeletal examinations did not reveal an increase in variations or malformations. There were no treatment-related effects in developmental parameters seen at any dose.

The developmental NOAEL is 15 mg/kg/day. The developmental LOAEL was not determined. This study is classified as acceptable/guideline.

In a developmental toxicity study (MRID 00095268), female CF-1 mice were administered chlorpyrifos by gavage on gestation days 6-15 at doses of 0, 1, 10, and 25 mg/kg/day (Experiment I). Because of severe maternal toxicity in the high dose group, additional groups of mice were exposed to 0, 0.1, 1.0, or 10 mg/kg/day on gestation days 6-15, inclusive (Experiment II). Maternal toxicity in the form of increased mortality (0/51, 1/40, 1/44, and 4/47 [$p<0.05$] at 0, 1, 10, and 25 mg/kg/day) and an increase in the number of mice showing clinical signs (0/51, 2/40, 9/44, and 32/47 at the above doses) were reported. Fetotoxicity was observed only at 25 mg/kg/day (decreased fetal body measurements and an increased incidence of minor skeletal variants). To determine the degree of RBC and plasma cholinesterase depression, additional groups of 4-10 mice were given 0, 1, 10, or 25 mg/kg/day of chlorpyrifos on day 6, days 6 through 10, or days 6 through 15 of gestation. Additionally, groups of 6-15 mice were given 0, 0.1, 1.0 or 10.0 mg/kg/day of chlorpyrifos concurrently with the animals for the low dose study (Experiment II) on day 6, days 6-10, or days 6 through 15 of gestation. Five hours after the final dosing (day 6, 10 or 15 of gestation, respectively), blood was obtained by cardiac puncture. A homogenate of fetuses from the mice sacrificed on day 15 of gestation was prepared to measure total fetal cholinesterase levels. Plasma cholinesterase levels decreased significantly in mice given 1, 10 or 25 mg/kg of chlorpyrifos on day 6, days 6 through 10, or days 6 through 15 of gestation. RBC cholinesterase levels also decreased significantly in mice given 10 or 25 mg/kg on day 6, days 6 through 10, or days 6 through 15 of gestation. Among mice given 1 mg/kg of chlorpyrifos on days 6 through 10 of gestation, a statistically significant decrease in RBC cholinesterase levels as compared to controls was observed. The fetal cholinesterase levels were decreased in fetuses from dams given 10 or 25 mg/kg of test material on days 6 through 15 of gestation.

The maternal LOAEL is 25 mg/kg/day, based on increased mortality and increased number of mice with clinical signs of cholinesterase inhibition. The maternal NOAEL is 10 mg/kg/day. The developmental LOAEL is 25 mg/kg/day, based on decreased fetal body measurements and increased incidence of minor skeletal variants. The developmental NOAEL is 10 mg/kg/day. The NOAEL for plasma and red blood cells cholinesterase is 0.10 mg/kg/day. This study is classified as unacceptable/non-guideline.

870.3700b Prenatal Developmental Toxicity Study – Rabbit

Chlorpyrifos was dosed via gavage at 0, 1, 9, 81, and 140 mg/kg/day to New Zealand rabbits for gestation days 7-19 (MRID 40436408).

Based on ChE inhibition the maternal NOAEL = 81 mg/kg with the maternal LOAEL = 140 mg/kg (based on decreased food consumption on gestation days 15-19; body weight loss during the dosing period followed by a compensatory weight gain; suggestion of post-implantation loss). The developmental NOAEL = 81 mg/kg/day with the LOAEL = 140 mg/kg/day (based on slight reduction fetal weights and crown-rump lengths; increased incidence of unmodified 5 th sternebra and/or xiphistrnum). The study is classified as acceptable/guideline.

A.4.3 Reproductive Toxicity

870.3800 Reproduction and Fertility Effects – Rat

In a two-generation reproduction study (MRID 41930301) chlorpyrifos (97.8-98.5% a.i.) was administered to 30 Sprague Dawley rats/sex/dose via the diet at dose levels of 0, 0.1, 1 and 5 mg/kg/day during the pre-mating period of 10 or 12 weeks (F_0 or F_1 generation, respectively); with exposure continuing in dams through gestation, lactation and weaning. The F_0-generation rats were mated once to produce F_1 litters. Plasma, red blood and brain cholinesterase (ChE) activity were determined for the first 10 F_0 and F_1 adult rats/sex/dose at the scheduled necropsy.

There were no treatment related effects on mortality, food consumption or clinical signs in either F_0 or F_1 animals. Parental toxicity was observed at 1 and 5 mg/kg/day as indicated by significant dose-related reductions in the ChE activities of the plasma (43-72% inhibition), and red blood cells (65-75% inhibition) in the F_0 and F_1 male and female adult rats. In addition, significant inhibition of brain ChE was noted in the high dose F_0 adult male and females (48 and 49% inhibition, respectively) and high dose F_1 males and females (53 and 58% inhibition, respectively). Parental F_0 and F_1 rats exposed to 5 mg/kg/day developed histopathological lesions of the adrenal gland that were confined to the cells of the zona fasciculata and were characterized as very slight to slight vacuolation. Also, histological changes in the adrenal gland were consistent with fatty changes in males and altered tinctorial properties in females. The body weights of the adult F_1 males were slightly lower than controls throughout the study in the 5 mg/kg/day dose group.

Neonatal effects were observed only in the presence of maternal toxicity and consisted of reduced pup weights and increased mortality at 5 mg/kg/day. There were no treatment-related effects on other reproductive parameters such as fertility indices, length of gestation, time to mating, pup sex ratio, pup survival, or litter size in either generation.

The NOAEL and LOAEL for parental toxicity are 0.1 and 1 mg/kg/day, respectively based on significant plasma, and red blood cell cholinesterase inhibition. The NOAEL and LOAEL for neonatal effects are 1 and 5 mg/kg/day, respectively, based on decreased pup weight and increased pup mortality. This study is classified as acceptable/guideline.

Chlorpyrifos was dosed to 10 males and 20 females per group at 58 days of age at levels of 0, 0, 0.03, 0.1 and 0.3 mg/kg/day for 1st generation and 0, 0, 0.1, 0.3 and 1.0mg/kg/day for subsequent generations (MRID 00029064 & 00064934). For each mating, conducted at 118 days of age, the number of conceptions, litter size, still births, resorptions, number and size of pups weaned, pup weight and growth rate were examined., Necropsy was performed upon death and on 5 rats/sex/group of Fla, F2a and F3a pups. Histology was conducted on control and F3a pups. Maternal RBC and plasma cholinesterase activity was measured at the time of Cesarian delivery. Only the b litters were used for reproduction study.

No clinical signs of toxicity were observed in parents or offspring. No treatment related effect was found on mortality, body weight gain, food consumption, number of pups, mean litter size, sex ratios, mean litter weight, growth rate (to weaning), gross and histological examinations (on F3a pups). The parental NOAEL is 0.1 mg/kg/day and the LOAEL is 0.3 mg/kg/day based on plasma and RBC ChE inhibition. The reproductive NOAEL not determined and the LOAEL is >1 mg/kg/day. The viability and lactation indices were decreased for F2a, F2b and F3a litters from the 1.0 mg/kg groups. Fetotoxicity may have arisen through the maternal milk. RBC and plasma ChE activity was depressed above 0.3 mg/kg level for female and at 1.0 mg/kg for male. No maternal toxic sign to 1.0 mg/kg/day. Reproduction indices are all normal for dose up to 1.0 mg/kg. This study is classified as acceptable/guideline.

Chlorpyrifos was dosed at 0.5, 0.8, 1.2 mg/kg/day in Sprague-Dawley rats (MRID 00130401).

Although not meeting core requirements for a reproduction study (primarily due to limited gross and no histological examination), the study is adequate to establish that the NOAEL for neonatal survival is 1.2 mg/kg/day (HDT), the primary purpose of the study. The NOAEL for other reproductive parameters is also 1.2 mg/kg/day and the NOAEL for general toxicity is 0.8 mg/kg/day based on decreased weight gain observed in the 1.2 mg/kg/day male dose level. In combination with the previous reproduction study (MRID No. 00029064 & 00064934), this study is adequate to meet the requirement for a core-minimum study.

A.4.4 Chronic Toxicity

870.4100a (870.4300) Chronic Toxicity

Chlorpyrifos was dosed to CD-1 mice at 0, 0.5 and 15 ppm for 2 years (MRID 00054352).

The systemic and oncogenic NOAEL was 2.25 mg/kg/day based on decreases in ChE activity of 90% in plasma and 50% in RBC. The study LOAEL values were not determined. The study is classified as acceptable and satisfies the requirement when taken together with MRID 00142902.

870.4100b Chronic Toxicity – Dog

The chronic toxicity study (MRIDs 00064933, 00146519) in dogs consisted of two phases. In Phase A, chlorpyrifos (97.2-98.8% a.i) as Dowco® 179 was administered to 3 beagle dogs/sex/dose in diet at dose levels of 0, 0.01, 0.03, 0.1, 1 or 3 mg/kg/day for one year (Phase A). One dog/group was sacrificed at one year, and the remaining 2 dogs/group were sacrificed after a 3 month recovery period. In Phase B, chlorpyrifos was administered to 4 beagle dogs/sex/dose at the same dose levels for a total of two years (Phase B), at which time all dogs were sacrificed.

The NOAEL and LOAEL for plasma ChE inhibition are 0.01 and 0.03 mg/kg/day based on consistent mean inhibition of 10% to 29% at 0.03 mg/kg/day compared to controls for both males and females in Phases A and B. HED did not identify a NOAEL and LOAEL for RBC ChE inhibition due to inconsistencies in the data and the large standard deviations that confounded the interpretation of the data at lower dose levels. The NOAEL and LOAEL for brain ChE were 1 and 3 mg/kg/day. The systemic NOAEL and LOAEL are 1 and 3 mg/kg/day based on liver weight effects. The chronic toxicity study in dogs in conjunction with the addendum that contains supplemental information are classified as ACCEPTABLE-GUIDELINE.

A.4.5 Carcinogenicity

870.4200a Carcinogenicity Study – Rat

In a carcinogenicity toxicity study (MRID 42172802), chlorpyrifos (96.1% a.i) was administered to 55 Fisher F344 rats/sex/dose in diet at dose levels of 0, 0.2, 5 or 100 ppm (equivalent to approximately 0, 0.0132, 0.33, or 6.99 mg/kg/day for males and 0, 0.0146, 0.365 or 7.78 mg/kg/day for females, respectively) for 104 weeks. Plasma cholinesterase (ChE) activity (10/animals/sex/group) was measured on weeks 14, 32, 45, 78 and 104, while red blood cell (RBC) ChE activity (10/animals/sex/group) was measured at weeks 45, 78 and 104. Plasma, RBC and brain ChE activities were measured on 5 animals/sex/group at week 50 and in 10 animals/sex/group at terminal sacrifice.

Rats in the 100 ppm group exhibited significantly decreased body weights in both sexes, and a significant increased incidence of non-neoplastic lesions (cataracts and diffuse retinal atrophy) in females. Plasma ChE activity was significantly inhibited at 5 and 100 ppm in both sexes. Significant plasma cholinesterase inhibition in the 5 ppm group ranged from 15 to 51% throughout the study in both sexes. In females exposed to 0.2 ppm, red blood cell ChE was also significantly inhibited 42% at the 50 week sacrifice, but was elevated 14% at the terminal sacrifice. Red blood cell ChE was also significantly inhibited in the 50 week sacrifice for the 5 and 100 ppm females (39 and 45% ↓, respectively), but inhibition was less pronounced at the terminal sacrifice where inhibition was 11 and 18%, respectively. At the week 50 measurements, the decrease in RBC ChE activity in the treated groups appeared to be seriously influenced by the high control value (3891 U/g tissue) compared to the other control values which ranged from 2092 to 2586 U/g tissue. Therefore, the RBC ChE inhibition in females at 50 weeks is discounted because of the unusually high control value. Brain ChE was significantly

reduced in both high dose males and females at the 50 week and terminal sacrifices (57-80% ↓), but was not significantly decreased at the other doses. At terminal sacrifice, males in the high dose group had significantly lower absolute liver and kidney weights that were not significant after correction for body weight, and therefore were not considered treatment-related. There were no treatment related effects in mortality, clinical signs, food consumption, or hematology.

At the doses tested, there was no treatment related increase in tumor incidence when compared to controls. Dosing was considered adequate based on decreased body weights and the increased incidence of non-neoplastic lesions.

The LOAEL and NOAEL for plasma inhibition are 5 and 0.2 ppm, respectively (0.33 and 0.0132 mg/kg/day, respectively). The LOAEL and NOAEL for systemic effects of decreased body weights in both sexes, and increased incidence of cataracts and diffuse retinal atrophy in females are 100 and 5 ppm, respectively (6.99 and 0.33 mg/kg/day, respectively). This carcinogenicity study in rats is classified as ACCEPTABLE-GUIDELINE.

In a chronic toxicity/carcinogenicity study (MRID 40952802), chlorpyrifos (98.5% a.i) was administered to 50 Fisher F344 rats/sex/dose in diet at dose levels of 0, 0.05, 0.1, 1 or 10 mg/kg/day for 104 weeks. Ten additional rats/sex/group were randomly allocated for the 12-month sacrifice. Plasma and red blood cell (RBC) cholinesterase (ChE) activities (10/animals/sex/group) were measured at months 6, 12, 18 and 24. Brain ChE activities were also measured at the 12-month (10/rats/sex/dose) and 24 month (20 rats/sex/dose) scheduled sacrifices.

Rats in the 10 mg/kg/day group exhibited a slight, but significant decrease in body weights (2-9%) in both sexes. Body weight gain was approximately 90% of controls in males and comparable among females. Male rats in the high dose group had an increase in the size of the adrenal gland characterized microscopically by increased fatty vacuolation of the zonal fasciculata. In addition, males exhibited changes in clinical chemistry parameters (decreased serum cholesterol, total protein, and globulin), an increase in urine specific gravity, and a decrease in some common geriatric conditions (renal disease and biliary hyperplasia), which may be secondary changes and do not reflect any deleterious effect on a specific organ or the overall health of the animals. Similar, but less severe effects were noted in the high dose female rats. There were no significant differences in food consumption, or survival in either sex.

There was a dose-related (in most cases) decrease in ChE activity (plasma, red blood cell and brain) at each time point in both sexes. Plasma ChE was significantly inhibited in both sexes at the 1 mg/kg/day (39-86%) and 10 mg/kg/day (56-95%) dose levels throughout the study. Brain ChE was significantly decreased at both the 1 mg/kg/day (5-9%) and 10 mg/kg/day (58-61%) dose levels at the 12 month sacrifice, but was only statistically reduced in the 10 mg/kg/day dose group at termination (56-57%). In the 1 mg/kg/day dose group, brain ChE activities were increased 3% in males, and decreased 4% in females at the 24 month sacrifice. RBC ChE was significantly depressed at the 1 mg/kg/day (14-34%) and 10 mg/kg/day (24-37%) dose levels in males throughout the study, although statistical significance was not attained at 12-months, and the value in the 1 mg/kg/day males at termination was only 14% lower than the control value. In

females, mostly non significant RBC ChE inhibition ranged from 16-22% for the 1 mg/kg/day dose group and 18-40% for the 10 mg/kg/day dose group during the 12, 18 and 24 month sacrifices.

At the doses tested, there was no treatment related increase in tumor incidence when compared to controls. Dosing was considered adequate based on decreased body weights coupled with the significant inhibition of plasma, red blood cell and brain ChE.

The LOAEL and NOAEL for systemic effects are 10 and 1 mg/kg/day, respectively based on effects on the adrenal gland and clinical chemistry alterations in males. The LOAEL and NOAEL for significant plasma (39-86%) and brain (5-9%) cholinesterase inhibition are 1 and 0.1 mg/kg/day, respectively. This combined chronic toxicity/oncogenicity study in rats is classified as ACCEPTABLE-GUIDELINE.

870.4200b Carcinogenicity (Feeding) – Mouse

This study evaluated the oncogenic potential of test compound, at dietary concentrations of 0, 5.0, 50 or 250 ppm chlorpyrifos (equivalent to approximately: 0, 0.89, 8.84, or 45.2 mg/kg/d (M); and 0, 0.938, 9.79, or 48.1 mg/kg/d (F), respectively) when administered to CD-1 mice for 78 weeks (MRID 42534201).

Systemic toxicity was observed in high-dose animals and included decreased body weight and feed consumption in males, lower mean water consumption in females, increased incidence of gross clinical findings (ocular opacity, hair loss on head and around eyes) and non-neoplastic lesions (keratitis, hepatocytic fatty vacuolation) in high dose males & females. Neoplastic lesions were observed in both sexes, but were not considered to be treatment-related. Plasma cholinesterase activities were significantly reduced at all treatment levels; brain activities were significantly decreased only in the high-dose animals.

The systemic NOAEL = 50 ppm (MDT). Systemic LOAEL = 250 ppm (HDT), based on decreased body weight in males, increase incidences of non-neoplastic lesions in males & females. Results of the study showed that the test compound does not have oncogenic potential. This study satisfies guideline requirements for an oncogenicity study in mice.

A.4.6 Mutagenicity

870.5100 Gene Mutation Bacterial Cell

Chlorpyrifos was tested in Salmonella strains TA 98, TA 100, TA 1535, TA 1537 and TA 1538 in the presence and absence of S-9 at concentrations of 30, 100, 300, 1000, 3000 and 10000 µg/plate (MRID 00157058 and 40436410). DMSO was the solvent and negative control. The positive controls were sodium azide, 9-aminoacridine, 2-nitrofluorene and 2-anthramine.

Chlorpyrifos was not toxic nor did it appear to increase over control values the number of revertant colonies/plate. Positive controls caused appropriate mutagenic responses. These studies were classified as Acceptable/Guideline.

870.5300 Gene Mutation Mammalian Cell

Chlorpyrifos was tested for gene mutation potential at 0, 10, 20, 25, 30, 40 & 50 µM in mammalian cells (MRID 00152683).

Chlorpyrifos was cytotoxic at 10 µM and above without metabolic activation and no toxicity with activation. Precipitate formed at 30 µM and higher concentrations with or without activation. Chlorpyrifos was negative for gene mutation. This study is classified as acceptable/guideline.

Chlorpyrifos was tested for gene mutation potential at the following concentrations: nonactivation from 5-75 µg/ml and with activation from 30-1000 µg/ml (MRID 40436410). Testing in the cytotoxicity assays at the following conditions: nonactivation from 1.5-3746 µg/ml and with activation from 1.5-500 µg/ml. Positive controls were ethyl-methane sulfonate (nonactivated) and dimethylnitrosamine (activated).

Cytotoxicity was detected only in non-activated assays at 50 µg/ml. There was no evidence of mutation. This study is classified as acceptable/guideline.

870.5375 In Vitro Cytogenetics

Chlorpyrifos was tested in an in vitro chromosomal aberration assay with and without S-9 activation (MRID 40436409). Concentrations assayed were as follows with non-activation in the 10 hour assay at 1.56, 3.12, 5.2, 10.4, 15.6, 31.2, 52, 104 & 156 µg/ml and in the 19-20 hour assay at 0.975, 1.47, 2.93, 4.89, 9.75, 14.7, 29.3, 48.9, 97.5 & 147 µg/ml. Concentrations tested with activation in the two 10 hour assays were 1, 1.5, 3, 5, 10, 15, 30, 50 & 100 µg/ml and 2.95, 4.95, 9.85, 14.8, 29.6, 49.4, 98.5 & 296 µg/ml, plus concentrations for the 19-20 hour assay were 9.75, 14.7, 29.3, 48.9, 97.5, 147 & 293 µg/ml. Positive controls were mitomycin C (non-activation) and cyclophosphamide (activation).

Cytotoxicity was shown in both non-activated as well as in activated assays. Chlorpyrifos did not appear to cause chromosomal aberrations. Positive controls caused appropriate mutagenic response. This study was classified as Acceptable/guideline.

870.5395 Micronucleus Assay in Mammalian Erythrocytes

Chlorpyrifos was tested at levels of 0, 7, 22, 70 mg/kg by gavage in corn oil in the mouse (MRID 00152684).

Chlorpyrifos was negative for clastogenic effects. This study is classified as acceptable/guideline.

870.5500 DNA Repair Assay in Bacteria

Increased damage to bacterial DNA was detected (Study 256040). This study is classified as acceptable/guideline.

870.5550 Unscheduled DNA Synthesis in Hepatocytes

Chlorpyrifos was tested with concentrations from 10E-06 M to 10E-04 M in isolated rat hepatocytes (MRID 00157057).

Chlorpyrifos was negative for UDS in isolated rat hepatocytes under the conditions of this study. The high dose was cytotoxic and also formed a precipitate. This study is classified as acceptable/guideline.

870.5575 Mitotic Gene Conversion in Yeast

Increased recombination frequency was detected in yeast (Study 256040). This study is classified as acceptable/guideline.

A.4.7 Neurotoxicity

870.6100 Delayed Neurotoxicity Study – Hen

Chlorpyrifos was dosed at 0, 50, 100 or 110 mg/kg in hens (MRID 00097144 and 40510601).

There is no evidence of histopathologically observed neurotoxicity in hens. The NOAEL is 110 mg/kg, negative for neurotoxicity at 110 mg/kg. The LOAEL was not determined. The LD_{50} in hens = 106 mg/kg. These studies are classified as acceptable/guideline.

870.6200a Acute Neurotoxicity Screening Battery

Male & female Fischer 344 rats were treated once, by oral gavage, with chlorpyrifos at doses of 0, 10, 50 or 100 mg/kg and evaluated for neurotoxicity on days 1 (at the peak time of toxicity, approximately 6 hours after dosing), 8 and 15 (MRID 42669101 and 42943101).

Systemic toxicity consisted of decreased body weights of animals in the 50 and 100 mg/kg groups. Neurotoxic effects consisted of decreased motor activity on day 1 through day 8 (females only). Significant FOB changes were limited to high dose females, of which 6 out of 10 could not perform the landing hind leg splay on day 1 of the study. Grip performance on day 1 revealed a possible treatment-related decrease with increasing dose. Neuropathological examinations did not reveal any treatment-related effects. Systemic NOAEL (M&F) = 10 mg/kg (LDT) with the systemic LOAEL (M&F) = 50 mg/kg (MDT). LOAEL is based on decrease in both body weight and motor activity and increased incidence of adverse clinical signs consistent with organophosphorus intoxication. These studies are classified as guideline.

870.6200b Sub-chronic Neurotoxicity Screening Battery

In this sub-chronic neurotoxicity study, male and female Fischer 344 rats were treated for 13 weeks with diets containing sufficient chlorpyrifos to yield doses of 0, 0.1, 1.0, 5.0 or 15 mg/kg/day (MRID 42929801). During the study, body weights, clinical signs, FOB, motor activity and neuropathology were examined. FOB, performed at pre-study and weeks 4, 8, 13 consisted of hand-held and open field observations and measurement of grip performance and landing foot splay.

The study indicated the treatment-related effects included decreased motor activity and an increased incidence of urine incontinence on females. Although a statistically significant depression in motor activity was present in high-dose animals at week 4. The transitory nature of the effect suggests that the differences were not treatment-related. In addition, a low, and statistically non-significant, increase in the incidence of urine incontinence was observed in several 5 and 15 mg/kg/day females during the clinical examinations and FOB evaluations. One high-dose female showed urine incontinence at weeks 4, 8, and 13 and another, only at weeks 4 and 8. None of the other animals showed urine incontinence in more than one FOB session. There was no clear dose- or time-relationships which would suggest that the incontinence was treatment-related. Body weights of treated animals were comparable to controls. Neuropathological examination did not reveal any differences which might be attributed to treatment. No neurotoxicity was noted at 15 mg/kg/day, a dose previously shown to markedly inhibit plasma (>80%), RBC (>45%) and brain (>62%) cholinesterase activities.

The NOAEL for neurotoxicity was established at 15 mg/kg/day (high dose tested); the LOAEL was not established. This study is satisfies guideline requirements for a sub-chronic neurotoxicity screening battery in the rat.

870.6300 Developmental Neurotoxicity Study

In this developmental neurotoxicity study (MRID 44556901 and companion 44648101 cholinesterase study), 25 pregnant Sprague-Dawley rats/group were administered chlorpyrifos (99.8% a.i.) by gavage from gestation day 6 (GD 6) through lactation day 11 at 0, 0.3, 1, or 5 mg/kg/day. An additional 5 pregnant females/group were dosed at the same levels for the cholinesterase (ChE) phase of the study. Dams were examined for body weight, reproductive performance, number of viable pups, and postpartum behavior. During the dosing period, dams were observed daily for signs of autonomic function toxicity. Satellite dams were sacrificed four to five hours post-dosing on GD 20 for ChE analyses to be performed on brain, plasma, and erythrocytes. Offspring were examined for viability at birth, pup/litter survival, body weight, sex ratio, physical development landmarks (eye opening and pinna detachment), observed nursing behavior, and sexual maturation. F_1 generation litters were randomly standardized on lactation day 5 and assigned to 4 subsets for continued observation. On postnatal day (PND) 12, fixed brain weight measurements (10 pups/sex/dose) and neuropathological evaluations including morphometrics (6 pups/sex/dose) were performed on Subset 1 pups, with the remaining 10 pups/sex/dose necropsied for gross lesions. In Subset 2, 8 pups/sex/dose were selected for evaluation of learning and memory; evaluations were performed on PNDs 23-25 and 62-92. These Subset 2 animals were sacrificed on PNDs 97-101, following the last evaluation. The Subset 2 pups not selected for evaluation were necropsied for gross lesions on PND 22. The

Subset 3 pups were tested for motor activity on PNDs 14, 18, 22, and 61 and auditory startle habituation on PNDs 23 and 62; all Subset 3 animals were sacrificed on PNDs 63 or 64 following the last evaluation. In Subset 4 pups, fixed brain weights were determined in 10 pups/sex/dose, neuropathological examinations were performed on 6 pups/sex/dose, and all remaining Subset 4 animals (10/sex/dose) were necropsied for gross lesions upon sacrifice on PND 66-77.

Maternal toxicity in the high-dose (5 mg/kg/day) animals was manifested as increased signs of autonomic function toxicity, apparent at the end of gestation as fasciculations (6/25 treated vs 0/25 controls), and during early lactation (days 1-5) as fasciculations (16/24 treated vs 0/25 controls), hyperpnea (8/24 treated vs 0/25 controls), and hyperreactivity (17/24 treated vs 2/25 controls). Dams with all pups dying were increased in the high-dose group (3/23 treated vs 0/25 controls). There were no significant effects on bodyweight, food consumption, or pregnancy parameters. There were no unscheduled deaths in the maternal animals.

Brain ChE activity was decreased in the high-dose (↓90%) and mid-dose (↓18%) dams as compared to control. Erythrocyte (↓41-99%) and plasma (↓43-92%) ChE activities were decreased in a dose-dependent manner in all treated groups.

For the F_1 generation pups, the high-dose group bodyweights were significantly reduced (↓8-15%) at PND 1 and 5 (pre- and post-culling). Bodyweights were also reduced from birth to PND 22 in Subset 4 high-dose animals (↓5-19%); bodyweight gains were reduced in these animals during the same period (↓5-30%). Additionally, compared to the controls, reduced terminal body weights were observed in the Subset 1 (PND 12) high-dose animals (↓17-19%) and the Subset 4 (PND 66) high-dose males (↓10%). For the F_1 generation adults, body weights of the high-dose males were decreased at PND 22 through 66 (↓11-17% vs controls). High-dose F_1 adult females also weighed less than controls at PND 22 (↓17% vs controls), but were of similar weight at PND 66. Bodyweight gains were also decreased in the high-dose males for the PND 22-40 interval (↓13% vs controls) and PND 40-66 interval (↓7%). Food consumption was decreased immediately after weaning (PND 23-30) in high-dose males and females (↓13% vs controls).

Development as assessed by pinna unfolding was delayed (4.0 days in treated vs 3.5 days in controls) in the high-dose group. Sexual maturation was delayed as assessed by time to preputial separation (106% of controls) and vaginal patency (103% of controls).

Pup viability was reduced as assessed by the following parameters: surviving pups/ litter (↓27%) and live litter size at culling (↓16%), pup viability index (↓29%), and pups found dead or presumed cannibalized (day 1 - 7.2 % treated vs 0.0% controls; days 2 to 5 - 24.7% treated vs 1.3% controls).

There were no statistically significant differences between the groups in the average acquisition and delay training. Additionally, there were no differences among dose groups when comparing retention of information during PNDs 23-25 and 62-92. Motor activity was decreased in high dose male and female pups on PND 14 (↓56% in males and ↓37% in females), and increased in

high dose females on PNDs 18 and 22 (↑51% on both days). On PND 61, motor activity was increased for both sexes (↑16-17%). There was a statistically significant increase (↑16-25%) in the latency to peak response during the auditory startle habituation assessments on PND 23 in the high-dose animals compared to concurrent controls. At PND 62, the latency to peak response in the high-dose animals was 10-12% higher than in the controls. Additionally, the peak response amplitudes in the high-dose animals were decreased by 9 to 29% on PNDs 23 and 62 (not statistically significant) compared to the controls.

There were no gross or microscopic lesions of the nervous system in Subset 1 or 4 offspring. Subset 1 high-dose males at PND 12 had reduced absolute brain weights (↓9% vs controls), increased relative brain weights (↑13% vs controls), reduced anterior to posterior measurement of the cerebellum (↓24% vs controls), reduced height of the cerebellum (↓14% vs controls), decreased thickness of the parietal cortex (↓6% vs controls), and decreased thickness of the hippocampal gyrus (↓9% vs controls). High-dose female pups had reduced absolute brain weights (↓9% vs controls), increased relative brain weights (↑14% vs controls), decreased thickness of the parietal cortex (↓6% vs controls), decreased width of the caudate-putamen (↓10% vs controls), and decreased thickness of the hippocampal gyrus (↓12% vs controls). In Subset 4 F1 animals, killed on PND 66, morphometric analysis revealed significantly decreased parietal cortex measurements in high-dose (↓5%) and mid-dose (↓4%) females, as compared to controls. Decreases in the thickness of the hippocampal gyrus in high-dose females (↓7%) resulted in contradictory statistical results when compared to controls; decreases in mid-dose (↓4%) females as compared to control were not found to be statistically significant. There was no evaluation of the morphometric data for low dose females at PND 66. Brain weight in high dose females was similar to control brain weight at day 66 (↓0.3%).

It is not possible to definitively classify findings in the preweaning offspring as having originated with pre- or postnatal exposure, nor as resulting from developmental perturbation versus direct systemic- or neurotoxicity. However, adverse findings in the adult (~PND 66) offspring, i.e., alterations in motor activity, auditory startle response, and brain structure (decreased measurements of the parietal cortex and hippocampal gyrus, in the absence of brain weight deficits) can be interpreted to represent the long-term sequellae of developmental exposure to chlorpyrifos.

Adverse effects in the offspring have been identified at the MDT of 1.0 mg/kg/day; these include a significant treatment-related decrease in the measurement of the parietal cortex, supported by possible (although non-significant) alterations in the hippocampal gyrus, in the brain of female rats at postnatal day 66.

The maternal toxicity NOAEL was not observed. The maternal LOAEL was < 0.3 mg/kg/day, based on plasma and RBC cholinesterase inhibition. However, due to the lack of morphometric data for low-dose (0.3 mg/kg/day) female rats at postnatal day 66, the offspring NOAEL and LOAEL cannot be determined. This study has been classified as guideline/unacceptable (but upgradeable).

A.4.8 Metabolism

870.7485 Metabolism – Rat

This study (MRID 44648102) was done to help construct and validate a physiologically-based pharmacokinetic model for chlorpyrifos (Unlabeled - 99.8% a.i., Lot # MM930503-17; Labeled - 89.4% a.i., Lot # B930-51 [INV1134]) a weak inhibitor of acetylcholinesterase activity, and its metabolites, chlorpyrifos-oxon (OXON), a strong cholinesterase inhibitor and 3,5,6-trichloropyridinol. Groups of 24 male rats were given a single gavage dose of 0.5, 1, 5, 10, 50, or 100 mg/kg chlorpyrifos in corn oil. Four rats from each group were killed 10 and 20 minutes and 1, 3, 6, and 12 hours after treatment. Cholinesterase activity was measured in the brain and plasma at each time point, as well as the plasma concentration of the test material and its OXON metabolite. In a separate portion of the study, four male rats were given a single gavage dose of labeled chlorpyrifos at a concentration of 5 or 100.0 mg/kg and were sacrificed three hours later. Blood was collected from the animals at sacrifice and the concentration of the test material and its metabolites 3,5,6-trichloropyridinol (TCP) and OXON determined.

Plasma cholinesterase activity decreased in a time- and dose-dependent manner. The plasma cholinesterase activities of rats treated with 0.5, 1, 5 or 10 mg/kg were maximally decreased 3-6 hours after treatment, with both the decrease and recovery of activity being dose-dependent. The decrease in activity of rats treated with 50 or 100 mg/kg began within 10 minutes of treatment. By 12 hours after treatment, both groups were approximately 11% of the control group and had not shown signs of recovery.

Brain cholinesterase activity was not affected as dramatically by test material treatment as plasma activity with only the 10, 50, and 100 mg/kg dose groups showing significant effects. The brain cholinesterase activity in the 50 or 100 mg/kg dose groups decreased significantly within one hour of treatment; mirrored each other; and by 12 hours, were approximately 30% and 20%, respectively, of control. The brain cholinesterase activity of rats treated with 10 mg/kg test material began to decline within three hours of treatment and was significantly decreased by six hours after treatment. In none of the affected groups did brain cholinesterase show signs of recovery.

Peak chlorpyrifos blood concentrations occurred within three hours of treatment in all but the lowest dose group. The area under the curve (AUC) was calculated as 0.4, 1.1, 5.0, and 12.5 µmole hr L-1 for the 5.0, 10.0, 50.0, and 100 mg/kg groups, respectively and yielded calculated blood half-lives of chlorpyrifos of 2.7,1.5, 2.1, and 7.3 hours for the 5.0, 10.0, 50.0, and 100.0 mg/kg dose groups, respectively. Regardless of dose, the highest concentration of OXON detected was 2.5 ng/g found in the blood of rats treated with 50 mg/kg test material one hour post-treatment. Following treatment with 5 or 100 mg/kg labeled test material, >=98% of the activity detected in the blood was identified as TCP metabolite with the remaining attributed to the parent compound. Since OXON is an intermediate in the formation of TCP and none of the metabolite was detected, these studies support that the half-life of the OXON metabolite is short (reportedly 10 seconds) and that in vivo metabolism of chlorpyrifos is rapid.

This study is considered acceptable (non-guideline).). It may partially fulfill guideline

requirements in other areas.

In another study of tissue distribution and metabolism (MRID 40458901), carbon-14 labelled chlorpyrifos was administered orally to Fischer 344 rats for 15 days (MRID 40458901).

The majority of the radioactivity was recovered in the urine (>84%) and feces (>5%) within 72 hours. Less than 0.2% of the radioactivity remained in tissues and carcass. No unchanged chlorpyrifos was found in the urine and the main urinary metabolites were identified as 3,5,6-TCP and conjugates (glucuronide and possibly sulfate) of 3,5,6-TCP.

This study is classified as acceptable-guideline.

870.7600 Dermal Absorption

Single doses of 0.5 mg/kg (N=1) and 5.0 mg/kg (N=5) of chlorpyrifos were administered to male humans (accession No. 249203).

Based on the urinary excretion of the 3,5,6-TCP metabolite, the minimum absorption was approximately 1-3% dermally. The proportion of the administered dose metabolized to this pyridinol is unknown, these estimates are considered minimum values (i.e. absorption could be higher).

A.4.9 Immunotoxicity

870.7800 Immunotoxicity

In an immunotoxicity study (MRID 48139304), chlorpyrifos technical (99.8% a.i., Lot No. KC28161419) was administered in the diet to 10 female Crl:CD(SD) rats/dose at nominal dose levels of 0, 0.4, 2, or 10 mg/kg/day (actual dose levels of 0, 0.416, 2.13, or 10.7 mg/kg/day) for 28 days. The female rat was determined to be the appropriate test species/sex for this study. Cyclophosphamide in sterile saline was intra-peritoneally administered to the positive control group on Days 24 to 28 at a rate of 20 mg/kg body weight/day. On Day 24, all animals received a 0.5 mL intravenous injection of sheep red blood cells (SRBCs) in isotonic saline (2×10^8 SRBCs)/mL). T-cell dependent antibody response (TDAR) was evaluated at day 29.

There were no statistically significant effects of treatment with chlorpyrifos on mean body weights, body weight gains, or food consumption. Statistically significant decreases in mean red blood cell (RBC) cholinesterase (ChE) activity were seen in all test substance treatment groups. Mean brain ChE activity was significantly decreased in the mid- and high-dose groups. There were no test substance treatment-related effects on clinical signs, gross anatomy, or hematological parameters. In the positive control group, mean body weights and body weight gains were lower than the control value throughout the study; these differences were attributed to normal body weight variability. No unscheduled mortalities occurred in any study group. For systemic toxicity related to treatment with chlorpyrifos, the NOAEL for female rats is 10 mg/kg/day (highest dose tested) based on no effects were seen in clinical observations, body

weight, food consumption, and hematological parameters. The LOAEL for systemic toxicity was not established. For neurotoxic effects, the LOAEL for female rats is 0.4 mg/kg/day (lowest dose tested), based on decreased RBC cholinesterase activity. The NOAEL for neurotoxic effects was not established (i.e., less than 0.4 mg/kg/day).

For immunotoxicity, there were no treatment-related effects on mean absolute and relative spleen and thymus weights or hematological parameters at any dose level. The anti-SRBC IgM titers did not show statistically significant differences among treatment and the control groups. Decreased anti-SRBC titers for the 2 and 10 mg/kg/day treatment groups (64% and 41%, respectively) were observed when compared with the control. However, the decreased response in these dose groups may have been due, in part, to a high mean value for the control group. The biological significance of these observations also was confounded by the lack of a clear dose response (the decrease was greater for the mid-dose group than for the high-dose group). The positive control demonstrated the validity of the assay. Considered the trend and distribution of individual animal data in treatment and control groups, there was no significant suppression of the anti-SRBC titers with chlorpyrifos exposure.

The NK cell activity was not evaluated. There were no treatment-related effects on spleen and thymus weights and histopathology parameters that would suggest the potential for immunotoxicity in repeat-dose studies (2-week, 28-day, 90-day, 2-year) studies in rats and mice. Under HED guidance, if the TDAR assay is negative and evaluation of observational endpoints from all available toxicology database provide no evidence of immunotoxicity, the test article is considered negative for immunotoxicity and evaluation of NK activity is not necessary.

Under conditions of this study, the NOAEL is 10 mg/kg/day (highest dose tested) based on the overall weight-of-evidence. A lack of dose-related response for anti-SRBC IgM titers at the mid- and high-dose levels, a lack of statistical significance at any dose level, and a lack of evidence of other immunological effects (absolute and relative spleen and thymus weights, hematological parameters). A LOAEL for immunotoxicity was not established. This immunotoxicity study in the rat is considered as acceptable/guideline and satisfies the guideline requirement for an immunotoxicity study (OPPTS 870.7800) in rats.

A.4.10 Special/Other Studies

Comparative Cholinesterase Assay

Comparison of cholinesterase (ChE) inhibition in young adult and preweanling CD rats after acute and repeated chlorpyrifos exposures were performed (MRID 48139301).

The following table illustrates the NOAEL and LOAELs derived from the acute dosing aspects of this study. Male pups had the same NOAELs and LOAELs as female pups.

Enzyme Source	Acute NOAEL/LOAEL mg/kg (% Inhibition at LOAEL) Pups (male/female %)	Adults (females only)
Plasma ChE:		
CPY – gavage	0.5/2(51%/47%)	0.5/2(54%)
CPY – milk/diet	0.5/2(39%/44%)	0.5/2(58%)
CPO - gavage	0.05/0.1(18%/21% but 51% at 0.5 mg/kg)	0.1/0.5(56%)
RBC AChE:		
CPY – gavage	0.5/2(35% /31%)	0.5/2(19%)
CPY – milk/diet	0.5/2(29%/27%)/	0.5/2(52%)
CPO - gavage	0.1/0.5(46%/47%)	0.1/0.5(36%)
Brain:		
CPY – gavage	2/5(51%/55%)	2/10(57%)
CPY – milk/diet	2/5(42%/56%)	2/10(22%)
CPO - gavage	Not inhibited	Not inhibited

Cpy = chlorpyrifos
CPO = chlorpyrifos oxon

The following table illustrates the NOAEL and LOAELs derived from the repeat dosing aspects of this study.

Enzyme Source	NOAEL/LOAEL mg/kg (% Inhibition at LOAEL) Pups	Adults
Plasma ChE:		
CPY	0.1/0.5(46%)	0.1/0.5 (46%)
CPO	0.01/0.5 (62%)	0.01/0.5 (76%)
RBC AChE:		
CPY	0.1/0.5 (18%)	0.1/0.5 (20%)
CPO	0.01/0.5 (84%)	0.01/0.5 (87%)
Brain:		
CPY	0.5/1 (19%)	0.5/1 (9%)
CPO	Not inhibited	Not inhibited

The classification of this *in vivo* comparative cholinesterase inhibition study is Acceptable/Non-Guideline.

Special Acute Neurotoxic Esterase Study in Rat

In a special study designed primarily to assess for the potential of chlorpyrifos to inhibit neurotoxic esterase (NTE), chlorpyrifos was administered by gavage to six groups of Fischer 344 strain female rats at dose levels of 0, 1, 5, 10, 50 or 100 mg/kg and sacrificed 24 hours later (MRID 44273901). NTE was assessed for by the method of Kayyali *et al* (J. Anal. Toxicol. 15:86-89 (1991). Dosing was by gavage at a dosing volume of 10 ml/kg. The rats were also assessed for cholinesterase inhibition in the plasma, red blood cells (RBCs), heart and brain and there was an additional group dosed at 0.5 mg/kg included for assessment of cholinesterase only.

The cholinesterase inhibition data indicated a NOAEL and LOAEL for plasma cholinesterase (ChE) and RBC and heart acetylcholinesterase (AChE) of 1 and 5 mg/kg, respectively. At 5 mg/kg, plasma ChE, RBC AChE and heart AChE were significantly inhibited approximately 45%, 17% and 19%, respectively. Brain AChE demonstrated a NOAEL and LOAEL of 10 and 50 mg/kg, respectively and at 50 mg/kg inhibition was approximately 53%. NTE was not inhibited at the highest dose level of 100 mg/kg and there was an apparent 9% increase in activity at this dose level.

This study is classified as ACCEPTABLE (Non-Guideline). The study contains data useful for evaluating the potential for chlorpyrifos to inhibit neurotoxic esterase following systemic administration.

Cognitive Rat Study

In this special study (MRID 44020901) the effects of repeated oral administration of chlorpyrifos technical (purity, 98.1%; lot no. #MM-890115-616) on the cognitive function of rats were evaluated with a delayed matching to position (DMTP) test. Groups of 10 female Long-Evans rats, pretrained in a DMTP apparatus were administered oral doses of chlorpyrifos in corn oil of 0, 1, 3, or 10 mg/kg/day for 5 days/week for 4 weeks. DMTP testing was conducted 6 days/week during treatment and continued post-dosing for 4 weeks. Testing for short-term memory (as evidenced by the retention rate) and attention/encoding deficits was based on the percent correct accuracy on several time delays. Slope over delay and intercept at time zero were calculated from these data for each rat and represented the "forgetting curve."

A satellite group of 6 rats/dose was sacrificed after the 4-week dosing period and plasma, erythrocyte and brain cholinesterase (ChE) were determined. Neurotoxic esterase (NTE) activity was determined in satellite rats from the control and high-dose groups one day after the last dose administration. Plasma (68%), RBC (56%) and brain (8%) ChE were inhibited at 1 mg/kg/day. At 3 mg/kg/day, plasma (83%), RBC (65%) and brain (63%) ChE inhibition was increased. At 10 mg/kg/day plasma (93%), RBC (65%) and brain (86%) ChE inhibition was further increased. NTE was minimally decreased (6%) in the high-dose group but this was not considered toxicologically significant. The clinical sign of miosis was observed in rats that received 3 and 10 mg/kg/day particularly at weeks 3 and 4. Salivation and tremors were observed primarily at 10 mg/kg/day with the tremors usually disappearing by the following morning.

A statistical analysis of the actual percent correct data was provided (supplemental report dated February 10, 1999) and no statistical differences (i.e., $p < 0.05$) indicative of treatment related decreases in percent correct choices were established for any dose or delay time. Thus, cognitive function is not obviously impaired. No consistent pattern in the intercept of the retention gradient was noted since it was increased at week 2 and decreased at week 3 but equivalent to the control at weeks 1 and 4 at 10 mg/kg/day. The DMTP parameters of actual total delay (increased by as much as 2.5 sec in the 0 delay trial at week 2), void trials per session (increased from about 5 in the control to about 15) and nosepokes (decreased ~42% at week 1 for the 15 sec delay) were affected in the 10 mg/kg/day Chlorpyrifos dose at most or all intervals during dosing. Although these effects can be possibly related to a decrease in motor activity known to be associated with organophosphates, the increase in void trials may also indicate a motivational or attention deficit.

The LOAEL for ChE inhibition is 1 mg/kg/day, with no NOAEL was established. The LOAEL for overt cholinergic signs is 3 mg/kg/day based on miosis. The NOAEL is 1 mg/kg/day. The LOAEL for DMTP performance (i.e. increase in void trials) is 10 mg/kg/day with the NOAEL at 3 mg/kg/day. This study is classified ACCEPTABLE (Non-guideline).

Acute Inhalation Study

Acute inhalation exposure of adult Crl:CD(SD) rats to particulate chlorpyrifos aerosols was assessed (MRID 48139303). The kinetics of concentration dependent cholinesterase (ChE) inhibition in red blood cells, plasma, brain and lung was measured. In the special acute inhalation study female rats were exposed nose only to atmospheric concentrations of up to mg/m^3 of particulate chlorpyrifos for six hours and allowed an additional 72 hours to recover (MRID No: 48139303 Hotchkiss *et al*. 2010). The peak inhibition for plasma and lung ChE was at 6 hours post-dosing. Significant lung (47%) and plasma (48%) ChE inhibition were noted at the lowest concentration tested of 3.7 mg/m^3, which is a LOAEL. RBC and brain ChE inhibition were noted at 12.9 mg/m^3 and 53.9 mg/m^3, respectively, indicating they are less sensitive than lung and plasma ChE inhibition following acute inhalation exposures. No NOAEL was established. EPA estimated a human equivalent concentration (HEC) of 0.62 mg/ m^3 based on the LOAEL of 3.7 mg/m3.

The LOAEL is 3.7 mg/m^3 based on lung cholinesterase testing (HEC of 0.62 mg/ m^3 estimated by EPA). A NOAEL was not identified. The classification of this special inhibition study is Acceptable/Non-Guideline.

Appendix B. Physical/Chemical Properties

Physicochemical Properties of Chlorpyrifos.		
Parameter	Value	Reference
Melting point/range	41.5-42.5 ºC	Chlorpyrifos IRED
pH	NR	
Density (21ºC)	1.51 g/mL	
Water solubility (25°C)	1.05 mg/L	
Solvent solubility (20°C)	Acetone >400 g/L Dichloromethane >400 g/L Methanol 250 g/L Ethyl acetate >400 g/L Toluene >400 g/L n-hexane >400 g/L	
Vapor pressure, (25°C)	1.87×10^{-5} torr[1]	
Dissociation constant, pK_a	NR	
Octanol/water partition coefficient, $Log(K_{OW})$	4.7	
UV/visible absorption spectrum	NR	

NR – not reported.

[1] R. Bohaty, June 2011, D368388 and D389480, *Chlorpyrifos Drinking Water Assessment for Registration Review* (CRF assessment, Oct. 16, 2009 product chemistry BC 2062713)

Appendix C. Current US Tolerances and International Residue Limits

Chlorpyrifos (059101)

Summary of US and International Tolerances and Maximum Residue Limits				
Residue Definition:				
US	Canada		Mexico[2]	Codex[3]
40CFR180.342 chlorpyrifos *per se* (*O,O* -diethyl *O* -(3,5,6-trichloro-2-pyridyl) phosphorothioate	*O,O*-diethyl-*O*-(3,5,6-trichloro-2-pyridyl) phosphorothioate (apples, grapes, tomatoes) *O,O*-diethyl-*O*-(3,5,6- trichloro-2-pyridyl) phosphorothioate, including the metabolite 3,5,6-trichloro-2-pyridinol (citrus fruits; fat, kidney, and liver of cattle; kiwifruit; peppers; rutabagas; meat and meat byproducts of cattle (calculated on the fat content)			Chlorpyrifos. The residue is fat soluble.
Commodity[1,]	*Tolerance (ppm) /Maximum Residue Limit (mg/kg)*			
	US	Canada	Mexico[2]	Codex[3]
Alfalfa, forage	3.0			
Alfalfa, hay	13			5 alfalfa fodder
Almond	0.2			0.05
Almond, hulls	12			
Apple	0.01	0.01		1 pome fruits
Apple, wet pomace	0.02			
Banana	0.1			2
Beet, sugar, dried pulp	5.0			
Beet, sugar, molasses	15			
Beet, sugar, roots	1.0			0.05
Beet, sugar, tops	8.0			
Cattle, fat	0.3	1.0		
Cattle, meat	0.05	1.0		1 (fat)
Cattle, meat byproducts	0.05	1.0		0.01 cattle, kidney and liver
Cherry, sweet	1.0			
Cherry, tart	1.0			
Citrus, dried pulp	5.0			
Citrus, oil	20			
Corn, field, forage	8.0			
Corn, field, grain	0.05	0.05		0.05 maize
Corn, field, refined oil	0.25			0.2 maize oil, edible
Corn, field, stover	8.0			10 maize fodder (dry)
Corn, sweet, forage	8.0			
Corn, sweet, kernel plus cob with husk removed	0.05	0.05		0.01 sweet corn (corn-on-the-cob)
Corn, sweet, stover	8.0			
Cotton, undelinted seed	0.2			0.3 cotton seed
Cranberry	1.0			1
Cucumber	0.05	0.05		
Egg	0.01			0.01 (*)
Fig	0.01			

Summary of US and International Tolerances and Maximum Residue Limits				
Residue Definition:				
US	Canada		Mexico[2]	Codex[3]
Fruit, citrus, group 10	1.0	1.0		1
Goat, fat	0.2			
Goat, meat	0.05			
Goat, meat byproducts	0.05			
Hazelnut	0.2			
Hog, fat	0.2			
Hog, meat	0.05			0.02 (fat)
Hog, meat byproducts	0.05			0.01 (*) pig, edible offal
Horse, fat	0.25			
Horse, meat	0.25			
Horse, meat byproducts	0.25			
Kiwifruit	2.0	2.0		
Lettuce	1.0			
Milk, fat (Reflecting 0.01 ppm in whole milk)	0.25			0.02 milk
Nectarine	0.05	0.05		
Onion, bulb	0.5			0.2
Peach	0.05	0.05		0.5
Peanut	0.2			
Peanut, refined oil	0.2			
Pear	0.05			1 pome fruits
Pecan	0.2			0.05 (*)
Pepper	1.0	1.0		2 peppers sweet including pimento or pimiento); 20 peppers chili, dried
Peppermint, tops	0.8			
Peppermint, oil	8.0			
Plum, prune, fresh	0.05			0.5 plums (including prunes)
Poultry, fat	0.1			
Poultry, meat	0.1			0.01 (fat)
Poultry, meat byproducts	0.1			0.01 (*) poultry, edible offal
Pumpkin	0.05			
Radish	2.0			
Rutabaga	0.5	0.5		
Sheep, fat	0.2			
Sheep, meat	0.05			1 (fat)
Sheep, meat byproducts	0.05			0.01 sheep, edible offal
Spearmint, tops	0.8			
Spearmint, oil	8.0			
Sorghum, grain, forage	0.5			
Sorghum, grain, grain	0.5			0.5
Sorghum, grain, stover	2.0			2 sorghum straw and fodder, dry
Soybean, seed	0.3			0.1 soya bean (dry)
Strawberry	0.2			0.3

Summary of US and International Tolerances and Maximum Residue Limits				
Residue Definition:				
US	Canada		Mexico[2]	Codex[3]
Sunflower, seed	0.1	0.1		
Sweet potato, roots	0.05			
Turnip, roots	1.0			
Turnip, tops	0.3			
Vegetable, brassica, leafy, group 5	1.0			2 Broccoli 1 Cabbages, head 0.05 Cauliflower 1 Chinese cabbage (type pe-tsai)
Vegetable, legume, group 6 except soybean	0.05	0.05 lentils		0.01 common bean (pods and/or immature seeds); peas (pods and succulent=immature seeds)
Walnut	0.2			0.05 (*)
Wheat, forage	3.0			
Wheat, grain	0.5			0.5
Wheat, straw	6.0			5 wheat straw and fodder, dry
MRLs with No US Equivalents				
Grapes		0.01		0.5
Tomatoes		0.01		
Carrot				0.1
Coffee beans				0.05
Cotton seed oil, crude				0.05 (*)
Cotton seed oil, edible				0.05 (*)
Dried grapes (=currants, raisins and sultanas)				0.1
Potato				2
Rice				0.5
Soya bean oil, refined				0.03
Tea, green, black (black, fermented and dried)				2
Wheat flour				0.1
Completed: M. Negussie; 04/12/2011				

[1] Includes commodities listed in the CFR as of 4/12/11. The 40CFR 180.342 (a) (3) also stipulates that "a tolerance of 0.1 part per million is established for residues of chlorpyrifos, per se, in or on food commodities (other than those already covered by a higher tolerance as a result of use on growing crops) in food service establishments where food and food products are prepared and served, as a result of the application of chlorpyrifos in microencapsulated form."

[2] Mexico adopts US tolerances and/or Codex MRLs for its export purposes.

[3] * = absent at the limit of quantitation; Po = postharvest treatment, such as treatment of stored grains. PoP = processed postharvest treated commodity, such as processing of treated stored wheat. (fat) = to be measured on the fat portion of the sample. MRLs indicated as proposed have not been finalized by the CCPR and the CAC.

(c) *Tolerances with regional registrations.* Tolerances with regional registration, as defined in 180.1(m), are established for residues of the pesticide chlorpyrifos *per se* (*O,O* -diethyl- *O* -(3,5,6-trichloro-2-pyridyl) phosphorothioate) in or on the following food commodities:

Commodity	Parts per million	Canada	Codex
Asparagus	5.0		
Grape	0.01	0.01	0.5

In addition, the following tolerances for chlorpyrifos are recommended under registration review:

Recommended/Reassessed Tolerances for Chlorpyrifos			
Commodity	**Established Tolerance (ppm)**	**Recommended Tolerance (ppm)**	**Comments** ***Correct Commodity Definition***
Grain, aspirated fractions	NA	22	
Cotton, gin by-products	NA	15	

Appendix D. Review of Human Research

ORE:

The chlorpyrifos occupational residential exposure assessment relies in part on data from studies in which adult human subjects were intentionally exposed to a pesticide or other chemical. These studies, which comprise AHETF, ORETF and PHED, as well as the majority of chemical-specific handler exposure data were determined to require a review of their ethical conduct, have received that review, and have been determined to be ethical. The chemical-specific handler exposure studies that were determined to be ethical and suitable for use in risk assessment are: MRID 42974501, Shurdut, B.A. *et al*. (1993); MRID 43138102, Honeycutt, R.C. & Day, E.W. Jr. (1994); MRID 44739302, Knuteson *et al*. (1999); and MRID 43027901, Contardi *et al*. (1993). A single handler exposure study, "Evaluation of Chlorpyrifos Exposure to Workers During Loading and Application of Lorsban 15 % Granular Insecticide During Corn Planting (MRID 44483501)," was determined to have been conducted in a manner which prohibits its use by the Agency.

Toxicology:

Deliberate dosing studies in adult (non-pregnant) humans are available which measure AChE activity and urinary levels of chlorpyrifos and/or its metabolites. Results from Nolan *et al* (1982; MRID 124144) have been used by the Agency in estimating (i.e. back-calculating) chlorpyrifos exposure based on urinary levels of TCP. This study has also been used to derive a dermal absorption factor in humans. The Nolan *et al* (1982)) study was reviewed by the Human Studies Review Board (June 24-25, 2009; http://www.epa.gov/hsrb/jun-24-25-2009-public-meeting.htm) and found to be scientifically and ethically conducted. EPA also determined that the study was ethically acceptable. Both the FIFRA SAP and HSRB supported the Agency's proposal to use this study for purposes of characterizing biomonitoring studies but not for purposes of deriving points of departure or in directly estimating uncertainty factors. Another intentional human dosing study was reviewed by the June 2009 HSRB (Kisicki *et al* (1999), MRID 44811002) and the HSRB concluded that the study was scientifically (and ethically) conducted. However, EPA ethics review had determined that "EPA is forbidden by 40 CFR §26.1704 to rely on the Kisicki *et al.* study, MRID 44811002, in actions taken under FIFRA or §408 of FFDCA. It is possible that the circumstances and purposes for which you propose to consider it may be such that the provisions of 40 CFR §26.1706 for an exception to the prohibition in 40 CFR §26.1704 may be satisfied." (J. Carley memo dated 5/29/09; http://www.epa.gov/hsrb/files/1d6-ethics-rvw-kisicki-etal-060109.pdf). The Kisicki data has not been used in the preliminary chlorpyrifos human health risk assessment.

Appendix E. Summary Benchmark Dose Values

As a preliminary analysis, the Agency has conducted BMD modeling on selected AChE studies. These studies were selected based on the availability of at least two treatment groups in addition to a control group. In addition, these studies were selected as they represented a variety of ages, lifestages, and durations. In the acute pup studies the Agency has focused on those studies representing rat ages (PND 10 and older) concordant with human post-natal exposure (i.e, birth and older) and durations of exposure.

BMD modeling was not performed on the 21-day dermal study or the subchronic inhalation studies in the rat since the highest doses tested were NOAELs. The recent acute inhalation CCA study (MRID 48139303) was not amenable to BMD analysis because of variability in the data (large standard deviations) and significant inconsistencies in baseline measurements over time.

The Agency has used a decreasing exponential dose-response model similar to that used for the OP and *N*-methyl carbamate cumulative risk assessments and previously reviewed and supported by the FIFRA SAP on several occasions (FIFRA SAP 2001, 2002, 2005a, 2005b, 2008). As shown below, the Agency has used two versions of the decreasing exponential model—R-based code similar to that used in the NMC cumulative risk assessment and the USEPA Benchmark Dose Software, version 2.1.1 (BMDS). The R-based program was derived from software written using version 1.2.1 of the open source statistical programming language R, and is based on methods utilized in the cumulative risk assessments. The Agency's benchmark dose software (BMDS) exponential model includes a family of nested exponential models from which an optimal model (based on statistical and model criteria) can be determined. The flexibility of the nested exponential models is reflected by the number of parameters considered in the models.

OPP has most often used R-based code to develop BMDs for risk assessment of cholinesterase inhibiting pesticides. However, recently, the Agency's BMDS has implemented the decreasing exponential model. As OPP transitions to using BMDS primarily for single chemical assessments, both approaches may be used in some assessments. It is notable that the two approaches provide remarkably consistent results for the selected studies.

Consistent with risk assessment on other OP and NMCs compounds, the Agency has used a benchmark response (BMR) level of 10% and has thus calculated BMD_{10}s and $BMDL_{10}$s. The BMD_{10} is the estimated dose where AChE is inhibited by 10% compared to background. The $BMDL_{10}$ is the lower confidence bound on the BMD_{10}. Extensive analyses conducted as part of the OP cumulative risk assessment (USEPA, 2002) have demonstrated that 10% is a level that can be reliably measured in the majority of rat toxicity studies, and is generally at or near the limit of sensitivity for discerning a statistically significant decrease in AChE activity across the brain compartment and is a response level close to the background AChE level. The Agency uses the BMDL, not the BMD, for use as the PoD since the BMDL accounts for variability of the data (USEPA, 2000). The BMD_{10} provides a point of comparison across studies and the BMD_{10} provides the basis for determining Toxicity Adjustment Factors (TAFs) for chlorpyrifos-oxon. Tables 1-4 provide the results of the BMD analysis of the parent, while Tables 5-8 provide the

results of BMD analysis of the oxon.

Typically, studies submitted for pesticide registration and most studies from the public literature only measure brain and/or blood ChEs. It is rare for data from peripheral tissues to be available for consideration. Chlorpyrifos is unique in that multiple studies are available which provide such peripheral data (Appendix B). Tables 10-13 do not include BMD results for plasma ChE measures. Consistent with OPP's ChE policy, plasma ChE data from animals are used for risk assessment when RBC AChE data are not reliable and/or when peripheral AChE measures are not available. This is not the case for chlorpyrifos; reliable RBC and peripheral data are both available. Thus, the plasma data have not been considered for PoD determination. When conducting BMD analysis for RBC AChE inhibition, the Agency generally starts with the standard BMR of 10% but will consider 15% or 20% in some cases. However, in the case of chlorpyrifos, data from peripheral tissues (e.g., heart, lung, liver) show these tissues are similar in sensitivity to RBC AChE inhibition. As such, when using RBC AChE inhibition as a surrogate for such peripheral data, the BMR of 10% has been used.

For the re-evaluation of endpoint selection for the oral route, OPP considered the quality of the all available studies, both previous and new.

The most robust studies for determining the acute oral PoD are from a new comparative cholinesterase (CCA) study (MRID 48139301) in the rat conducted by the registrant and the results of cholinesterase (ChE) analyses in male PND17 rats performed by EPA's ORD (Moser *et al*, 2006). Both of these studies involved a wide range of doses and provided high quality AChE data. The results of published studies (e.g., Timchalk *et al*. 2006 and Zheng *et al*. 2000) add support the findings of the Dow CCA Study and Moser *et al* (2006).

Table 1. Results of BMD Modeling of Male and Female Rat Pup Brain and RBC ChE Inhibition following a Single Oral Dose of Chlorpyrifos

Dataset	Sex/age	Endpoint/Route	BMD Program/Software			
			R-based Program		EPA BMDS V2.1.1	
			BMD_{10}	$BMDL_{10}$	BMD_{10}	$BMDL_{10}$
Moser *et al*, 2006	Male PND 17	Brain ChE/ Acute Gavage	0.84	0.75	1.89[a]	1.54[a]
Moser *et al*, 2006	Male PND 17	Whole Blood ChE/Acute Gavage	0.38	0.35	0.62[b]	0.43[b]
CCA Study MRID 48139301	Male PND 11	Brain ChE/ Acute Gavage	2.13	1.51	2.13[c]	1.53[c]
CCA Study MRID 48139301	Male PND 11	RBC ChE/ Acute Gavage	0.83	0.66	0.82	0.65
CCA Study MRID 48139301	Male PND 11	Brain ChE/ Acute Milk	(no computation)[d]	(no computation)[d]	4.4	2.4
CCA Study MRID 48139301	Male PND 11	RBC ChE/ Acute Milk	0.5	0.35	0.47	0.36
CCA Study MRID 48139301	Female PND 11	Brain ChE/ Acute Gavage	2.17	1.53	2.18	1.56
CCA Study MRID 48139301	Female PND 11	RBC ChE/ Acute Gavage	0.97	0.76	0.96	0.75
CCA Study MRID 48139301	Female PND 11	Brain ChE/ Acute Milk	1.53	1.03	1.42	0.91
CCA Study MRID 48139301	Female PND 11	RBC ChE/ Acute Milk	0.5	0.35	0.5	0.36

[a]High dose dropped to improve fit.
[b] High dose dropped to improve fit.
[c] P = 0.071.
[d] No computation (technical issues e.g., no convergence).

Table 2. Results of BMD Modeling of Adult Female Rat Brain and RBC ChE Inhibition following a Single Oral Dose of Chlorpyrifos

Dataset	Sex/route	Endpoint/ Route	BMD Program			
			R-based Single-Sex		EPA BMDS V2.1	
			BMD_{10}	$BMDL_{10}$	BMD_{10}	$BMDL_{10}$
CCA MRID 48139301	Adult Female Acute Gavage (8 hr)	Brain	No convergence	No convergence	4.11[a]	2.26[a]
CCA MRID 48139301	Adult Female Acute Gavage (8 hr)	RBC	1.5	1.13	1.9[b]	1.2[b]
CCA MRID 48139301 (a)	Adult Female 12 hr diet (6 pm-6 am)	Brain	(no computation)[c]	(no computation)[c]	4.47 (8 hr after feeding; 20 hr after food introduction)	3.30 (8 hr after feeding; 20 hr after food introduction)
CCA MRID 48139301	Adult Female 12 hr diet (6 pm-6 am)	RBC	0.66	0.55	1.03 (8 hr after feeding; 20 hr after food introduction)	0.6 (8 hr after feeding; 20 hr after food introduction)

[a] The homogeneous variance resulted in a lower BMDL than the model variance model and also provided an acceptable p value..
[b] An acceptable p value was not achieved.
[c] No computation (technical issues e.g., no convergence).

For exposure scenarios longer than acute duration, several high quality oral studies were available for BMD analyses and determination of oral PoDs for short- and intermediate-term incidental oral and chronic dietary scenarios. These included the new CCA study (MRID 48139301) in the rat, a developmental neurotoxicity rat study (MRID 44556901) and a special ChE study in the rat (MRID 44648101).

Table 3. Results of BMD Modeling of Pup Rat Brain and RBC ChE Inhibition following Repeat Oral Doses of Chlorpyrifos

Dataset	Sex/time of dosing	Endpoint/Route	BMD Program			
			R-based Program		EPA BMDS V2.1	
			BMD_{10}	$BMDL_{10}$	BMD_{10}	$BMDL_{10}$
CCA MRID 48139301)	PND 11-25 F (11 days) Gavage corn oil	Brain	0.60	0.48	0.80[a]	0.69[a]
CCA MRID 48139301)	PND 11-25 F (11 days) Gavage corn oil	RBC	0.17	0.15	0.17	0.15
CCA MRID 48139301)	PND 11-25 M (11 days) Gavage corn oil	Brain	0.32	0.3	0.63	0.52
CCA MRID 48139301)	PND 11-25 M (11 days) Gavage corn oil	RBC	0.077	0.04	0.11	0.09

[a] An acceptable p value was not achieved with BMDS program, however, there was good visual fit and value was similar to R-based program.

Table 4. Results of BMD Modeling of Adult Rat Brain, RBC and Heart ChE Inhibition following Repeat Oral Doses of Chlorpyrifos

Dataset	Sex/Time of Dosing	Endpoint/ Route	BMD Program/Software			
			R-based Program		EPA BMDS V2.1	
			BMD_{10}	$BMDL_{10}$	BMD_{10}	$BMDL_{10}$
Dow (Hoberman *et al.* 1998a,b MRID 44556901); Maurissen, 2000	Dams, GD6-20	Brain	0.65	0.51	0.65[a]	0.54[a]
Dow (Hoberman *et al.* 1998a,b MRID 44556901); Maurissen, 2000	Dams, GD6-20	RBC	0.06	0.04	0.06[a]	0.03[a]
Dow (Mattsson *et al.* 1998 44648101): Mattson, 2000	Dams, GD6-20	Brain	Hindbrain 1.1 Forebrain (no computation)[b]	Hindbrain 0.8 Forebrain (no computation)[b]	Hindbrain 1.1 Forebrain 1.2	Hindbrain 0.8 Forebrain 0.98
Dow (Mattsson *et al.* 1998 44648101): Mattson, 2000	Dams, GD6-20	RBC/ Heart	RBC 0.14 Heart 0.30	RBC 0.08 Heart 0.26	RBC 0.14[a] Heart 0.85[c]	RBC 0.08[a] Heart 0.22[c]
Dow (Mattsson *et al.* 1998 44648101): Mattson, 2000	Dams, LD1	Brain	Hindbrain 1.45 Forebrain (no computation)	Hindbrain 0.54 Forebrain (no computation)	Hindbrain 1.33 Forebrain 1.13	Hindbrain 0.65 Forebrain 0.89
Dow (Mattsson *et al.* 1998 44648101): Mattson, 2000	Dams, LD1	RBC	RBC 0.055 Heart 0.23	RBC 0.045 Heart 0.21	RBC 0.050 Heart 0.21	RBC 0.044 Heart 0.18
CCA MRID 48139301)	Adult F (11 days) Gavage corn oil	Brain	(no computation)	(no computation)	1.03 (8 hr)	0.95 (8 hr)
CCA MRID 48139301)	Adult F (11 days) Gavage corn oil	RBC	0.45	0.35	0.45[d]	0.35[d]

[a] The homogeneous variance provided a BMDL value and an acceptable p value.
[b] No computation (technical issues e.g., no convergence).
[c] An acceptable p value was not achieved with BMDS program. Submodel 5 had best AIC and a $BMDL_{10}$ value comparable to R-based program. Submodel 3 had BMD_{10} and $BMDL_{10}$ values similar to R-based runs but not the best AIC value.
[d] An acceptable p value was not achieved with BMDS program, however visual fits were good and values same as R-based program.

Table 5. CCA Acute BMD_{10} /$BMDL_{10}$ results for Chlorpyrifos Oxon: pup rats

	Male	Female
Chlorpyrifos oxon BMD_{10}/$BMDL_{10}$	Brain: 1.06/0.36 RBC: 0.093/0.050	Brain: No reliable fit[a] RBC: 0.081/0.063

[a]No reliable fit with BMDS program and no convergence in R-based program.

Table 6. CCA Acute BMD_{10} /$BMDL_{10}$ results for Chlorpyrifos Oxon: adult rats

	Male	Female
Chlorpyrifos oxon BMD_{10}/$BMDL_{10}$	Brain and RBC: Not examined	Brain; 1.66/0.80[a] RBC: 0.214/0.150

[a]BMD value from r-based program. Submodels 4 and 5 of the BMDS program failed to compute values

Table 7. CCA Chronic (11 day) BMD_{10} /$BMDL_{10}$ results for Chlorpyrifos Oxon: pup rats

	Male	Female
Chlorpyrifos oxon BMD_{10}/$BMDL_{10}$	Brain: No reliable fit[a] RBC: 0.029/0.024	Brain: 0.60/0.13 RBC: 0.027/0.025

[a]No convergence in r-based program. Bad completion or failure to compute BMD value in BMDS submodels 4 and 5.
[b]No acceptable P value with BMDS but good visual fit and comparable to value obtained with R-based program.

Table 8. CCA Chronic (11 day) BMD_{10}/$BMDL_{10}$ for Chlorpyrifos Oxon:adult rats

	Male	Female
Chlorpyrifos oxon BMD_{10}/$BMDL_{10}$	Brain and RBC: Not examined	Brain: No reliable fit [b] RBC: 0.025/0.011 (p=0.08)

[a]No acceptable P value not achieved in BMDS but good visual fit and same value as R-based program.
[b]No convergence in r-based program. Failure to compute BMD value in BMDS submodels 4 and 5.

ATTACHMENT G

Reader's Guide to the Preliminary Human Health Risk Assessment for Chlorpyrifos
Docket # EPA-HQ-OPP-2008-0850

Publication Date: July 1, 2011

Purpose of This Reader's Guide:

The purpose of this note to reader is to highlight what the Agency believes are the key areas of uncertainty and continuing evaluation in its human health assessment of chlorpyrifos.

Health Effects Associated with Chlorpyrifos

Chlorpyrifos is an organophosphate (OP) insecticide that binds to and phosphorylates the enzyme, acetylcholinesterase (AChE), in both the central (brain) and peripheral nervous systems leading to accumulation of acetylcholine and, ultimately, to clinical signs of toxicity at sufficiently high doses. In 2000, the Agency concluded for chlorpyrifos that inhibition of AChE was the most sensitive effect in all of the animal species evaluated (rats, mice, rabbits, and dogs) and in humans, regardless of exposure duration. The Agency is maintaining at this time, based on available data, that AChE inhibition, particularly in blood, still provides the most sensitive dose-response data for the chlorpyrifos human health risk assessment.

There is, however, a growing body of literature with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent behavioral effects into adulthood. The results of both *in vivo* and *in vitro* studies on chlorpyrifos have led some research groups to propose that changes in brain neurochemistry may underlie behavioral changes into adulthood. Although there are several biologically plausible hypotheses for these changes, a mode of action, including key events, temporal and dose-response concordance, has not yet been described.

In addition, there are three prospective epidemiology cohort studies evaluating pre- and post-natal pesticide, chlorpyrifos or OP, exposure in mother-infant pairs that have reported associations with birth outcomes, childhood neurobehavioral and neurodevelopment outcomes in neonates, infants, and young children. The Agency has not yet performed a comprehensive weight of evidence analysis on these studies, but based on preliminary reviews, there appears to be consistency across three studies in reported behavioral delays in cognitive achievement, motor control, social behavior, and intelligence measures. The Agency has not completed its integrated analysis of the epidemiologic studies with experimental toxicology studies. Thus, definitive conclusions regarding to what extent the reported neurodevelopmental effects in the epidemiologic studies can be attributed to chlorpyrifos exposure are not presented in the preliminary risk assessment.

Key considerations As You Review the Preliminary Risk Assessment:

Human Health Risk Assessment:

- Previously published results from the three epidemiology studies were reviewed and discussed by the FIFRA Scientific Advisory Panel (SAP) in 2008[1]. At that time, the SAP concluded these epidemiological studies have utility for risk assessment but only in a qualitative manner, and not as the principal basis for quantitative risk assessment.
- Recently, study results from the same three cohort studies were published for 7 year olds. The Agency is evaluating the current epidemiology database with experimental laboratory animal data using the draft framework reviewed by the SAP in 2010[2].
- The Agency plans to conduct a full weight of evidence evaluation integrating the epidemiology studies with the experimental toxicology studies for the neurodevelopmental outcomes using the *Draft Framework for Incorporating Epidemiologic and Human Incidence Data in Human Health Risk Assessment*[3]. Such a weight of evidence analysis requires explicit consideration of such criteria as strength, consistency, specificity, dose response, temporal concordance and biological plausibility.
- Due to the preliminary nature of this assessment, the Agency is providing assessments reflecting both the retention of the 10X FQPA Safety Factor as presented in the previous June 2000 assessment, and a proposal to reduce the FQPA Safety Factor to 1X based on recently submitted data. The Agency will continue to reevaluate the existing data to determine whether a reduction of the FQPA Safety Factor is appropriate in the final risk assessment.

Drinking Water Assessment

- Estimated concentrations of chlorpyrifos in source water are based on modeling. Currently available monitoring data is judged to be insufficient to capture peak, daily water concentrations and do not represent high use areas of chlorpyrifos.
- As the Agency moves forward to finalize the risk assessment, the Agency will further characterize estimated chlorpyrifos concentrations in source water by considering more typical use rates and agronomic practices for all chlorpyrifos uses.
- Laboratory-scale experiments indicate rapid conversion of chlorpyrifos to chlorpyrifos oxon (more toxic metabolite) during chlorination; therefore quantitative (100%) conversion of chlorpyrifos to chlorpyrifos oxon is assumed in drinking water utilities. Because chlorpyrifos oxon is more toxic than

[1] http://www.epa.gov/scipoly/sap/meetings/2008/091608_mtg.htm
[2] http://www.epa.gov/scipoly/sap/meetings/2010/020210meeting.html
[3] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0851-0004

chlorpyrifos, this preliminary assessment considers the oxon metabolite as the residue of concern in treated drinking water.

- The impact of alternative drinking water treatment processes on chlorpyrifos and chlorpyrifos oxon concentrations in finished drinking water is an area of uncertainty. In the final assessment the Agency will attempt to further characterize the effect of treatment processes on estimated chlorpyrifos oxon concentrations in finished drinking water.
- The Agency is asking for any pertinent data and information that could inform or possibly revise existing monitoring data, model input data or the assumption that chlorpyrifos is completely oxidized to the oxon metabolite during drinking water treatment and distribution to end users.

Residential Bystander Post-application Inhalation Exposure

- Recently, the Agency has begun to explore the development of an approach for assessing inhalation exposure resulting from the field volatilization of conventional pesticides based on recommendations provided by the SAP in December 2009[4].
- In the preliminary risk assessment, the Agency has assessed residential bystander exposure from field volatilization of applied chlorpyrifos based on available ambient and application site air monitoring data; application site studies were conducted in California and Washington.
- The limitations and assumptions have been highlighted in the assessment and the Agency seeks public comment on those assumptions. In addition, the Agency will continue to evaluate the 2009 SAP comments and where appropriate, the Agency will revise the approach, which may warrant a reanalysis of the preliminary residential bystander exposure and risk assessment for chlorpyrifos.

Current Status and Next Steps

- This preliminary human health risk assessment on chlorpyrifos is being presented for public comment for a period of 60-days to seek input on the above issues.
- EPA is asking for any pertinent data and information that will inform or possibly revise the preliminary human health risk assessment.
- After the comment period, the Agency plans to fully analyze and integrate all the available scientific data and considerations of public comments to ensure a scientifically sound, technically robust, human health risk assessment for chlorpyrifos.

How to Submit Comments

Comments on risk mitigation options for chlorpyrifos must be submitted to EPA no later than September 5, 2011. Submit your comments, identified by docket number EPA-HQ-OPP-2008-0850, by one of the following methods.

[4] http://www.epa.gov/scipoly/sap/meetings/2009/december/120309meetingminutes.pdf

Federal eRulemaking Portal, http://www.regulations.gov : Follow the on-line instructions for submitting comments. Alternatively, paste this address into your browser:
http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2008-0850

- Mail: Office of Pesticide Programs (OPP), Regulatory Public Docket (7502P), Environmental Protection Agency, 1200 Pennsylvania Avenue, NW, Washington, DC 20460-0001.

- Delivery: OPP Regulatory Public Docket (7502P), Environmental Protection Agency, Room S-4400, One Potomac Yard (South Building), 2777 S. Crystal Drive, Arlington, VA. Deliveries are accepted only during the Docket's normal hours of operation (8:30 a.m. to 4:00 p.m., Monday through Friday, excluding legal holidays). Special arrangements should be made for deliveries of boxed information. The Docket telephone number is 703-305-5805.

Contact

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ATTACHMENT H

Meeting of the FIFRA Scientific Advisory Panel

Draft Issue Paper: Scientific Issues Concerning Health Effects of Chlorpyrifos

April 10-13, 2012

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1.0 Introduction

Chlorpyrifos (*0*,*0*-diethyl-*0*-3,5,6-trichloro -2-pyridyl phosphorothioate) is a broad-spectrum, chlorinated organophosphate (OP) insecticide. In 2000, nearly all residential uses were voluntarily cancelled by Dow AgroSciences but agricultural use remains. The 2000 human health risk assessment was largely based on adult laboratory animal data (rat or dog) for cholinesterase (ChE) inhibition and the application of default uncertainty factors (U.S. Environmental Protection Agency, 2000). In 2008, the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP) reviewed a draft science issue paper on the human health effects of chlorpyrifos which provided a preliminary review of the scientific literature on experimental toxicology and epidemiology studies available since the 2000 risk assessment. This draft issue paper considered a growing body of literature with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent effects into adulthood in addition to epidemiology studies evaluating prenatal chlorpyrifos exposure in mother-infant pairs that have reported associations with birth outcomes, childhood neurobehavioral and neurodevelopment outcomes in the offspring when evaluated in neonates, infants, and young children. In 2008, the SAP agreed with the Agency that although the epidemiology studies were high quality with respect to design, conduct and analyses and provided information for hazard characterization, ChE inhibition remained the most robust and sensitive data for deriving points of departure. In 2011, the Agency released a preliminary human health risk assessment for chlorpyrifos. The focus of the 2011 risk assessment was on the ChE inhibiting potential of chlorpyrifos including in young animals. Like other OPs, chlorpyrifos binds to and phosphorylates the enzyme acetylcholinesterase (AChE) in both the central (brain) and peripheral nervous systems. This can lead to accumulation of acetylcholine and, ultimately, at sufficiently high doses, to clinical signs of toxicity. Consistent with the focus on ChE inhibition, in the 2011 preliminary risk assessment, EPA evaluated the extensive database of ChE data for multiple lifestages and selected points of departure based on consideration of all quality and reliable data.

In 2010, the Agency developed a draft "Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment" which provides the foundation for evaluating multiple lines of scientific evidence in the context of the understanding of the adverse outcome pathway (or mode of action; (U.S. Environmental Protection Agency, 2010). The draft framework was reviewed favorably by the SAP in 2010 (FIFRA Scientific Advisory Panel (SAP), 2010). Mode of action (Boobis et al., 2006; Boobis et al., 2008; U.S. Environmental Protection Agency, 2005) and adverse outcome pathway (Ankley et al., 2010) provide important concepts in this integrative analysis. Both a mode of action and an adverse outcome pathway are based on the premise that an adverse effect caused by exposure to a compound can be described by a series of causally linked biological key events that result in an adverse human health or ecological outcome. One of the key components of the Agency's draft framework is the use the MOA framework /AOP concept as a tool for organizing and integrating information from different sources to inform the causal nature of links observed in both experimental and observational studies. Specifically, the modified Bradford Hill Criteria are used to evaluate the experimental support that establishes key events within a mode of action or an adverse outcome pathway, and explicitly considers such concepts as strength, consistency, dose response, temporal concordance and biological plausibility in a weight of evidence analysis.

The draft framework is oriented around the source to outcome pathway (Figure 1) as discussed in the National Research Council's (NRC) report on toxicity testing in the 21st Century (National Research Council, 2007). Since the 2008 SAP on chlorpyrifos, the Agency has reviewed new experimental

toxicology studies evaluating adverse effects in laboratory animals and performed further analyses on the existing and new epidemiology results in mothers and children along with relevant biomonitoring data. In addition, the Agency has reviewed *in vivo* and *in vitro* studies evaluating mechanistic aspects of chlorpyrifos evaluating proposed adverse outcome pathways related to the effects on the developing brain. The Agency is in the process of developing the weight of evidence analysis integrating the epidemiology studies with the experimental toxicology studies for chlorpyrifos; the 2012 SAP review is a key milestone in the development of the weight of evidence analysis. This 2012 draft issue paper includes review of scientific information in the various areas provided in Figure 1: chlorpyrifos exposure, pharmacokinetics, toxicity pathways, and effects on individuals and/or populations. The state of the science on each of these areas is summarized in this document.

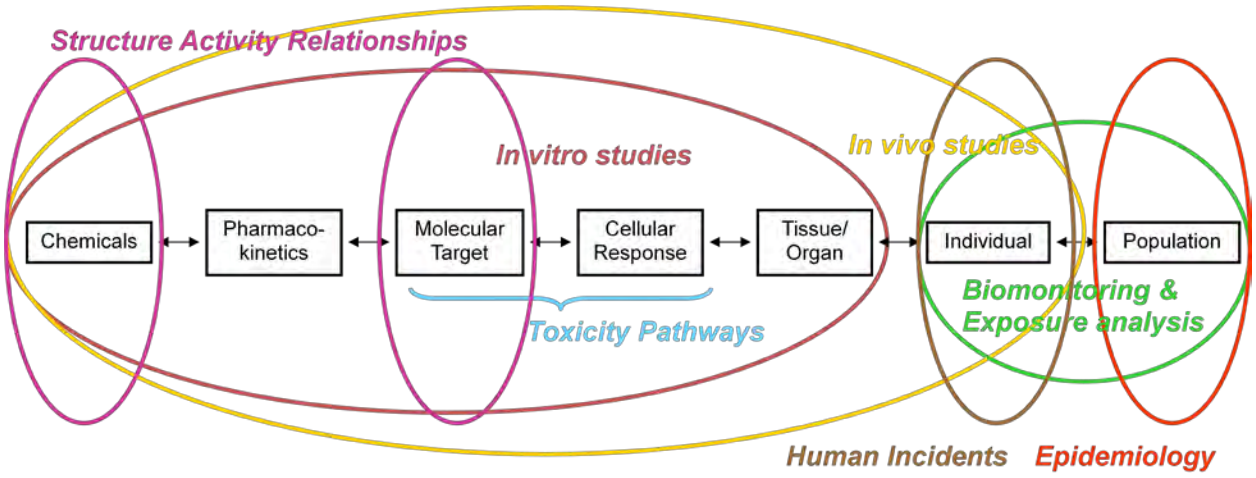


Figure 1 Source to effects pathway (adapted from NRC, 2007).

This draft science issue paper is organized in a manner consistent with the source-to-outcome paradigm (Figure 1). As such, the document begins with a summary of the exposure pathways which exist for chlorpyrifos to provide the Panel and the public with scientific context on the routes and duration of exposure for chlorpyrifos in addition to the potentially sensitive lifestages. These exposure pathways, potentially sensitive lifestages and relevant toxicity pathways and health outcomes are organized in a conceptual framework which provides the foundation for the Agency's analysis (Chapter 2). The AChE inhibiting mode of action for chlorpyrifos (Section 3.1) along with several biologically plausible hypotheses on modes of action leading to effects on brain development are discussed (Section 3.2.1). The next sections of the document consider the adverse effects observed in laboratory animals (Section 3.2.2) epidemiology studies with mothers and children (Chapter 4). Chapter 5 discusses biomonitoring data available for chlorpyrifos along with the state of the science with respect to physiologically based pharmacokinetic modeling (PBPK) and possible approaches for interpreting biomonitoring data. The Agency is soliciting comment from the SAP on scientific issues related to the interpretation of epidemiology, biomonitoring data, and experimental toxicology studies as the Agency prepares to integrate information and data from these scientific areas in a weight of the evidence analysis considering the extent to which chlorpyrifos may cause long term adverse effects from early life exposure (*i.e.*, gestational, postnatal) and that such effects may (or may not) occur below doses established from ChE inhibition including in postnatal animals and used for regulatory purposes.

2.0 Source-to-Outcome Considerations

2.1 Exposure Pathways for Chlorpyrifos

Chlorpyrifos is an insecticide, acaricide and miticide that controls many insect pests. Chlorpyrifos was first registered in 1965 for use in a variety of food and feed crops. As chlorpyrifos use expanded in agriculture, use also grew in the residential marketplace. For several years thereafter, it maintained a large percentage of the residential marketshare. Residential product labels at that time allowed for use indoors as a pest control product. Examples of how it was used included: indoor space and surface sprays; as a lawn treatment; as a termiticide; for the control of fleas and ticks on pets; and for treatments of residential turf and gardens. The risks from these broad uses of chlorpyrifos were evaluated by the Agency in a 2000 risk assessment (Smegal and Leighton, 1999).[1] The outcome of this assessment was that most residential uses of chlorpyrifos were phased-out (sales in those markets ended by December 2001).[2] Currently, chlorpyrifos is still widely used in agriculture and it still has a variety of remaining non-agricultural uses. Agricultural uses can occur on a variety of food and feed crops including grapes, many vegetable and fruit row crops (*e.g.*, cauliflower, strawberry), tree crops (*e.g.*, citrus and many nut varieties), and field crops (*e.g.*, sorghum, alfalfa). Non-agricultural uses include in industrial settings (*e.g.*, industrial plants, railcars, warehouses), on ornamental plants and in their production, on some types of turf (*e.g.*, sod and golf course), and on wood products (*e.g.*, logs, pallets, utility poles). Some non-agricultural products are highly specialized for use in niche markets including cattle ear tags, as a mosquito adulticide, and in residential ant bait stations that are sold in child resistant packages.

Chlorpyrifos is being reevaluated under the Registration Review process,[3] and a preliminary risk assessment has recently been completed that reflects the current use patterns (U.S. Environmental Protection Agency, 2011). Risk assessments for pesticides address all manner of exposure pathways as well as all pertinent routes and durations of exposure. Metabolites and/or degradate compounds found to be of significance are also considered as appropriate. The oxygen analog of chlorpyrifos, referred to as its oxon, is known to form *in vivo* in humans after exposure and in the environment after application which can lead to exposures. The oxon of chlorpyrifos is also known to be more toxic than chlorpyrifos, *per se*. The pertinent sources of the oxon and the levels at which direct human exposures can occur are described in detail in the Agency's 2011 risk assessment.

The Food Quality Protection Act (FQPA) of 1996 requires that the Agency complete risk assessments that consider all possible exposure routes and pathways and to aggregate the resulting exposures as appropriate. This ensures that all sources of exposure for a particular pesticide are reasonably considered in regulatory decision making. In this approach, exposures through the diet, from drinking water, and residential sources are calculated. Risk assessment for chlorpyrifos considers exposures through the diet, from drinking water, and from all other sources that can lead to exposures in the general public (referred to as residential exposure). Residential exposures occur because people buy products to treat a pest inside their homes or outside (*e.g.*, on lawns). They can also occur because people live in or near treated environments where they can be exposed because residues move away

[1] http://www.epa.gov/pesticides/chem_search/cleared_reviews/csr_PC-059101_5-Oct-99_426.pdf
[2] http://www.epa.gov/oppsrrd1/REDs/factsheets/chlorpyrifos_fs.htm
[3] http://www.epa.gov/oppsrrd1/registration_review/chlorpyrifos/index.htm

from their intended application area (*e.g.*, through spray drift during application or by volatization from treated areas). They can also occur because they frequent areas (*e.g.*, golf course). Dietary and drinking water exposures are calculated using information which couples how much of a commodity (or drinking water) is consumed based on survey data with possible residues in the food or water which is consumed. Consumption is defined using a national food intake survey. Commodity residue values are based on monitoring data but can be refined using many techniques (*e.g.*, at what point is commodity sampled in distribution chain, how much of a given crop is treated with a particular pesticide). Residential exposures are calculated using a scenario-driven approach which reflects anticipated behaviors for different aged children and adults in various residential settings. Exposures are calculated using methodologies that couple what is known about the use of a pesticide and available information on its residues in the environment with the scenarios that correctly match how the product can be used.

Risk assessments also consider occupational exposures individuals can receive through their jobs. For chlorpyrifos, as is typical for most agricultural use pesticides, such occupational exposures can occur for those involved in the application of a pesticide and those who may be exposed because they are work in areas that have been previously treated (*e.g.*, while harvesting fruit or vegetables). Occupational exposures are evaluated based on the requirements of the FIFRA. In a manner analogous to residential exposures described above, occupational exposures are also evaluated using a scenario based process. Scenarios are defined by the types of crops (or non-agricultural areas) and pests that a pesticide is used on coupled with any restrictions regarding its use (*e.g.*, certain requirements for protective clothing). For those involved in application, a series of standardized exposure rates (*i.e.*, referred to as unit exposure values[4]) and standard production throughput values (*e.g.*, acres treated per day are commonly used for agriculture) are coupled with specific information about each crop where a pesticide can be used (*e.g.*, application rates and levels of personal protection required). Exposures for those who work in previously treated areas are evaluated using a similar approach. Exposure rates associated with the type of crop and activity individual applicators may perform (*i.e.*, known as transfer coefficients[5]) are coupled with specific information related to how persistent residues are for a pesticide in the environment (*i.e.*, typically referred to as dislodgeable foliar residues) to calculate risks. The results of these types of assessments assist in defining risk management approaches for occupational tasks. Such approaches might entail requiring additional protective clothing or equipment or extending periods after application to allow residues to dissipate before allowing certain tasks because exposures are too great.

The exposure and risk assessment methods used by the Agency have been extensively vetted using mandated public peer review processes. They are also based on guidance used across the Agency and throughout the Federal government.[6,7] Additionally, testing guidelines have been established for developing the data required to evaluate pesticides for regulatory purposes which have also undergone extensive review and input.[8] Science policy papers have also been developed describing how input values should be developed and results should be interpreted.[9] Specifically, dietary and drinking water exposure and risk assessment methods have been peer reviewed by the SAP on several occasions.[10,11]

[4] http://epa.gov/pesticides/science/handler-exposure-data.html
[5] http://www.epa.gov/pesticides/science/post-app-exposure-data.html
[6] http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=15263
[7] http://www.epa.gov/risk_assessment/history.htm
[8] http://www.epa.gov/pesticides/science/guidelines.htm
[9] http://www.epa.gov/oppfead1/trac/science/
[10] http://www.epa.gov/scipoly/sap/tools/atozindex/dietexp.htm

Similarly, the methods used to evaluate both residential and occupational exposures have also been extensively peer reviewed.[12]

For detailed information documenting the kinds of exposures identified for chlorpyrifos and the calculated exposure values, please refer to Appendix 1. Residential risk estimates calculated prior to the removal of those uses from the label in 2001 are presented. These are included because some cohorts of interest in this analysis were initiated prior to such uses being removed from the label (*e.g.*, Columbia University cohort of lower income women in New York City was initiated in 1997). The remaining values included in Appendix 1 are based the 2011 risk assessment. The values in Appendix 1 represent a subset of exposure scenarios considered in the 2011 risk assessment; these have been selected as they are a representative cross section of current uses of chlorpyrifos. The Agency is not soliciting comment from the Panel on the methods or data used to estimate the values in Appendix 1; these are provided for informational purposes only in order to provide context for the discussions below regarding exposure. [Please refer to the 2011 risk assessment for more information concerning the exposure potential of chlorpyrifos associated with current use practices].

2.2 Conceptual Framework

As an aid in organizing the available information and to identify the complex scientific issues being considered, the Agency developed a conceptual framework (Figure 2). This conceptual framework is consistent with the source-to-outcome pathway (Figure 1) and provides the foundation for the analysis being undertaken by the Agency to delineate outcomes derived from AChE inhibition in different lifestages and from alternative modes/mechanisms. This conceptual framework provides key information on which lifestages are thought be the most susceptible to chlorpyrifos, the exposure pathway by which these individuals are exposed, possible adverse outcome pathways and related critical duration(s) of exposure leading from exposure to adverse health outcome.

[11] http://www.epa.gov/scipoly/sap/tools/atozindex/drinkrisk.htm
[12] http://www.epa.gov/scipoly/sap/tools/atozindex/workerexposure.htm, http://www.epa.gov/scipoly/sap/tools/atozindex/residentexp.htm, http://www.epa.gov/scipoly/sap/meetings/2009/100609meeting.html, and http://www.epa.gov/scipoly/sap/meetings/2008/120208_mtg.htm

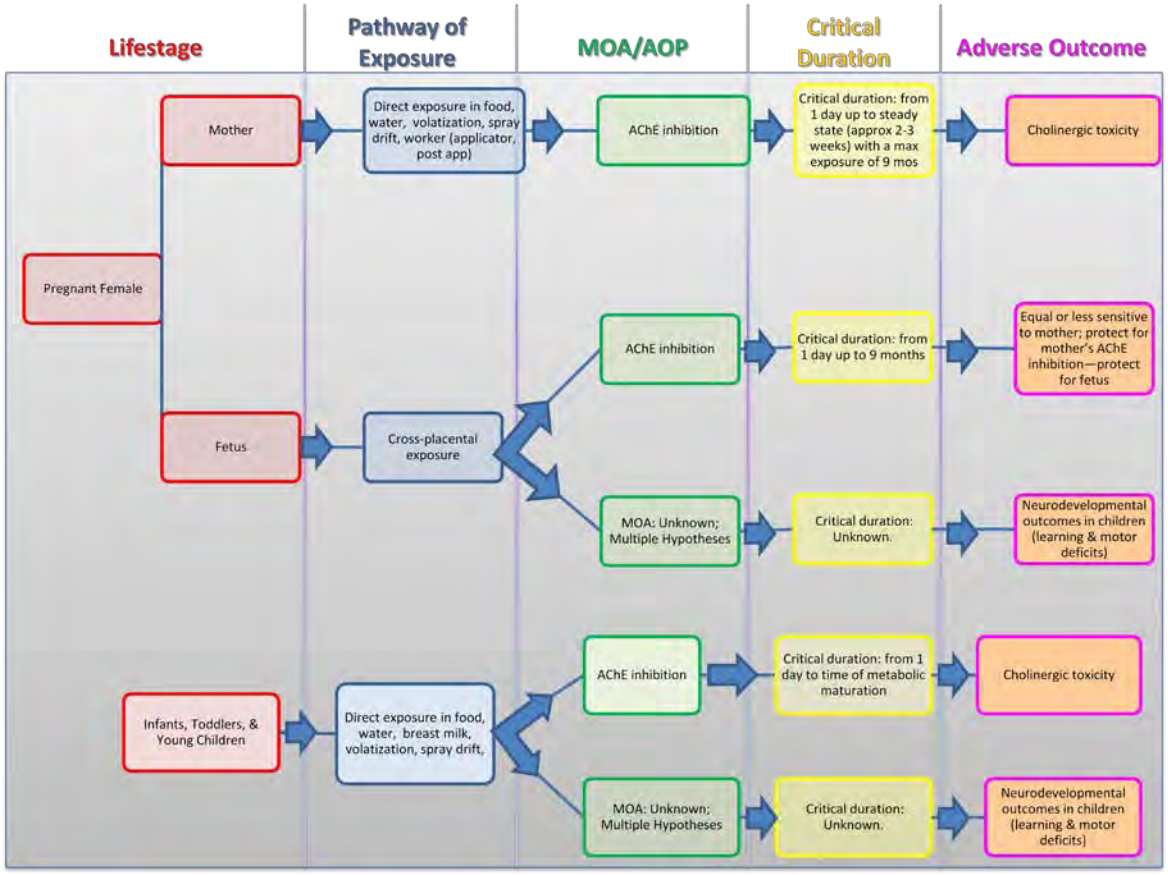


Figure 2 Conceptual framework considering chlorpyrifos exposure, lifestages, critical duration, modes of action/adverse outcome pathways, and health outcomes of interest.

2.2.1. Adverse Outcomes

AChE inhibition is the well-established mode of action for OPs and is typically used as the critical effect in hazard characterization for members of this class of pesticides. The 2008 SAP concurred with the Agency that AChE/ChE data when taking into account sensitive lifestages remains the most robust dose response data for use in derivation of points of departure. However, over the last 10-15 years, experimental toxicology studies on neurotoxicological as well as epidemiology studies have been published which suggest that the developing brain of the fetus and young children may also be affected. There are a significant number of literature studies evaluating neurobehavioral outcomes in experimental animals (rats, mice). These studies vary in their study design but many involve gestational and/or early postnatal dosing with behavioral evaluation in adulthood. Epidemiological studies from three cohorts of mothers and children, funded in part by EPA and National Institutes of Environmental Health, have reported birth and neurodevelopmental outcomes associated with prenatal exposures to OPs. Two of these (Mt. Sinai and Berkeley) have focused primarily upon non-specific urinary biomarkers of OP exposure, the dialkyl phosphate metabolites (*i.e.*, DAPs), and thus present some uncertainty as to the extent these results relate to chlorpyrifos *per se*. The Columbia University investigators are studying a cohort of mothers and children in inner-city New York and have published multiple papers on associations among level of prenatal chlorpyrifos exposure and birth and neurodevelopmental outcomes at multiple time points from birth through age seven years. With respect to neurodevelopmental outcomes, multiple investigators have reported hypothesized modes/mechanisms

of action. In the conceptual framework, both adverse health outcomes of interest for chlorpyrifos are being tracked—AChE inhibition and potential for neurodevelopmental effects.

2.2.2. Potentially Sensitive Lifestages

The Agency believes that pregnant women and their fetuses, newborns, and young children represent potentially the most sensitive lifestages for exposure to chlorpyrifos. This conclusion is based on multiple lines of evidence.

- *Pregnant Women:*
  Chlorpyrifos RBC AChE data from pregnant rats provides the most sensitive data for use in deriving a point of departure from repeated dosing studies. Although it is unclear whether metabolic changes during pregnancy are sufficient to affect metabolism at environmental exposure, some have suggested that there may be reduced ability to detoxify chlorpyrifos and/or the oxon may affect sensitivity during pregnancy.

  Metabolic activities can be altered during pregnancy (Anderson, 2006; Anger & Piquette-Miller, 2008; Bologa, Tang, Klein, Tesoro, & Koren, 1991; Carpintero, Sanchez-Martin, Cabezas-Delamare, & Cabezas, 1996; Czekaj, Wiaderkiewicz, Florek, & Wiaderkiewicz, 2000, 2005; Dickmann et al., 2008; Ejiri, Katayama, Kiyosawa, Baba, & Doi, 2005; Ferre et al., 2006; Hines, 2007; Homma et al., 2000; R. M. Howard & Sugden, 1993; Tsutsumi et al., 2001). For example, Chanda et al. (2002) showed that pregnant female rats had lower plasma, brain, and liver carboxylesterase activity compared to non pregnant females. Regarding A-esterase activity, Ferre *et al.* (2006) showed that the paraoxonase (*i.e.*, A-esterase in serum decreased from a nonpregnant background of 146 U/L to 111 U/L in late gestation, indicating 76% of normal activity in late gestation pregnant women. Carpintero et al. (1996), however, found that phenyl acetate metabolism increased from 23.6 to 33.5 μkat/g in the third trimester, but it must be kept in mind that phenyl acetate metabolism is not necessarily a specific measure of A-esterase activity. Data in mice support the findings of Ferre et al. (2006) in humans suggesting a reduction in A-esterase activity during pregnancy. In mice, Weitman et al. (1983) found that PON1 activity after exposure to parathion was 50 nmol/min/ml in non-pregnant females, but it decreased as low as 14 nmol/min/ml during gestation (Weitman, et al., 1983). With regard to plasma butylcholinesterase (BuChE) activity, Howard et al. (1978) have shown that in six healthy pregnant women levels of plasma BuChE activity dropped by approximately 30% during the first trimester, but returned to close to pre-pregnancy levels in the third trimester. Similarly, other investigators have also reported decreases in plasma BuChE activity in pregnant women (de Peyster, Willis, & Liebhaber, 1994; Venkataraman, Iyer, Narayanan, & Joseph, 1990; Whittaker, Crawford, & Lewis, 1988). Evans et al. (1988) showed that serum ChE levels in 39 of 44 pregnant women dropped after conception; in 20 of those women, the decline in ChE activity continued throughout pregnancy.

  Pregnancy is a remarkably dynamic biological process in which rapid changes occurring in both the developing system (embryo/fetus) and the mother can significantly impact the pharmacokinetics of chemicals. In addition to the potential for decreased clearance of chemicals due to immature metabolic systems in the developing embryo/fetus (mentioned above), other pregnancy-related changes can also have a pronounced effect on pharmacokinetics. For a typical human pregnancy, total body weight gain is in the order of 10-30% while cardiac output can increase as much as 50% (Corley, Mast, Carney, Rogers, & Daston, 2003; Young et al., 1997).

A significant fraction of the body weight gain is due to increases in total body water and fat that can lead to a greater volume of distribution by simple dilution. For instance, increases in total body water dilute plasma proteins which can increase the free fraction of highly plasma protein-bound chemicals (*e.g.*, chlorpyrifos) available for distribution. Increases in fat, on the other hand, can provide a larger storage volume for lipophilic chemicals (Corley, et al., 2003). Additionally, significant changes take place in the maternal circulatory system to accommodate the development of placental blood flow which undergoes considerable increases throughout pregnancy along with embryo/fetus which increases in volume over a billion-fold from conception to birth (Young, et al., 1997).

- *Fetuses, Infants, Toddlers & Young Children:*
  With respect to AChE inhibition, there are multiple studies on chlorpyrifos (Appendix 2) and other OPs (U.S. Environmental Protection Agency, 2006) which show that the pregnant dam exhibits similar or more AChE inhibition than the fetus at a given dose to the dam. As such, for AChE inhibition, by protecting against AChE inhibition in the pregnant female is expected to be protective for AChE inhibition in the fetus. With respect to neurodevelopmental outcomes, as discussed in Section 4.0 below, numerous epidemiological investigations have observed a link between prenatal, and notably not post-natal, exposure to chlorpyrifos (measured as parent, 3,5,6-trichloro-2-pyridinol [TCPy], or DAPs) and adverse effects on fetal growth and neurodevelopment through age seven years. Within this database, notable consistencies are observed and areas of inconsistencies may be attributed to differences in study methods (*i.e.*, measurement of surrogate for prenatal chlorpyrifos exposure), among other sources of variablity. Overall, these studies are strong, well-conducted studies in which likely sources of error or bias would more likely tend to underestimate, rather than overestimate an effect size (*e.g.*, relative risk measure). However, additional analyses are needed to determine with certainty whether chlorpyrifos is the etiological causative agent in the adverse neurodevelopmental outcomes observed. In addition, there a growing body of literature with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent effects into adulthood. The results of both *in vivo* and *in vitro* studies on chlorpyrifos have led some research groups to propose that changes in brain connectivity and/or neurochemistry may underlie these changes into adulthood.

  Potential sensitivity, particularly from acute, single dose exposures, is largely derived from immature metabolic systems that have less capacity to detoxify the parent OP and its toxicologically active metabolite chlorpyrifos oxon. Differential inhibition of the AChE enzyme itself does not appear to account for the observed age-related sensitivity found in young animals as suggested by *in vitro* studies (Atterberry, Burnett, & Chambers, 1997; Benke & Murphy, 1975; Chanda, Harp, Liu, & Pope, 1995; Mortensen, Chanda, Hooper, & Padilla, 1996). Rat fetuses and juveniles and newborn humans, however, have lower capacity to detoxify than adults. This decreased capacity to detoxify has been associated with increased sensitivity in rats. Specifically, in rats, A-esterase activity is virtually nonexistent in the fetus (Lassiter et al., 1998) and increases from birth to reach adult levels around PND21 (W. F. Li, Matthews, Disteche, Costa, & Furlong, 1997; Mortensen, et al., 1996). Mortenson et al. (1996) showed that in the plasma level of CPOase[13] was 1/11 that of adult animals. The animal data regarding the role of carboxylesterase in mediating OP toxicity are also quite extensive (*e.g.*, Clement, 1984; Fonnum, Sterri, Aas, & Johnsen, 1985; Maxwell, 1992a, 1992b). Fetal rats possess very little

[13] CPOase is A-esterase (PON1) activity specific to chlorpyrifos oxon

carboxylesterase activity (Lassiter, et al., 1998) with increasing activity as the postnatal rat matures, reaching adult values after puberty (50 days-of-age; Karanth & Pope, 2000; Morgan, Yan, Greenway, & Parkinson, 1994; Moser, Chanda, Mortensen, & Padilla, 1998). The temporal pattern of A-esterase and carboxylesterase activities correlates reasonably well with studies on OP sensitivity. Several studies have shown an increased sensitivity of newborn rats to OP compounds, and specifically chlorpyrifos, which are detoxified via the A-esterase and/or carboxylesterase pathways (Benke & Murphy, 1975; Chambers & Carr, 1993; Chanda, et al., 2002; Gagne & Brodeur, 1972; Karanth & Pope, 2000; Morgan, et al., 1994; Mortensen, et al., 1996; Moser, et al., 1998; C. N. Pope, Chakraborti, Chapman, Farrar, & Arthun, 1991). Although maturation of the cytochrome P450s may also play a role in age-related differences in the ability of young and adults to detoxify or activate chlorpyrifos to the oxon, the degree to which this maturation impacts juvenile sensitivity to chlorpyrifos has not been extensively studied.

There are, however, relatively little data in human tissues which could evaluate age-related maturation of expression of these detoxification esterases. For example, Pope et al. (2005) evaluated maturational expression of liver carboxylesterases in human liver tissues from infants (2–24 months) and adults (20–36 years). The authors report relatively small (and not statistically significant) differences in activities between children ages 2–24 months and adults (20–36 years). More recently, others have measured expression and activity of human carboxylesterases 1 and 2 in human fetuses, newborns, and children, and while there are considerable inter-individual differences, the lowest activities are recorded at the youngest ages (Yang et al., 2009; Zhu, Appel, Jiang, & Markowitz, 2009).

There are also studies in the literature that have assessed A-esterase (also known as PON1) activity in children. Based on these studies, it appears that serum A-esterase levels are very low in human infants compared to adults (Augustinsson & Barr, 1963; Augustinsson & Brody, 1962; J. Chen, Kumar, Chan, Berkowitz, & Wetmur, 2003; Cole et al., 2003; Holland et al., 2006; Mueller et al., 1983). After birth, there is a steady increase of this activity (Augustinsson & Barr, 1963) with approximately a 2x increase in A-esterase activity between birth and adulthood (Ecobichon & Stephens, 1973). In a related study of the age-dependence of total serum arylesterase (ARase) activity (of which a large component is A-esterase activity), adult levels were achieved by two years-of-age (Burlina, Michielin, & Galzigna, 1977). Recent studies have shown age-dependant increases in PON1 activity from birth to aged 7, and that by age 7 the activity were similar to or slightly less than mothers in the study (Huen et al., 2011; Huen, Harley, Bradman, Eskenazi, & Holland, 2010; Huen et al., 2009). In addition, other studies (J. Chen, et al., 2003; Holland, et al., 2006) have provided A-esterase and/or carboxylesterase activities in newborns and their mothers which show that newborns have lower activities than their mothers. Thus, the lower detoxification potential of chlorpyrifos and other OPs in young rats is also suggested in humans, leading to the potential of greater sensitivity of infants and children due to kinetic factors.

There are critical windows of vulnerability (Rice & Barone, 2000; Rodier, 2004) with regard to toxicant effects on brain development. This vulnerable period in humans spans early pregnancy to adolescence (Rice & Barone, 2000). In fact, recent evidence shows that synapse formation peaks quite late in human brain development at 4-8 years of age (Glantz, Gilmore, Hamer, Lieberman, & Jarskog, 2007). Within these vulnerable periods there are key neurodevelopmental processes (*e.g.* cell division, migration, differentiation, synaptogenesis, and myleination) and each of these is region and stage specific. Consequently, the time of toxicant

exposure will be a major determinate in the spectrum of neurotoxic effects. In addition, young children may be more susceptible with respect to exposure, as they age because they engage in more active behaviors for more sustained periods that can lead to higher overall exposures. These types of considerations are accounted for in the processes used by the Agency to evaluate the residential exposures of pesticides.

2.2.3. Critical Duration(s) of Exposure

It is important to appropriately match the exposure potential with the toxicology profile in risk assessment. As such, it is important to consider a variety of toxicokinetic and toxicodynamic factors such as the half life of the chemical in the body and the duration of the toxic effect (*i.e.*, the half life of recovery). AChE inhibition can occur from a single exposure or from repeated exposures. Chlorpyrifos exposure can occur from a single exposure (*e.g.*, eating a meal) or from repeated days of exposure (*e.g.*, worker). With respect to OPs, repeated exposures generally cause more inhibition at a given administered dose compared to acute studies. Moreover, AChE inhibition in repeated dosing guideline toxicology studies with OPs show a consistent pattern of steady state inhibition at or around 2-3 weeks of exposure in adult laboratory animals (U.S. Environmental Protection Agency, 2002). Specifically, with repeated dosing at a given dose, the amount of inhibition comes at equilibrium with production of new enzyme. As such, AChE studies of 2-3 weeks generally show the same degree of inhibition with those of longer duration (*i.e.*, up to 2 years of exposure). Thus, for the health outcomes relevant to the AChE inhibition mode of action in adults, the critical durations range from a single day up to 2-3 weeks (*i.e.*, the time to reach steady state). In newborns and young children, there are uncertainties associated as the concept of steady state may not hold as the metabolic capacity of children is changing over time (*i.e.*, increasing towards adult levels with age).

Very little is known about the duration of chlorpyrifos exposure needed to precipitate adverse effects in the developing brain. Because of the dynamic processes in the developing brain (*i.e.*, vulnerable windows) it is difficult to determine if the effect or differences in effects is due to duration of exposure or if different vulnerable windows were affected. As such, it is impossible at this time to rule out even a single day of exposure having a potential effect in humans.

3.0 Adverse Outcome Pathways: AChE Inhibition & Plausible Pathways Leading to Neurodevelopmental Outcomes

Mode of action and adverse outcome pathways provide important concepts in the draft "Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment" (U.S. Environmental Protection Agency, 2010). As mentioned above, both a mode of action and an adverse outcome pathway (AOP) include a set of measureable key events that describe the biological processes leading to an apical effect. However, an adverse outcome pathway explicitly considers the steps linking a molecular initiating event and an adverse outcome at a biological level of organization that is relevant for risk assessment. Figure 3 is a graphical presentation of a generic adverse outcome pathway (Ankley, et al., 2010). This figure is an extension of the source-to-outcome pathway (Figure 1) and provides additional detail on the types of scientific information from various levels of biological organization used in establishing an adverse outcome pathway. As shown in the conceptual framework (Figure 2), two different adverse outcomes in laboratory animals are being evaluated: AChE/ChE inhibition and neurodevelopmental outcomes. Each of these is discussed below.

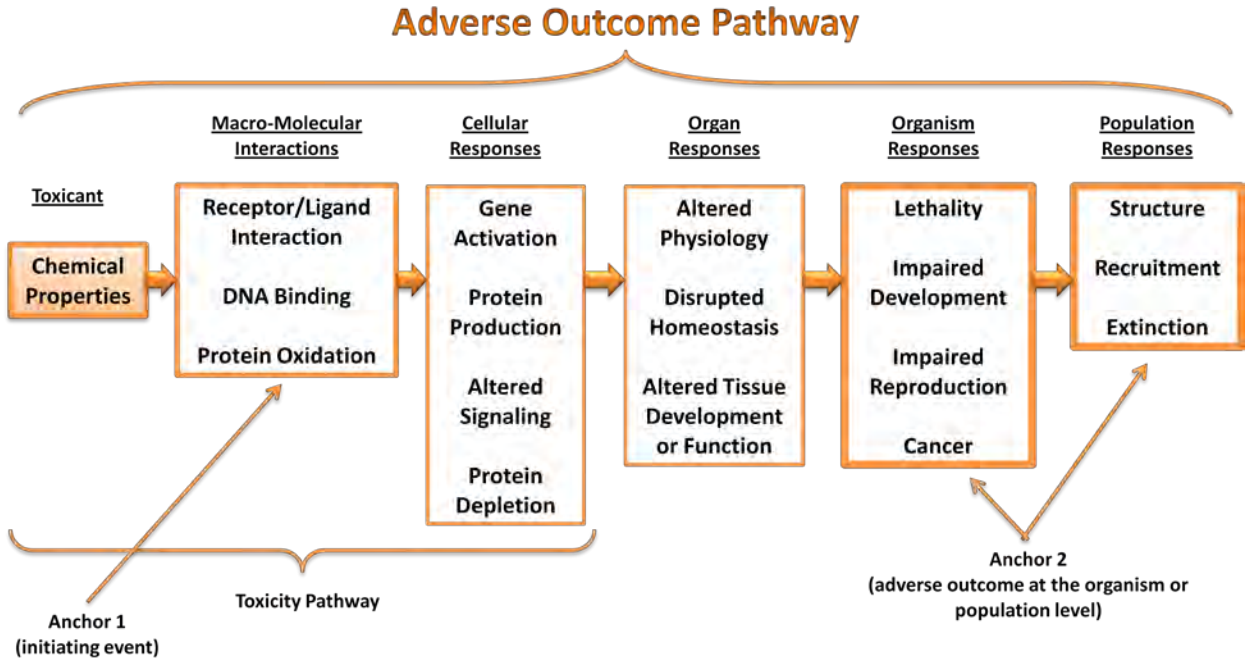


Figure 3 Generic Adverse Outcome Pathway (Adapted from Ankley et al., 2010)

3.1 Adverse Outcome Pathway: AChE Inhibition

3.1.1 Initiating Event & Health Outcomes

AChE inhibition is the well-established mode of action for OPs and is typically used as the critical effect in hazard characterization for members of this class of pesticides. Specifically, the initiating event for OP mediated neurotoxicity is interaction at the serine residue at the active site of the AChE. This interaction leads to accumulation of acetylcholine and ultimately to clinical signs of neurotoxicity at high doses. In experimental toxicity studies in laboratory animals, high acute doses produce signs of

autonomic stimulation, ataxia, fasciculations, tremors, respiratory difficulties, and convulsions. The Agency's recent review of human incident reporting databases (Recore & Oo, 2011) indicates that humans exposed to high levels of chlorpyrifos may complain of gastrointestinal and neurological effects consistent with acute AChE poisoning. Specifically, individuals have reported: nausea, vomiting, diarrhea, tremors, headaches, dizziness, numbness and tingling sensations, muscle spasm, shortness of breath, and seizures (Recore & Oo, 2011).

Although inhibition of AChE as the initiating event in this adverse outcome pathway has long been established, the quantitative dose response linkage between AChE inhibition and toxicity has been shown to be more variable. In other words, the amount of AChE inhibition required to elicit such responses from OP exposure is not consistent but instead varies among different OPs, different study designs, and different neurotoxic outcomes. Specific to chlorpyrifos, there are limited human data which inform the quantitative dose-response linkage between AChE inhibition and apical neurotoxic effects. There is one human deliberate dosing study in human subjects (Nolan, Rick, Feshour, & Saunders, 1982)[14] which provides plasma ChE and RBC AChE data from a single oral dose (0.5 mg/kg). AChE/ChE activity (plasma and RBC) was monitored at 2, 6, 12, 24 hours, and 2, 3, 4, 8, 14, 22, 27, and 30 days post dose. Despite plasma ChE inhibition of 83-89% at the peak time of effect, no clinical signs of toxicity or inhibition of RBC ChE activity were observed. In addition to this deliberate dosing study, several observational worker studies have monitored urinary TCPy and ChE activity. The Agency has previously reviewed these monitoring studies in its 2008 SAP meeting on chlorpyrifos and concluded that urinary TCPy was not predictive of AChE inhibition. Further description of the Agency evaluation of the relationship between TCPy and AChE inhibition is described in Section 5.4 of this report.

3.1.2 Dose Response Analysis for AChE Inhibition

Points of departures can be no-observed-adverse-effect-levels (NOAELs), low-observed-adverse-effect-levels (LOAELs), or derived from benchmark (BMD) modeling. Numerous scientific peer review panels over the last decade have supported the Agency's application of the BMD approach as an improvement over the historically applied approach of using NOAELs or LOAELs and as a scientifically supportable method for deriving points of departure in human health risk assessment. The NOAEL/LOAEL approach does not account for the variability and uncertainty in the experimental results, which are due to characteristics of the study design, such as dose selection, dose spacing, and sample size (U.S. Environmental Protection Agency, 2000). With the BMD approach, all the dose response data are used to derive a point of departure.

For the chlorpyrifos risk assessment, the Agency has used a decreasing exponential dose-response model similar to that used for the OP and *N*-methyl carbamate cumulative risk assessments and multiple risk assessments for individual AChE inhibiting pesticides (U.S. Environmental Protection Agency, 2006, 2007). The use of this empirical dose-response procedure has been previously reviewed and supported by the FIFRA SAP on several occasions (FIFRA Scientific Advisory Panel (SAP), 2001, 2002, 2005a, 2005b, 2008b). In lieu of a defined response level (*i.e.*, percent inhibition) that quantitatively links AChE inhibition to neurotoxic effects, the Agency typically uses a 10% decrease in brain and peripheral ChE inhibition as the response level in deriving points of departure. This 10% response level is called the benchmark response level (BMR). This level has been shown to be a level which is protective for neurotoxic outcomes for OPs and can also be reliably measured in most guideline experimental

[14] Nolan (1982) has been reviewed by the Human Studies Review Board (HSRB) and found to be ethically and scientifically conducted (HSRB, 2009).

toxicology studies (U.S. Environmental Protection Agency, 2002, 2006). The Agency has calculated BMD_{10}s and $BMDL_{10}$s where the BMD_{10} is the estimated dose where AChE is inhibited by 10% compared to background and the $BMDL_{10}$ is the lower confidence bound on the BMD_{10}. As a matter of science policy, the Agency uses the BMDL, not the BMD, for use as the point of departure since the BMDL accounts for variability of the data (U.S. Environmental Protection Agency, 2000).

As part of the 2008 SAP, the Agency performed a comprehensive review of the literature on available AChE data across multiple lifestages, durations, and routes (FIFRA Scientific Advisory Panel (SAP), 2008a). This literature review was considered by the SAP at that time and provided part of the scientific rationale for the Panel's recommendation that the Agency continue to use AChE data in the most sensitive lifestages for dose-response analysis and deriving points of departure. This extensive literature is not summarized here but can be found in Draft Appendix B of the 2008 draft science issue paper presented to the SAP (FIFRA Scientific Advisory Panel (SAP), 2008a). Instead, the following text focuses on the most robust dose-response studies used in the deriving points of departure for the 2011 risk assessment.

For endpoint selection, the Agency considered the quality of all the available studies, both previously available at the time of the 2008 SAP and newest ones. Route-specific dermal and inhalation studies are available for chlorpyrifos and are being used to assess dermal and inhalation exposure, respectively. Information on these studies can be found in the 2011 risk assessment (U.S. Environmental Protection Agency, 2011). There are also high quality dose-response data in adults and juvenile rats for chlorpyrifos oxon. Dose response analysis for the oxon data has been conducted and can also be found in the 2011 risk assessment.

For the preliminary risk assessment, the Agency has conducted BMD modeling on selected oral AChE studies. These studies were selected based on the availability of at least two treatment groups in addition to a control group and those that showed good dose response. The Agency has also focused on those studies where AChE was measured at or near the time of peak effect, typically within one to several hours after dosing. AChE data measured 24 hours or longer after dosing will underestimate the amount of AChE inhibition and are thus not appropriate for deriving points of departure. In addition, the studies used in the dose response assessment were selected as they represented a variety of ages, lifestages, and durations. The Agency has focused on those studies representing single and repeating dosing in post-natal exposure to rat ages (PND 10 and older); PND10 and older is considered to be approximately concordant with human postnatal exposure (Benjamins & McKhann, 1981; Dobbing & Smart, 1974). AChE data from adult laboratory animals (both pregnant and non-pregnant) from single dose and repeating dosing studies has also been collected. The focus has been on AChE data from rats as these are generally the most robust and sensitive studies. Data in fetuses have not been evaluating using BMD techniques as these studies tend to be less sensitive than those in post-natal juveniles and adults. The Agency has conducted BMD analysis on available data for blood (RBC), brain (whole or sections as appropriate), and peripheral (*i.e.*, heart) tissues.

The Agency typically performs risk assessments for oral exposure to food and water, in addition to non-dietary ingestion to young children. For assessment of food, water, and aggregate dietary risk, the Agency typically conducts single day (*i.e.*, acute) and chronic assessments. For assessment of non-dietary ingestion to young children, the Agency conducts short- and intermediate-term assessments which involve durations of 1-30 days and 1 month to 6 months, respectively.

There are several studies available which inform the acute oral point of departure. For the assessment of infants and children, the most robust studies are 1) the new comparative cholinesterase[15] (CCA) study (MRID 48139301) which provides brain and RBC data in PND11 pups and 2) (Moser, Simmons, & Gennings, 2006) which provides data from brain and whole blood ChE data in PND17 pups. In both of these studies, the rat pups were dosed directly and ChE was measured at the time of peak effect. In addition, two studies (Timchalk, Poet, & Kousba, 2006; Q. Zheng, Olivier, Won, & Pope, 2000) provide supporting data. These two studies show AChE data in post-natal rats which are consistent with the comparative cholinesterase study and Moser et al (2006). However, Timchalk et al. (2006) and Zheng et al. (2000) are not robust datasets for dose-response modeling (U.S. Environmental Protection Agency, 2011).

For the assessment of adults, single dosing AChE data from the comparative cholinesterase study (MRID 48139301) provide the most robust data for BMD analysis[16]. Tables 1 and 2 provide a summary of the BMD results for the single dosing studies in post-natal rat pups and adult rats, respectively. Generally for the both the pups and the adults, the BMD and BMDL estimates for the blood measures are lower than those for the brain. In addition, the results from the pups are approximately 2-fold lower than those for adults (*i.e.*, when comparing the same compartment, administration vehicle). The $BMDL_{10}$ for RBC AChE inhibition of 0.36 mg/kg from the PND11 pups exposed to chlorpyrifos in milk provides the most sensitive data for deriving a point of departure. These results are supported by the Moser et al (2006)$BMDL_{10}$ of 0.43 mg/kg from whole blood ChE, from the RBC AChE measures from the gavage administered PND11 pups in the comparative cholinesterase study ($BMDL_{10}$s of 0.65 and 0.75 mg/kg), and the adult female results from exposure in the diet for 12 hours (0.60 mg/kg).

[15] Comparative cholinesterase (CCA) studies are specially designed studies in juvenile and adult rats which provide direct comparative ChE/AChE data for evaluating lifestage sensitivity. For OPs, data are typically collected from acute dosing (typically PND11, young adult) and from 11-days repeated dosing (PND11-21 and young adults).

[16] Only adult female rats were tested in MRID 48139301.

Table 1. Results of BMD Modeling of Rat Pup Brain and Blood ChE Inhibition following a Single Oral Dose of Chlorpyrifos

Reference	Sex & Age at Dosing	Endpoint	BMD_{10} (mg/kg)	$BMDL_{10}$ (mg/kg)
Moser et al., 2006	Male PND 17 Gavage	Brain AChE	1.89	1.54
Moser et al., 2006	Male PND 17 Gavage	Whole Blood AChE	0.62	0.43
CCA Study (MRID 48139301)	Male PND 11 Gavage	Brain AChE	2.13	1.53
CCA Study (MRID 48139301)	Male PND 11 Gavage	RBC AChE	0.82	0.65
CCA Study (MRID 48139301)	Female PND 11 Gavage	Brain AChE	2.18	1.56
CCA Study (MRID 48139301)	Female PND 11 Gavage	RBC AChE	0.96	0.75
CCA Study (MRID 48139301)	Male PND 11 Milk	Brain AChE	4.4	2.4
CCA Study (MRID 48139301)	Male PND 11 Milk	RBC AChE	0.47	0.36
CCA Study (MRID 48139301)	Female PND 11 Milk	Brain AChE	1.42	0.91
CCA Study (MRID 48139301)	Female PND 11 Milk	RBC AChE	0.5	0.36

Table 2. Results of BMD Modeling of Adult Rat Brain and RBC AChE Inhibition following a Single Oral, Dose of Chlorpyrifos

Reference	Sex & Administration Method	Endpoint	BMD_{10} (mg/kg)	$BMDL_{10}$ (mg/kg)
CCA (MRID 48139301)	Female Gavage	Brain AChE	4.11	2.26
CCA (MRID 48139301)	Female Gavage	RBC AChE	1.9	1.2
CCA (MRID 48139301)	Female 12 hr diet	Brain AChE	4.47	3.30
CCA (MRID 48139301)	Female 12 hr diet	RBC AChE	1.03	0.6

Several high quality repeated dosing oral studies are available determination of oral points of departure for chronic dietary scenarios (Tables 3 and 4). For BMD analysis in the preliminary risk assessment, the Agency selected the repeated dosing portions of the comparative cholinesterase study (MRID 48139301) in juvenile and non-pregnant adult rats;, the data from pregnant dams in the developmental neurotoxicity rat study (Maurissen, Hoberman, Garman, & Hanley, 2000) (MRID 44556901) and the data

from pregnant dams in the special cholinesterase study in which dams where administered chlorpyrifos from gestation day 6 through lactation day 10 (GD6 – LD10) (Mattsson, Maurissen, Nolan, & Brzak, 2000); MRID 44648101). Although the Zheng et al (2000) data are not robust data for BMD modeling, the repeated dosing portion of this study provides additional supportive data for the findings from the comparative cholinesterase study in both pups and adults.

Table 3. Results of BMD Modeling of Pup Rat Brain and RBC AChE Inhibition following 11-days Repeated Oral, Gavage Doses of Chlorpyrifos

Reference	Sex & Age/Duration of Dosing	Endpoint	BMD_{10} (mg/kg/day)	$BMDL_{10}$ (mg/kg/day)
CCA (MRID 48139301)	Female PND 11-21 Gavage	Brain AChE	0.80	0.69
CCA (MRID 48139301)	Female PND 11-21 Gavage	RBC AChE	0.17	0.15
CCA (MRID 48139301)	Male PND 11-21 Gavage	Brain AChE	0.63	0.52
CCA (MRID 48139301)	Male PND 11-21 Gavage	RBC AChE	0.11	0.09

Table 4. Results of BMD Modeling of Adult, Female Rat Brain, RBC and Heart Cholinesterase Inhibition following Repeat Oral Doses of Chlorpyrifos

Reference	Sex, Age/Duration & Administration method	Endpoint	BMD_{10} (mg/kg/day)	$BMDL_{10}$ (mg/kg/day)
Dow (MRID 44556901; Maurissen et al., 2000)	Dams, GD6-20, Gavage	Brain AChE	0.65	0.54
Dow (MRID 44556901; Maurissen et al., 2000)	Dams, GD6-20, Gavage	RBC AChE	0.06	0.03
Dow (MRID 44648101; Mattsson et al., 2000)	Dams, GD6-20, Gavage	Brain AChE	1.1	0.8
Dow (MRID 44648101; Mattsson et al., 2000)	Dams, GD6-20, Gavage	RBC AChE	0.14	0.08
Dow (MRID 44648101; Mattsson et al., 2000)	Dams, GD6-20, Gavage	Heart ChE	0.85	0.22
Dow (MRID 44648101; Mattsson et al., 2000)	Dams, GD6-LD10 Gavage	Brain AChE	1.13	0.89
Dow (MRID 44648101; Mattsson et al., 2000)	Dams, GD6-LD10 Gavage	RBC AChE	0.050	0.044
Dow (MRID 44648101; Mattsson et al., 2000)	Dams, GD6-LD10 Gavage	Heart ChE	0.21	0.18
CCA (MRID 48139301)	Adult, non-pregnant 11 daysGavage	Brain AChE	1.03	0.95
CCA (MRID 48139301)	Adult, non-pregnant 11 days Gavage	RBC AChE	0.45	0.35

As shown in Tables 3 and 4, there is no meaningful difference between BMD_{10}s and $BMDL_{10}$s from pups and adults in repeated dosing studies. This is not unexpected given that over the duration of the repeated dosing study, the metabolic system in pups is maturing closer to adult levels. Thus, it is not unexpected that as the metabolic system matures that the degree of sensitivity decreases. Typically, studies submitted for pesticide registration and most studies from the public literature only measure brain and/or blood ChEs. It is rare for data from peripheral tissues to be available for consideration. Chlorpyrifos is unique in that multiple studies are available which provide such peripheral data. It is notable that the BMDs and BMDLs for heart ChE are more sensitive than brain and closer in magnitude than those for RBC AChE. This finding supports the use of the blood measures.

There is good concordance in the estimates across the BMD_{10} and $BMDL_{10}$ estimates across the adult studies evaluated in this analysis. Notably, brain AChE estimates range from approximately 0.5 to 1 mg/kg/day and the RBC AChE estimates range from 0.03 to 0.35 mg/kg/day. In addition, for the OP cumulative risk assessment (U.S. Environmental Protection Agency, 2006), the Agency performed BMD modeling using a sophisticated meta-analysis approach on brain AChE data from adult, non-pregnant female and male rats from one subchronic oral toxicity study and two chronic oral toxicity studies (MRID nos. 40952801, 40952802, and 42172802). The BMD results reported in the OP cumulative risk assessment for chlorpyrifos are BMD_{10}s of 1.48 and 1.50 mg/kg/day and $BMDL_{10}$s of1.26 and 1.27, mg/kg/day respectively in females and males. The values for brain AChE inhibition reported in the cumulative assessment from subchronic and chronic dosing are quite similar to those reported in Table 4 above for both pregnant and non-pregnant females and males in the selected studies and add further support to the analysis conducted for the preliminary risk assessment for chlorpyrifos. For purposes of deriving a point of departure for risk assessment, the Agency is using the $BMDL_{10}$ of 0.03 mg/kg/day for RBC AChE inhibition from the DNT study (MRID 44556901, also called (Maurissen, et al., 2000).

3.1.3 Summary of the Dose Response Analysis

Acute oral point of departure is $BMDL_{10}$ = 0.36 mg/kg based on inhibition of RBC AChE in male and female rat pups from the CCA study and is supported by RBC BMDs from data on PND17 males (Moser et al.,(2006), and adult female in the comparative cholinesterase study exposed in the diet. These studies are further supported by Timchalk et al. (2006) and Zheng et al. (2000) studies. The chronic oral point of departure is $BMDL_{10}$ = 0.03 mg/kg/day based on inhibition of RBC AChE in rat dams (GD6-20) from the DNT study (MRID 44556901, also called Maurisse et al., 2000) with support from pregnant (GD6-LD10) rats (MRID 44648101).

3.2 Adverse Outcome Pathway: Neurodevelopmental Outcomes

With respect to modes of action/adverse outcome pathways leading to neurodevelopmental effects, at the present time, there is no established series of causal key events at a biological level of organization relevant to the risk assessment (*i.e.*, adverse neurodevelopmental effects from gestational and/or postnatal exposure). The Agency does not believe that any of the current lines of research support a coherent set of key events and that much work remains to elucidate the MOAs and AOP of chlorpyrifos toxicity. Even though a rigorous demonstration of an adverse outcome pathway for chlorpyrifos has not been constructed at this time, there are multiple studies from numerous laboratories that show that exposure to a variety of OPs (including chlorpyrifos, other pesticides, nerve gases) during the developmental period affects key events in nervous system development (see below). These studies

conducted to date generally report a correlation between OP exposure and a tested effect without consideration of temporal concordance (*i.e.*, the sequence of biological events leading to effects) and quantitative linkages (*i.e.*, degree of change that results in neurodevelopmental consequences). Some of this experimental work specific to chlorpyrifos is summarized below, and may be shown in the future actually to be steps in the same AOP and not discreet pathways. However, at this time, the experimental studies below do not provide all the information needed to demonstrate linkages among initiating event(s), subsequent events at the molecular and cellular level, and the adverse outcome of interest and are thus described separately.

3.2.1 Biologically Plausible Toxicity Pathways Leading to Neurodevelopmental Outcomes

3.2.1.1 Acetylcholinesterase (AChE) as a morphogen

The classically understood role of AChE is the rapid hydrolysis of acetylcholine at synapses in the brain and at neuromuscular junctions, thereby regulating cholinergic neurotransmission. Consistent with this role, AChE is predominant at cholinergic synapses at neurons and in muscle, and inhibition of its catalytic activity results in the signs and symptoms of cholinergic overstimulation. Several lines of evidence, however, suggest that AChE can also serve as a morphogen, influencing the growth of cells during neurodevelopment. Alterations in the expression or structure of the AChE protein can disrupt various aspects of neuronal differentiation and growth. Because chlorpyrifos can interact with AChE, perturbation of this morphogenic activity represents a plausible adverse outcome pathway leading to developmental neurotoxicity.

One of the first indications that AChE had roles other than the termination of cholinergic neurotransmission was the timing and distribution of its expression in the developing nervous system. In certain brain regions containing non-cholinergic neurons and few cholinergic synapses, AChE levels are still high. In addition, AChE is highly expressed throughout the brain during periods of active axonal outgrowth in the absence of other cholinergic markers and before synaptic connections are made (Bigbee, Sharma, Gupta, & Dupree, 1999; Brimijoin & Koenigsberger, 1999). For example, AChE levels are high during early cerebellar development while there is very little evidence for the presence of cholinergic cells or cholinergic neurotransmission (Appleyard & Jahnsen, 1992; Parvari, 1983). A second line of evidence for a morphogenic role arose after the cloning and sequencing of rodent and human AChE. Surprising sequence homologies were found with a family of proteins lacking a catalytically active esterase site but pocessing a similar extacellular domain. Several of the homologs of AChE are extracellular matrix components essential to neuronal adhesion, axon guidance, and synapse formation (Brimijoin & Koenigsberger, 1999; Grisaru, Sternfeld, Eldor, Glick, & Soreq, 1999). Thus, the sequence of AChE is similar to other proteins which have a morphogenic role during the development of the nervous system. Finally, experimental evidence indicates that manipulation of AChE levels or activity can influence neuronal growth *in vitro*. Studies in cultured neuroblastoma cells (Koenigsberger, Chiappa, & Brimijoin, 1997) or PC12 cells (Grifman, Galyam, Seidman, & Soreq, 1998) have shown that antisense suppression of AChE decreased neurite outgrowth, and that neurite outgrowth could be rescued or increased by transfection with sense AChE. In cultures of rat dorsal root ganglion cultures, treatment with an antibody against AChE (which did not inhibit the catalytic activity) decreased neurite outgrowth, and neurite outgrowth was restored upon removal of the antibody (Bigbee, et al., 1999). Expression in frog embyros of a recombinant human AChE lacking the ability to hydrolyze acetylcholine resulted in increased neurite outgrowth from cultured spinal neurons (Sternfeld et al., 1998). Thus, the catalytic ability of AChE does not seem to be required for its morphogenic properties.

Studies using pharmacologic inhibition of AChE activity to alter neurite outgrowth also suggest a dissociation of the catalytic and morphogenic activities. Several laboratories using different *in vitro* systems have demonstrated that some potent AChE inhibitors including BW284c51 supress neurite outgrowth while other, equally potent AChE inhibitors like ecothiophate do not (Bigbee, et al., 1999; Koenigsberger, et al., 1997; Layer, Weikert, & Alber, 1993). These differential effects may be related to the degree of chemical binding to separate sites on the AChE enzyme. There are at least two sites that can bind inhibitors: the catalytic (active) site and the peripheral anionic site (Grisaru, et al., 1999). It has been suggested that inhibitors that bind directly to the peripheral anionic site, or that bind to the catalytic site in such a way that changes the conformation of the peripheral anionic site, will alter the morphogenic function of AChE (Bigbee, et al., 1999; Yang et al., 2008). Thus inhibitors that bind only to the catalytic site (*e.g.* edrophonium, tacrine) did not affect neurite outgrowth, while ligands that bind to the peripheral anionic site (propidium, gallamine) or both sites (BW284c51) inhibited neurite outgrowth (Koenigsberger, et al., 1997; Munoz, Aldunate, & Inestrosa, 1999). At least one study indicates that chlorpyrifos oxon can bind at both sites (Kousba, Sultatos, Poet, & Timchalk, 2004).

Data showing a potential action of chlorpyrifos on the morphogenic function of AChE are derived primarily from *in vitro* studies. Chlorpyrifos (as well as the activated oxon form and the inactive metabolite TCPy) has been examined for effects on neurite outgrowth using a number of cell lines and in primary neuronal cultures. In some cases concurrent assessments of cholinesterase inhibition or cytotoxicity were performed. Li and Cassida (W. Li & Casida, 1998) examined the effect of chlorpyrifos oxon on neurite outgrowth in the PC12 cell line. In this study cells were exposed prior to, but not during the active phase of neurite growth, which was assessed after 5 days. Chlorpyrifos oxon inhibited neurite outgrowth by 50%, but only at a relatively high concentration (200 uM) that was both cytotoxic and decreased cholinesterase activity. Sachana et al. (Sachana, Flaskos, & Hargreaves, 2005; Sachana, Flaskos, Sidiropoulou, Yavari, & Hargreaves, 2008) examined the effect of 3 uM chlorpyrifos on the initiation of neurite growth in mouse N2a neuroblastoma cells. The number of cells exhibiting neurites after 4 or 8 hours of exposure (considered as a measure of cell differentiation) was decreased by approximately 50% in the absense of cytotoxicity. Cholinesterase activity was not measured. Axelrad et al. (2002) exposed mouse NB2a cells to chlorpyrifos for 24 hr and directly measured neurite length. Chlorpyrifos inhibited neurite outgrowth by 50% at 25 uM. Again, cholinesterase activity was not measured. While the studies cited above suggested that chlorpyrifos and chlorpyrifos oxon could affect measures related to neuronal differentiation and neurite growth, they were not specifically designed to examine the morphogenic role of AChE.

Using PC12 cells, Das and Barone (1999) examined the concentration-related effects of chlorpyrifos, chlorpyrifos oxon, and TCPy on both neurite outgrowth and cholinesterase inhibition. Exposure to chlorpyrifos for 24 hr inhibited neurite outgrowth at a concentration (3 uM) 10-fold below that which inhibited cholinesterase activity, while chlorpyrifos oxon inhibited both measures at equivalent concentrations (1 nM). TCPy, which is inactive against AChE, inhibited neurite growth at 5 uM. Similar studies of chlorpyrifos and its metabolites were performed in a series of experiments in the Lein laboratory using primary neuronal cultures derived directly from the mammalian nervous system. Using rat sympathetic neurons, Howard et al. (2005) showed that 24 hr exposure to chlorpyrifos and chlorpyrifos oxon decreased axonal outgrowth at concentrations (0.001 uM and 0.001 nM, respectively) well below the concentrations that inhibited AChE activity (1 uM and 1 nM). In the same study chlorpyrifos, chlorpyrifos oxon and TCPy enhanced dendrite outgrowth. A follow-up study from the same laboratory used sensory neurons from the dorsal root ganglion, which extend only axons (Yang, et al., 2008). Similar results were observed, with both chlorpyrifos and chlorpyrifos oxon decreasing axonal

outgrowth at concentrations below those that inhibited AChE activity. To establish whether the target of chlorpyrifos and chlorpyrifos oxon was AChE or some other molecule, Yang et al. (Yang, et al., 2008) repeated the experiments in cultures from AChE-null animals. Chlorpyrifos and chlorpyrifos oxon had no effect in the AChE-null cultures, suggesting that inhibition of neurite outgrowth required AChE. These later three studies provide the most convincing evidence for selective inhibition of the morphogenic activity of AChE. It is not yet clear whether the effects are a direct action of chlorpyrifos on AChE, or the result of *in vitro* conversion to the active oxon form. It should be mentioned that studies from the Slotkin laboratory suggest an effect of chlorpyrifos on neurite outgrowth *in vitro* (Song, Violin, Seidler, & Slotkin, 1998). In those experiments, however, neurite outgrowth is inferred from biochemical measurements and cholinesterase activity is not assessed concurrently.

Testing the hypothesis that exposure to chlorpyrifos (or any chemical) can inhibit the morphogenic function of AChE *in vivo* and alter brain development is difficult. Neuronal differentiation and the subsequent development of axonal and dendritic networks occurs in a temporal- and region-specific manner. In the absence of prior information extensive studies would be required to survey brain morphology during both the prenatal and postnatal period. In addition, the methods needed to assess the morphology of axonal and dendritic growth *in vivo* and detect potentially subtle chemical-induced changes are tedious and time consuming. Nevertheless, some attempts have been made to detect chlorpyrifos-induced changes in neurite growth *in vivo*. In particular, several laboratories have examined the effect of chlorpyrifos and chlorpyrifos oxon on neuronal morphogenesis in developing zebrafish. Jacobson et al. (2010) exposed zebrafish embryos at 3 hr post fertilization to 300 nM chlorpyrifos oxon. When examined 24 hr later (1 day post fertilization), AChE was inhibited by 50%. Gross morphology was only slightly affected and muscular development (including the neuromuscular junction) was normal; however, there was a decrease in the number of Rohon-Beard sensory neurons accompanied by abnormal extension of their axons. As a follow up to this study and to their *in vitro* work described above, Yang et al. (2011) exposed zebrafish embryos from 24 to 72 hr post fertilization to chlorpyrifos and chlorpyrifos oxon. Chlorpyrifos had no effect, but chlorpyrifos oxon significantly decreased AChE activity (30 nM), decreased touch-induced swimming (100 nM) and inhibited axon growth from both sensory neurons and motor neurons (100 – 1000 nM). These two studies show that exposure to chlorpyrifos oxon in an intact, developing organism can alter neurite outgrowth. However, because catalytic activity was also inhibited, it is not clear that this effect was solely due to disruption of the morphogenic role of AChE.

As noted above, neurite outgrowth has been assessed indirectly *in vitro*. This method has also been applied *in vivo* in mammals. Qiao et al. (2003) exposed rats to 1 or 5 mg/kg chlorpyrifos on gestational days 17-20. In a previous study from the same laboratory this exposure paradigm resulted in a non-significant 5% decrease in AChE activity after 1 mg/kg and a significant 50% decrease in AChE activity after 5 mg/kg when assessed 24 hr after the last dose (Qiao, Seidler, Padilla, & Slotkin, 2002). The ratio of membrane protein to total protein in several brain regions was assessed as a surrogate index of neurite outgrowth. The results indicated that there was no effect of treatment during early postnatal development (PND 4-21), but that both doses decreased this ratio on PND 30 and 60. While the authors interpret this biochemical measurement as a decrease in neurite growth, there has not been data presented to correlate the changes in the membrane protein to total protein ratio with actual quantification of neurite length. In addition, no effects were observed early in development when axons and dendrite growth is high.

In summary, while perturbation of the morphogenic activity of AChE is a plausible adverse outcome pathway for chlorpyrifos, a number of questions remain. There is substantial evidence for a

morphogenic role of AChE in nervous system development distinct from its role as an esterase to hydrolyze acetylcholine. *In vitro* evidence suggests that AChE can regulate neurite outgrowth, and that cholinesterase inhibitors (including chlorpyrifos and its metabolites) can interfere with this process at concentrations that do not inhibit the esterase activity. There is, however, no direct evidence showing that disruption of the morphogenic function of AChE can alter axon or dendritic growth *in vivo*. While limited *in vivo* studies using zebrafish indicate that chlorpyrifos or its metabolite chlorpyrifos oxon can disrupt axonal growth, it has not been demonstrated that this effect is due to alteration of the morphogenic function of AChE versus other potential mechanisms.

3.2.1.2 Cholinergic system

There are several lines of evidence showing that signaling through cholinergic receptors is involved in neurodevelopment. Activation of muscarinic and/or nicotinic cholinergic receptors can regulate neural progenitor cell proliferation and differentiation (Resende & Adhikari, 2009), and *in vivo* studies demonstrate that cholinergic signaling is likely involved in brain morphogenesis (Hohmann & Berger-Sweeney, 1998). While cholinesterase inhibitors can affect cholinergic signaling by inhibition of the catalytic activity of AChE and subsequent increase in acetylcholine, some, including chlorpyrifos and chlorpyrifos oxon, can also directly interact with cholinergic receptors. Thus, direct interaction with cholinergic receptors by chlorpyrifos represents a potential adverse outcome pathway for disruption of neurodevelopment distinct from AChE/ChE inhibition.

Some OPs have been shown to directly interact with cholinergic muscarinic receptors at relatively low concentrations. The muscarinic receptors are members of the G-protein receptor family and five subtypes (m1-m5) have been identified. Volpe et al. (1985) reported that nanomolar concentrations of several OPs decreased binding of quinuclidinyl benzilate (QNB), which binds equally to all five subtypes. This effect was noncompetitive and occurred at only a small fraction of the total QNB binding sites. A further study from the same laboratory proposed these receptor subtypes to be m2 and/or m3 (L. S. Katz & Marquis, 1989). Bakry et al. (1988) found that a number of OP nerve agents and an OP therapeutic agent inhibited QNB binding to rat brain membranes with low potency, but competitively inhibited binding of cis-methyldioxolane (CD) with high potency. In rat cardiac tissue, OPs also competitively inhibited CD binding with very high potency (Silveira, Eldefrawi, & Eldefrawi, 1990). CD is a muscarinic agonist that binds to the high affinity state of the m2 receptor in mammalian brain and heart (Closse, Bittiger, Langenegger, & Wanner, 1987; Vickroy, Roeske, & Yamamura, 1984; Watson, 1986). Subsequent research using Chinese hamster ovary (CHO) cells transfected with cDNA for the five distinct genes for muscarinic receptors (ml-m5) has identified the CD binding site as the high affinity state (GDP bound G-protein) of the m2 receptor (Huff & Abou-Donia, 1994). In light of these findings, Ward et al. (1993) examined the relationship between cholinesterase inhibition and direct binding to muscarinic receptors for a series of OPs and their active “oxon” metabolites. The results indicated a strong correlation between anticholinesterase activity of OPs, including chlorpyrifos and chlorpyrifos oxon, and the ability to compete for CD binding sites (m2 receptors) in rat brain homogenates. Binding affinities of the oxons were in the nanomolar range, at or below concentrations that inhibited AChE (Huff, Corcoran, Anderson, & Abou-Donia, 1994); specifically, chlorpyrifos oxon had a binding affinity of 22 nM in rat striatum and 2 nM in rat cortex (Huff, et al., 1994; Ward & Mundy, 1996). In total, these studies suggest that direct interactions with muscarinic receptors, and especially the m2 subtype, represent an alternative site of action for OPs including chlorpyrifos and chlorpyrifos oxon, with the oxon forms having high affinity.

Further studies determined whether binding of OPs to the m2 receptor had functional consequences on downstream second messenger signaling. The m2 and m4 receptors are coupled to an inhibitory G-protein (G_i) and activation inhibits adenylate cyclase and decreases cAMP formation. In contrast, m1, m3, and m5 receptors increase PI hydrolysis via the stimulatory G-protein G_p (Lameh et al., 1990). Jett et al. (1991) reported that paraoxon could inhibit both CD binding and cAMP formation in rat striatum. The effects of paraoxon were similar to the classical muscarinic agonist carbachol, and were blocked by the muscarinic antagonist atropine. Similar results were observed in rat frontal cortex for paraoxon and malaoxon (Ward & Mundy, 1996). In addition, Ward and Mundy (1996) observed that the OPs had no effect on PI hydrolysis. These data suggest that OPs can act as agonists at the m2 receptor (but not the m1, m3, or m5 receptor) and decrease cAMP formation. Huff et al. (1994) extended these findings to chlorpyrifos oxon, which had an IC_{50} for inhibition of cAMP of 155 nM. Unlike the other OPs, however, the effects of chlorpyrifos oxon were not completely blocked by atropine; an observation confirmed by Ward and Mundy (1996). Thus, like other OPs chlorpyrifos oxon can act directly as an agonist at the m2 receptor, but may also act at another site downstream of the receptor to inhibit adenylate cyclase.

Cholinergic receptor signaling has been shown to be involved in apoptosis, cell proliferation and neuronal differentiation (Resende & Adhikari, 2009). While the m2 receptor subtype is widely expressed in proliferating neuroepithelial cells in the ventricular zone of embryonic brain (Ma, Li, Zhang, & Pant, 2004), evidence for a specific role of in the modulation of neurodevelopment is derived primarily from *in vitro* studies. Neural precursor cells derived from embryonic rat cortex expressed the m2 receptor, and exposure to muscarinic agonists increased cell proliferation and enhanced differentiation into neurons (Ma et al., 2000). Using a P19 embryonic cell line as a model for neurogenesis, Resende et al. (2008) used pharmacologic agonists and antagonists to demonstrate the ability of m2 receptors to induce cell differentiation. The m2 receptor is highly expressed in developing cells of the dorsal root ganglia, and is thought to regulate both neuronal and non neuronal cell differentiation (Biagioni, Tata, De Jaco, & Augusti-Tocco, 2000; Tata, Cursi, Biagioni, & Augusti-Tocco, 2003). Another line of evidence for involvement of m2 receptor signaling in neurodevelopment is the role of cAMP in neurite outgrowth. Studies in both cell lines and primary neuronal cultures show that activation of adenylate cyclase and subsequent formation of cAMP stimulate neurite outgrowth (Kamei & Tsang, 2003; Mattson, Guthrie, & Kater, 1988), while inhibition of adenylate cyclase decreases neurite outgrowth (Tam, Rosenberg, & Maysinger, 2006; Wong, Bruch, & Farbman, 1991). Thus, *in vitro* evidence supports a role of m2 receptor signaling in neurodevelopment at the cellular level. However, evidence showing that manipulation of m2 receptors can alter brain development *in vivo* is lacking.

Because they can also influence neurodevelopment, it should be noted that there are several studies demonstrating that OPs including chlorpyrifos and chlorpyrifos oxon can bind directly to and desensitize nicotinic receptors (E. J. Katz, Cortes, Eldefrawi, & Eldefrawi, 1997; Smulders, Bueters, Vailati, van Kleef, & Vijverberg, 2004). This binding, however, occurs at relatively high concentrations (5 – 30 uM) and has not been demonstrated in neuronal tissue.

In summary, *in vitro* studies have shown that chlorpyrifos oxon can bind to and activate m2 receptors at levels similar to those that inhibit AChE activity. Other work has shown that m2 receptor signaling can regulate various aspects of neurodevelopment. Together, the studies cited above outline a plausible adverse outcome pathway for chlorpyrifos and chlorpyrifos oxon to affect brain development via actions at the m2 subtype of muscarinic receptors. However, while there are studies showing that chlorpyrifos oxon can affect neurite outgrowth *in vitro* and decrease cell proliferation and differentiation both *in vitro* (Jameson, Seidler, Qiao, & Slotkin, 2006; Qiao, Seidler, & Slotkin, 2001; Song, et al., 1998) and *in*

vivo (Dam, Seidler, & Slotkin, 1998; Qiao, et al., 2003), there is no experimental evidence that these effects are a result of direct actions on the m2 receptor.

3.2.1.3. Endocannabinoid system

Several lines of research have suggested that disruption of the endocannabinoid (EC) system due to chlorpyrifos exposure could play a role in its acute and/or long-term toxicity, and could also be extended to potential developmental toxicity. The EC system modulates neurotransmission as well as playing a morphogenic role during development of the nervous system. Chemicals *e.g.*, drugs of abuse, which act on this system, produce long-term neurodevelopmental disorders in animal models and human studies. Chlorpyrifos also interacts with this system, both *in vitro* and *in vivo*. By this reasoning, the EC system represents a possible adverse outcome pathway for developmental effects of chlorpyrifos.

The EC system includes inhibitory G-protein-coupled receptors (CB1 and CB2), endogenous ligands (2-arachidonoylglycerol, or 2-AG, and anadamide, or AEA), and hydrolases (monoacylglycerol, or MAG, and fatty acid amide hydrolase, or FAAH) that end the receptor actions of these ligands. CB1 receptors are predominant in certain brain regions, and the EC system modulates neuronal transmission and is involved in several physiological processes including appetite, pain sensation, mood, and cognition (Wilson & Nicoll, 2002). Emerging lines of research indicate that during development, this system plays a major role in controlling the morphological and functional specification of neurons (*e.g.*, Fride, 2008; Harkany et al., 2007; Harkany, Keimpema, Barabas, & Mulder, 2008; Harkany, Mackie, & Doherty, 2008); reviewed in Campolongo, Trezza, Palmery, Trabace, & Cuomo, 2009). Its involvement in neurogenesis, proliferation, and axon guidance suggests a role in overall neuronal connectivity.

There are epidemiological data on neurodevelopmental outcomes following prenatal exposure to cannabinoids such as cannabis and THC through maternal drug abuse, but interpretations are often somewhat difficult given co-exposures to other neurotoxicants (*e.g.*, ethanol). Despite limitations, an overall picture has emerged of long-lasting impaired cognitive processes, including attention and problem-solving deficits, as well as anxiety and depressive symptoms in humans (*e.g.*, Campolongo, et al., 2009; Fried & Smith, 2001; Jutras-Aswad, DiNieri, Harkany, & Hurd, 2009; Trezza, Cuomo, & Vanderschuren, 2008). In addition, animal models have indicated various changes in neurotransmission of catecholaminergic and indolaminergic systems, ontogeny of motor function, and cognitive function in adults developmentally exposed to cannabis as well as other CB1 agonists (*e.g.*, WIN55,212-2) (reviewed in Campolongo, Trezza, Ratano, Palmery, & Cuomo, 2011; Marco et al., 2009). This supports the possibility that other chemicals, unrelated to cannabis, that also act on the EC system early in development could lead to long-term neurological dysfunction. Despite the number of studies in this area, however, the specific responsible cellular responses have not been delineated, and most studies have not evaluated basic dose-response such as correlations of the degree of receptor binding with any structural or functional outcomes.

In addition to drugs of abuse (*e.g.*, cannabis), there are several classes of chemicals that act on the EC system, including organosulfonyl fluorides and some OPs (*e.g.*, Casida & Quistad, 2004; Segall, Quistad, Sparks, Nomura, & Casida, 2003). OPs phosphorylate and thereby block the action of the hydrolases FAAH and MAG. This inhibition of the hydrolysis of endogenous cannabinoids could prolong their actions on the receptor, which inhibits several neurotransmitter systems, including ACh. OPs also bind the CB1 receptor and block the binding of specific receptor ligands. The OPs do not bind at the agonist site, however, and may have no inherent agonist or antagonists consequences; however, this has not been adequately studied. Thus, prolongation of the ligand residence time, plus blockade of ligand

binding at the receptor, could lead to downstream modulation of cholinergic activity and signaling, separate from AChE inhibition. It is unclear which actions could predominate *in vivo*.

Casida and colleagues have produced several papers relating FAAH inhibition to that of AChE and neurotoxic esterase (NTE), and considered its involvement in acute cholinergic, intermediate, and delayed toxicity syndromes (*e.g.*, Casida & Quistad, 2004; Quistad, Klintenberg, Caboni, Liang, & Casida, 2006; Quistad, Nomura, Sparks, Segall, & Casida, 2002; Quistad, Sparks, & Casida, 2001; Quistad, Sparks, Segall, Nomura, & Casida, 2002; Segall, et al., 2003). Several studies have included chlorpyrifos. They have reported that *in vitro* chlorpyrifos oxon inhibits FAAH and MAG with IC50s of 40 nM and 34 nM, respectively, compared to an AChE IC50 of 19 nM. Other OPs (paraoxon, dichlorvos, diazoxon) have higher hydrolase inhibitory actions *in vitro* (IC50s 540-14,000 nM). *In vitro* receptor binding assays of CB1 agonists reveal an IC50 of 14 nM for chlorpyrifos oxon, and higher IC50s (>64 nM) for other OPs (chlorpyrifos methyl oxon, diazoxon, dichlorvos).

Casida's group has also dosed adult mice with OPs and evaluated motor behavior and cholinergic signs, followed by *ex vivo* biochemical assays. These studies report inhibition of FAAH activity and CB1 binding only at highly toxic doses of chlorpyrifos and/or chlorpyrifos oxon. Chlorpyrifos dose-response differs somewhat across studies, but in general, FAAH is inhibited more than MAG and AChE, and CB1 binding is moderately inhibited, but only at doses producing cholinergic signs. Chlorpyrifos inhibition of FAAH does not correlate with the hypomotility (akinesia, rigidity) produced by the EC agonist amandamide. Furthermore, chlorpyrifos does not potentiate the effects of amandamide, suggesting that it does not appear to have functional impacts in the EC system. In contrast, treatment with other chemicals that do produce marked (>76%) FAAH inhibition does indeed potentiate anandamide effects in mice; however, chlorpyrifos doses that would be required to produce this level of FAAH inhibition are likely to be lethal. This line of research has led Casida and colleagues to conclude that chlorpyrifos actions on the EC system *in vivo* are less important to its acute toxicity than AChE inhibition.

A series of studies from the laboratories of Pope and colleagues has evaluated the influence of CB1 agonists administered *in vivo* on the signs of toxicity produced by several OPs (Baireddy, Liu, Hinsdale, & Pope, 2011; Nallapaneni, Liu, Karanth, & Pope, 2006, 2008; C. Pope, Mechoulam, & Parsons, 2010; Wright, Liu, Nallapaneni, & Pope, 2010). They reported that CB1 agonists and other cannabinomimetics block the acute signs of exposure to paraoxon or DFP (chlorpyrifos was not tested). Studies with chlorpyrifos show increased extracellular levels of 2-AG but not amandamide in hippocampus. Somewhat different conclusions were drawn, however, in a paper showing that the toxicity and AChE inhibition produced by an acute dose of chlorpyrifos is not altered in mice without the CB1 receptor (knockout) compared to their wild type littermates. Pope et al. also measured chlorpyrifos oxon-induced ACh release from these tissues and saw only a small difference. These somewhat contradictory results suggest that potential EC involvement in the acute toxicity of certain OPs is still uncertain.

Only one study has examined chlorpyrifos effects on the EC system in developing animals. Carr et al. (2011) dosed preweanling rats for 5 days (1-5 mg/kg/day, p.o.) and took brain tissue 4 hr after the last dose. The lowest dose produced 40% inhibition of FAAH, 14% inhibition of MAG, and only 18% inhibition of AChE; in addition, the highest dose eliminated FAAH activity and produced about 52-55% inhibition of AChE. This suggests a greater sensitivity of the EC system, at least in terms of the hydrolase compared to AChE activity, in the pups. However, there was no time-course, dose-response, or other ages tested, and no downstream or correlative measure of changes in EC system function. Additional studies along these lines are needed.

In summary, most of the research in this area has evaluated acute responses in the EC system, even though such changes during a critical period of development could have long-term consequences. While some steps along a chlorpyrifos adverse outcome pathway for developmental neurotoxicity are plausible, there are remaining questions as to whether the EC system is sufficiently sensitive to low doses of chlorpyrifos to alter its function during development. Despite the increasing understanding of the EC system, there are no data during development on 1) inhibition of the CB1 receptor at any time during development (animal studies used only agonists); 2) inhibition of FAAH during development; 3) time-course and dose-response of OPs *in vivo* compared to AChE inhibition; and 4) whether these actions on the receptors and/or hydrolases actually change EC signaling to a point that would impact downstream nervous system development.

3.2.1.4. Reactive Oxygen Species

The production of reactive oxygen species (ROS) and resulting cellular damage has been proposed as a mechanism for a wide variety of neurotoxicants. Due to lower levels of protective enzymes and antioxidants, and relatively low numbers of glia relative to the adult, the developing brain may be particularly sensitive to neural cell damage caused by oxidative stress. In addition, recent work suggests that ROS can act as second messengers. Relatively small changes the oxidative status of the cell (redox potential) can lead to changes in redox sensitive signaling pathways that regulate cell physiology. In the nervous system, redox signaling is involved in the regulation of neurodevelopmental processes including neural stem cell proliferation and differentiation (Le Belle et al., 2011; Vieira, Alves, & Vercelli, 2011). A number of studies suggest that chlorpyrifos and chlorpyrifos oxon can induce oxidative stress in various neural cell types. Thus, generation of reactive oxygen species and/or alteration of cellular redox potential by chlorpyrifos represent a possible initiating event leading to developmental neurotoxicity.

Both chlorpyrifos and chlorpyrifos oxon have been shown to induce oxidative stress in neuronal cells *in vitro*. A series of studies using PC12 cells were performed by the Slotkin laboratory. Crumpton et al. (2000) reported that acute exposure of undifferentiated PC12 cells to 1.5 – 150 uM chlorpyrifos for 10 min resulted in a rapid increase in ROS production. Exposure to 30 uM chlorpyrifos oxon for 10 min had no effect. Consistent with the generation of ROS, further studies demonstrated an increase in oxidative damage (lipid peroxidation) in both undifferentiated and differentiated PC12 cells after exposure to chlorpyrifos (3 – 100 uM) for 24 hr (Qiao, Seidler, & Slotkin, 2005). Finally, Slotkin and Seidler (2009) used microarrays to examine the expression of genes involved in the response to oxidative stress after exposure of PC12 cells to 30 uM chlorpyrifos for 24 or 72 hr. They observed a significant effect on transcription of genes including glutathione peroxidase, glutathione-S-transferase, and superoxide dismutase, which was greater in differentiated cells compared to undifferentiated cells. PC12 cells were also used by a separate group of researchers who reported that very high concentrations of chlorpyrifos (1.4 – 14 mM) increased ROS production during a 5 hr exposure period (Geter et al., 2008). ROS production has also been reported in other neuronal cell types. Giodano et al. (2007) examined the effects of chlorpyrifos and chlorpyrifos oxon on ROS levels and lipid peroxidation in primary cultures of cerebellar granule cells from mice. Exposure to 1 uM chlorpyrifos for 60 min increased both ROS and lipid peroxidation, and a similar response was observed with 1 uM chlorpyrifos oxon. Saulsbury et al. (2009) examined oxidative stress in oligodendrocyte progenitor cells, which are the precursors to glial cell types in the brain. Exposure to 15 – 120 uM chlorpyrifos for 3 hr resulted in a significant increase in ROS production. These *in vitro* studies suggest that exposure of cells to the micromolar concentrations of the parent compound chlorpyrifos (which is not a potent AChE inhibitor) can result in oxidative stress. The active metabolite chlorpyrifos oxon was found to be active in cerebellar granule cells but not in PC12 cells. Inhibition of AChE was not determined in these studies.

A limited number of studies have examined the oxidative stress response in rat brain after *in vivo* exposure to chlorpyrifos. Bagchi et al. (1995) dosed adult female rats p.o. twice with 41 mg/kg chlorpyrifos, once at 0 hr and once at 21 hr. Animals were sacrificed at 24 hr. Both ROS production and lipid peroxidation were increased in homogenates from whole brain. In a follow-up study, the same dosing paradigm resulted in an up-regulation of heat shock protein, which in some cases can be a response to oxidative stress (Bagchi, Bhattacharya, & Stohs, 1996). There are two studies of oxidative stress in response to chlorpyrifos exposure in developing animals. Slotkin et al., (2005) used three dosing paradigms: s.c. exposure of pregnant rats on GD 17-20 followed by collection of the fetal brain on GD 21, s.c. exposure of rat pups on PND 1-4 followed by brain collection on PND 5, or s.c. exposure of rat pups on PND 11-14 followed by brain collection on PND 15. Lipid peroxidation was measured in brain regions including the brainstem, forebrain and cerebellum. There was a 20% increase in lipid peroxidation in the brain only after exposure to 5 mg/kg chlorpyrifos during the later postnatal period (PND 11-14) and only in males. AChE inhibition was not assessed. Ray et al. (Ray, Liu, Ayoubi, & Pope, 2010) dosed 7 day old rat pups p.o. with 0.1, 0.5, 1, or 2 mg/kg chlorpyrifos and examined gene expression in the forebrain 24 hr later using microarrays. Only the highest dose inhibited AChE (25%). Gene expression changes were observed at all doses, and 'oxidative stress' was one of the six canonical pathways that was significantly affected.

Data from both *in vitro* studies with neuronal cells (including neural precursors) and *in vivo* studies in developing brain demonstrate chlorpyrifos can induce oxidative stress. The *in vitro* data suggests that this effect may not be due to AChE inhibition, since the parent compound chlorpyrifos is either equipotent or more potent than the oxon. There was, however, no concurrent analysis of AChE inhibition in most of these studies. Several known developmental neurotoxicants have been shown to disrupt neural precursor cell proliferation *in vitro* through a common pathway that is initiated by increasing the oxidative state of the cell (Z. Li, Dong, Proschel, & Noble, 2007), and the antioxidant vitamin E protected PC12 cells from the anti-proliferative effect of chlorpyrifos (Slotkin, MacKillop, Ryde, & Seidler, 2007). Thus, the *in vitro* data suggest that chlorpyrifos can affect a critical neurodevelopmental process, at least in part, via generation of ROS. Though limited, *in vivo* studies show both direct evidence (lipid peroxidation) and indirect evidence (alteration in the expression of oxidative stress response genes) of oxidative stress in the developing brain after exposure to chlorpyrifos. Recent evidence suggests that oxidative stress can alter neurodevelopment *in vitro* and *in vivo* by the dysregulation of signaling pathways controlling neuroprogenitor cell function (Le Belle, et al., 2011; Vieira, et al., 2011). Thus, while there is the potential for initiation of an adverse outcome pathway via induction of oxidative stress, it has yet to be demonstrated *in vivo* that treatments preventing the induction of oxidative stress can ameliorate the developmental neurotoxicity of chlorpyrifos.

3.2.1.5. Serotonergic system

The serotonergic system (reviewed in Rho & Storey, 2001; *Serotonin Receptors in Neurobiology*, 2007) includes 7 distinct families of serotonin receptors regulating diverse functions such as neuronal excitability, appetite control, memory, learning, circadian rhythm, sexual behavior, anxiety, aggression, and respiration. While the majority of the receptors are either negatively or positively coupled to adenylate cyclase, the 5-HT_3 serotonergic receptor is a ligand-gated ion channel. Serotonin receptors are distributed throughout the brain and body. Synthesized from L-tryptophan, the rate limiting step in serotonin biosynthesis is catalyzed by tryptophan hydroxylase. After release, the action of serotonin is primarily terminated by reuptake into the presynaptic terminal via monoamine transporters. Various

drugs inhibit the reuptake of serotonin, effectively increasing the available synaptic concentrations and actions: Ecstasy, amphetamine, cocaine, tricyclic antidepressants and the newer antidepressants: SSRIs (selective serotonin reuptake inhibitors).

Beyond its classical neurotransmitter actions (described above), serotonin has other roles during development. In their review, Thompson and Stanwood (Thompson & Stanwood, 2009) described serotonin as a pleiotropic molecule, meaning that it can produce multiple, diverse effects, regulating different functions at different times during development. The serotonergic system is integral in many developmental processes including, but not limited to, neurogenesis, migration, and differentiation, synaptogenesis, and cardiac development before assuming its more well-known function as a neurotransmitter in the adult nervous system (reviewed in (Frederick & Stanwood, 2009). Serotonin also plays crucial roles in thalamocortical patterning (reviewed in (Frederick & Stanwood, 2009). As serotonin is present extremely early in development, it is thought that it modulates cellular function even before neurogenesis. Later in development, serotonin is temporarily taken up by so-called transient serotonergic neurons mainly involved in sensory processing, and is involved in activity-dependent patterning of the brain. Later in development, serotonin has also been shown to modulate differentiation in the brain.

Perturbation of the development of the serotonergic system in laboratory animals causes permanent changes in the neurochemical, behavioral and structural aspects of the adult nervous system (reviewed in Borue, Chen, & Condron, 2007; Daubert & Condron, 2010; Frederick & Stanwood, 2009; Oberlander, Gingrich, & Ansorge, 2009). Moreover, it is generally recognized that the development stage of serotonergic perturbation determines the developmental outcome (reviewed in Oberlander, et al., 2009; Olivier, Blom, Arentsen, & Homberg, 2011), meaning that the time at which the developing animal experienced the insult to the serotonergic nervous system would determine the degree and the result. The review by Daubert and Condron (2010) includes a table summarizing many laboratory studies where different aspects of the serotonergic system were manipulated (either through knock-out, conditional knock-out, or chemical treatment) during development accompanied by the behavioral and morphological results of that manipulation. For example, SERT (serotonin reuptake receptor) knock-out mice had alterations in sensory patterning, sleep pattern abnormalities, and long-term behavioral deficits (Q. Li, 2006), and animals treated with SSRIs during development show many similarities to the SERT knock-out mice. Developmental exposure to SSRIs in mice produced permanent (adulthood) effects on behavior (Lisboa, Oliveira, Costa, Venâncio, & Moreira, 2007; Noorlander et al., 2008) and brain chemistry (Noorlander, et al., 2008) and changes in morphology in the hippocampus (J. Zheng et al., 2011) and somatosensory cortex (L. J. Lee, 2009).

Because of the extensive usage of SSRI antidepressants in the human population, there is a rich literature SSRI effects during pregnancy and lactation (reviewed in Alwan & Friedman, 2009; Ellfolk & Malm, 2010; Gentile, 2005; Oberlander, et al., 2009; Olivier, et al., 2011; Tuccori et al., 2009). The main effects of SSRI usage are on cardiac development, and while there is some support for concluding that actions on the serotonergic system early in development can lead to long-term neurological dysfunction, there are only few studies that have studied this relationship in humans in detail. A recent study has noted that children born to mothers who used SSRIs have increased likelihood for social-behavioral abnormalities (Klinger et al., 2011).

There are numerous studies of the effects of perinatal chlorpyrifos administration on the patency of the serotonergic system. Endpoints in various brain regions include serotonin levels (Aldridge, Meyer, Seidler, & Slotkin, 2005a; Slotkin & Seidler, 2007b, 2007c), serotonin turnover (Aldridge, Meyer, et al.,

2005a; Slotkin & Seidler, 2007b, 2007c), serotonin receptor levels (Aldridge, Meyer, Seidler, & Slotkin, 2005b; Aldridge, Seidler, Meyer, Thallai, & Slotkin, 2003; Aldridge, Seidler, & Slotkin, 2004; Slotkin & Seidler, 2005), serotonin reuptake receptor levels (Aldridge, Meyer, et al., 2005b; Aldridge, et al., 2003; Aldridge, et al., 2004; Slotkin & Seidler, 2005), serotonin elicited second messenger activity (Aldridge, Meyer, et al., 2005b; Aldridge, et al., 2003; Aldridge, et al., 2004), gene expression of serotonin receptor and metabolism related genes (Slotkin & Seidler, 2007a), serotonin related behavioral assessments (Aldridge, Levin, Seidler, & Slotkin, 2005; W.-Q. Chen et al., 2011; Venerosi, Ricceri, Rungi, Sanghez, & Calamandrei, 2010), and behavior after serotonergic drug challenge (Aldridge, Levin, et al., 2005; Venerosi, et al., 2010). Chlorpyrifos was administered during gestation (Aldridge, Meyer, et al., 2005a; Aldridge, et al., 2003; Aldridge, et al., 2004; Slotkin & Seidler, 2007c; Venerosi, et al., 2010), postnatally (Aldridge, Levin, et al., 2005; Aldridge, Meyer, et al., 2005a, 2005b; Aldridge, et al., 2003; Aldridge, et al., 2004; Slotkin & Seidler, 2005, 2007a, 2007b) or during adolescent (W.-Q. Chen, et al., 2011) and the animals were assessed either shortly after dosing ceased (Aldridge, et al., 2003; Slotkin & Seidler, 2007a), or after a matter of weeks (Aldridge, Meyer, et al., 2005b; Aldridge, et al., 2003; W.-Q. Chen, et al., 2011; Slotkin & Seidler, 2007b, 2007c) or in adulthood (Aldridge, Meyer, et al., 2005a; Aldridge, et al., 2004; Slotkin & Seidler, 2005; Venerosi, et al., 2010). All the data indicate that there are acute, as well as permanent, effects of neonatal chlorpyrifos treatment on the maturation of the serotonergic nervous system. The effects are often gender-specific, region-specific and dose-related. For example, a group of papers found permanent effects of late gestational chlorpyrifos administration in rats (Aldridge, Meyer, et al., 2005a; Aldridge, et al., 2004) or mice (Venerosi, et al., 2010) on the development of the serotonergic nervous system. In rats (Aldridge, Meyer, et al., 2005a; Aldridge, et al., 2004), chlorpyrifos was administered on GD17-20 (1 or 5 mg/kg, sc in DMSO), and the adult brain showed changes in brain regional serotonin content (5 mg/kg group), increased serotonin turnover (1 and 5 mg/kg groups), increases in 5-HT_{1A} and 5-HT_2 receptor populations (1 mg and 5 mg/kg groups), increases in serotonin reuptake receptor (1 and 5 mg/kg groups), and changes in the adenylate cyclase response to serotonin in the cerebral cortex and mid brain (1 mg/kg group)—all indicative of permanent changes in the serotonergic tone of the adult brain. In a related study in mice (Venerosi, et al., 2010), chlorpyrifos was also administered during late gestation, but at a higher dose and by a different route (GD14-17; 6 mg/kg, peanut oil, gavage); the offspring were assessed upon reaching adulthood (≅ 90 days). The chlorpyrifos-treated rats showed minor differences in the behavioral tests, but when the tests were combined with a fluvoxamine (serotonin reuptake inhibitor) pharmacological challenge, there were marked differences in responses between the animals that had been exposed to chlorpyrifos during gestation and those that had not—another indication that developmental chlorpyrifos exposure had permanently altered the serotonergic tone of the adult brain.

There is ample evidence that chlorpyrifos exposure during development causes permanent changes in the serotonergic nervous system; there are, however, few papers that assessed concurrently the cholinesterase inhibition (either brain or blood) in those same animals. In some cases, although cholinesterase activity was not assessed concurrently, a dosing regimen was used that had been characterized previously with regard to cholinesterase activity. It does appear, however, that most of the studies on the effects of chlorpyrifos on the serotonergic nervous system were conducted with doses of chlorpyrifos that likely produced marked inhibition of cholinesterase activity. There is, however, one of the dosing regimens [GD17-20 (daily subcutaneous dosing with chlorpyrifos in DMSO] that has been shown to produce no fetal brain cholinesterase inhibition measured 24 hours after the last dose at the 1 mg/kg , and inhibition (~20%) at 2 mg/kg (Qiao, et al., 2002). As noted above, this level of gestational exposure to chlorpyrifos permanently altered the development of the serotonergic nervous system (Aldridge, Meyer, et al., 2005a; Aldridge, et al., 2004).

In summary, with regard to development of the chlorpyrifos adverse outcome pathway for the serotonergic nervous system: the serotonergic system is intregal to mammalian brain development and function; perturbation of that system during development leads to lasting effects in mammals; and exposure to chlorpyrifos during the perinatal period can permanently change the function of serotonergic system. Therefore, as many steps in this chlorpyrifos adverse outcome pathway are possible and plausible, and in laboratory animals the serotonergic nervous system is sufficiently sensitive to low doses of chlorpyrifos during development to alter its function, it is plausible that exposure to chlorpyrifos during development could alter brain development and the function of the serotonergic nervous system. Although chlorpyrifos effects on the serotonergic nervous system in laboratory animals likely is initiated within 24 hours (Slotkin & Seidler, 2007a), the actual initiating event of this potential adverse outcome pathway is unknown.

3.2.1.6. Tubulin, Microtubule Associated Proteins and Axonal Transport

Microtubules, one component of the dynamic cytoskeletal scaffolding within each cell, are composed of heterodimers of α- and β-tubulin, as well as microtubule associated proteins. The microtubule associated proteins appear to have three main functions: (1) to stabilize the microtubules; (2) to aid in tubulin dissociation and (3) to act as motor proteins moving substances forward and backward along the microtubules (Avila, Dominguez, & Diaz-Nido, 1994; Pellegrini & Budman, 2005; Sánchez, Díaz-Nido, & Avila, 2000). Not only does the microtubule cytoskeleton determine neuronal morphology (Matus, 1988, 1990; Sánchez, et al., 2000), but the dynamic reorganization of the microtubules and microtubule associated proteins within a cell may also coordinate neurite extension/retraction, as well as growth cone advancement. In addition to these integral roles in brain structure and growth, microtubules and the microtubule associated motor proteins kinesin (Hirokawa & Noda, 2008) and dynein (Vallee, Williams, Varma, & Barnhart, 2004) also provide a "railway" for transport of materials throughout the cell, *i.e.*, axonal transport (Fukushima, Furuta, Hidaka, Moriyama, & Tsujiuchi, 2009), another process which is integral to the health of the central and peripheral nervous system, playing a pivotal role in neuronal network formation and synapse maturation (Hirokawa & Takemura, 2004).

During brain development, the pattern of expression of microtubules (Bond & Farmer, 1983; Denoulet, Edde, & Gros, 1986; Meininger & Binet, 1989; Moskowitz & Oblinger, 1995), the post-translational modification of the tubulin (Fukushima, et al., 2009; M. K. Lee, Rebhun, & Frankfurter, 1990), and the spectrum of microtubule associated proteins (Fischer & Romano-Clarke, 1990; Nunez, 1986; Sánchez, et al., 2000) are all developmentally regulated. Because the phosphorylation of both microtubules and microtubule associated proteins are integral to their functional roles in the nervous system (Avila, et al., 1994; Fischer & Romano-Clarke, 1990; Fukushima, et al., 2009; Sánchez, et al., 2000), there is concern that exposure to OPs may affect the phosphorylation status and consequently the function of these molecules.

One of the shortcomings of constructing this adverse outcome pathway is that there are no studies on tubulin dynamics, microtubule associated proteins or axonal transport in developing animals, but because tubulin dynamics, microtubule associated proteins and axonal transport are so integral to the development and maintenance of the nervous system, the available adult data are first given in detail and then summarized below.

Adult rats treated for 14 days with chlorpyrifos (sc in peanut oil; 2.5, 10, 18, or 25 mg/kg; all doses produced plasma cholinesterase inhibition) show depressed anterograde and retrograde axonal transport in the sciatic nerve 6 days after cessation of treatment 2003 (Terry et al., 2003). This same

laboratory followed up in 2007 (Terry et al., 2007) with another, slightly different, study where axonal transport was assessed in adult rats given chlorpyrifos repeatedly (sc, 3% DMSO, 97% peanut oil vehicle, every other day for 30 days; 18 mg/kg). Depressed bidirectional axonal transport was noted after 1 dose, after the 30 days of dosing or 2 weeks after cessation of treatment. This dose produced about 30-50% brain cholinesterase inhibition 14 days after last dose. In that same year, the Terry laboratory published an *in vitro* paper (Gearhart, Sickles, Buccafusco, Prendergast, & Terry, 2007) studying the effect of chlorpyrifos or chlorpyrifos oxon on kinesin-dependent microtubule motility (the mechanism of anterograde axonal transport). Using tubulin and kinesin prepared from bovine brain, they assessed kinesin-mediated movement of microtubules. This was done by either preincubating the tubulin or the kinesin fraction with chlorpyrifos or chlorpyrifos oxon and then combining the fractions and assessing movement of the microtubules. They obtained the same interesting pattern using chlorypyrifos or chlorpyrifos oxon: there was no effect if preincubated with microtubules, but marked effect (increased detachment of the microtubules) if preincubated with kinesin. This assay contains no cholinesterase activity, thereby precluding inhibition assessment. A recently published *in vitro* paper (Middlemore-Risher, Adam, Lambert, & Terry, 2011) assessed the effect of chlorpyrifos or chlorpyrifos oxon transport of mitochondria in rat cortical neurons. They found dose-related effects of chlorpyrifos or chlorpyrifos oxon on total distance moved and number of moving mitochondria. All doses produced cholinesterase inhibition except lowest dose of chlorpyrifos (1 uM) or chlorpyrifos oxon (5 nM), and those doses still affected movement. Interestingly the toxic effect was not altered by co-treatment of either a nicotinic or muscarinic receptor blocker, an indication that the depressed transport was not mediated through cholinergic receptors. Subsequent to demonstrating that *in vitro* chlorpyrifos oxon appeared to alter tubulin dynamics (Grigoryan & Lockridge, 2009) and also covalently bound to tubulin (Grigoryan et al., 2008), the Lockridge laboratory assessed tubulin and microtubule associated proteins in the brains of mice that had been dosed *in vivo* with chlorpyrifos or chlorpyrifos oxon (Jiang et al., 2010). Repeated (14 days) dosing with chlorpyrifos (sc, 3% DMSO/97% peanut oil, 3 mg/kg) produced a transient decrease in blood AChE activity, no change in blood carboxylesterase activity and a steady decline in blood BuChE activity. Brain β-tubulin from the animals treated with chlorpyrifos had been covalently modified (diethosyphosphorylated) on tyrosine 281 (the residue also found in the *in vitro* study (Grigoryan, Schopfer, et al., 2009). This same paper also described treating adult mice with a single dose of chlorpyrifos oxon (3 mg/kg; ip); this dose produced toxic signs and marked AChE and BuChE inhibition in the plasma. Brain tubulin was then purified, polymerized, the proteins separated by gel electrophoresis and the proteins identified by mass spectrophometry. The polymerized tubulin was also subjected to atomic force microscopy nanoimaging to assess the structure (width and height) of the microtubules. Microtubules from chlorpyrifos oxon- treated brains were missing 6 microtubule associated proteins. This observation was supported by the nanoimaging showing that microtubules from chlorpyrifos oxon-treated animals were narrower and shorter than control brain microtubules and also had fewer attached (decorated) proteins than do the microtubules from control brains. It interesting to note that an *in vitro* assessment (Prendergast et al., 2007) of the effects of chlorpyrifos oxon tubulin/microtubule associated protein dynamics in a 8 day old rat hippocampal slice preparation showed all concentrations of chlorpyrifos oxon decreased mircrotubule associated protein- staining and decreased microtubule associated protein mediated tubulin polymerization by about 45%; all chlorpyrifos-oxon doses inhibited AChE.

In summary, in the late 2000s, a number of papers were published on the *in vitro* modification of various proteins by chlorpyrifos or chlorpyrifos oxon (Grigoryan, Li, et al., 2009; B. Li et al., 2009), including tubulin (Grigoryan, Li, et al., 2009; Grigoryan & Lockridge, 2009; Grigoryan, Schopfer, et al., 2009; Grigoryan, et al., 2008). Although interesting and provocative, these studies were usually conducted with exceedingly high concentrations (high micromolar to millimolar) of the OP compound,

making the connection to a "real world" human exposure tenuous. In 2010, however, Jiang and coworkers published an *in vivo* study in which they found covalently labeled tubulin in the brains of mice treated with chlorpyrifos or chlorpyrifos oxon, showing the real-world possibility of chlorpyrifos-induced covalent modification of brain tubulin in mammals at doses that produced minimal blood cholinesterase inhibition (Jiang, et al., 2010). Other studies have shown perturbations in tubulin dynamics, perturbations in the amount of tubulin protein or microtubule associated proteins, and changes in axonal transport elicited by treatment with chlorpyrifos or its oxon. Submicromolar concentrations of chlorpyrifos oxon markedly inhibited tubulin polymerization *in vitro* (Prendergast, et al., 2007), while *in vitro* treatment with chlorpyrifos oxon (but not chlorpyrifos) reduced that amount tubulin in rat C6 glioma cells (Sachana, et al., 2008). Decreases in the amount of microtubule associated proteins after chlorpyrifos or chlorpyrifos oxon treatment has been reported by three separate studies: two *in vitro* studies both showed decreases after chlorpyrifos oxon treatment (Prendergast, et al., 2007; Sachana, et al., 2008), and one *in vivo* study showed extensive reduction in specific microtubule associated proteins in the brains of mice treated with chlorpyrifos oxon (Jiang, et al., 2010). Depressed axonal transport has been noted by many laboratories both *in vivo* (Terry, et al., 2007; Terry, et al., 2003) and *in vitro* (Gearhart, et al., 2007; Middlemore-Risher, et al., 2011) using both chlorpyrifos (*in vivo* and *in vitro* studies) and chlorpyrifos oxon (*in vitro* studies). In two instances (Gearhart, et al., 2007; Middlemore-Risher, et al., 2011), the effects of the chlorpyrifos and oxon were noted at concentrations of chlorpyrifos or chlorpyrifos oxon that did not inhibition cholinesterase activity (Middlemore-Risher, et al., 2011) or in systems that were so basic (purified microtubule/kinesin) that no cholinesterase was present (Gearhart, et al., 2007).

The construction of an adverse outcome pathway using chlorpyrifos-induced effects on tubulin and microtuble associated proteins is still in its infancy mainly because although it is thought that tubulin, microtubule associated proteins and axonal transport are integral to nervous system development and maintainence, there is no experimental evidence that perturbations of these endpoints by chlorpyrifos during development has neurotoxic outcomes.

3.2.1.7 Summary/Conclusions

As can be seen from the section above, there are several lines of evidence for actions of chlorpyrifos distinct from the classical mode of action of cholinesterase inhibition. This information has been generated from model systems representing different levels of biological organization, and provide support for molecular initiating events (binding to the morphogenic site of AChE, muscarinic receptors, or tubulin), cellular responses (alterations in neuronal proliferation, differentiation, neurite growth, or intracellular signaling) and responses at the level of the intact nervous system (serotonergic tone, axonal transport). In some cases these effects occur at chlorpyrifos exposure levels below or equivalent to those which result in cholinesterase inhibition in the same test system. When taken in aggregate, data from individual studies can be used to develop plausible hypotheses for a mode of action leading to developmental neurotoxicity (*e.g.* inhibition of the morphogenic action of AChE leading to reduced neurite outgrowth and subsequent miswiring or dysmorphology of the brain); however, as is the case for many other developmental neurotoxicants, most of these studies have not been designed with the specific goal of construction or testing an adverse outcome pathway. Thus, there are not sufficient data available to test rigorously the causal relationship between effects of chlorpyrifos at the different levels of biological organization in the nervous system. This is not surprising in light of the complex, intricate, and interrelated dynamic processes ongoing in the developing brain and multiple critical windows for exposure. There are a number of known developmentally neurotoxic chemicals with well-established relationships between exposure and neurological disorders in humans for which a definitive mode of

action has not been established: for example lead, methyl mercury, and ethanol (Alfonso-Loeches & Guerri, 2011; Castoldi et al., 2008; Farina, Rocha, & Aschner, 2011; Johansson et al., 2007; Verstraeten, Aimo, & Oteiza, 2008). Even today, armed with thousands of published papers on these three, accepted, developmental neurotoxicants, no coherent adverse outcome pathway or pathways can be constructed, because there are a multitude of initiating toxic events and cellular responses put forth, and they are not positively connected with one another. The only adverse outcome pathway available for any form of neurotoxicity has been developed for domoic acid in adult animals (Watanabe et al., 2011). Domoic acid was chosen for this case study due to a large amount of preexisting data and a molecular initiating event at a receptor (glutamate) with known consequences to its overactivation in the adult nervous system.

3.2.2 Adverse Outcomes in Laboratory Animals: Effects on the Developing Brain

There is a considerable and growing body of literature on the effects of chlorpyrifos on the developing brain of laboratory animals (rats and mice) indicating that gestational and/or postnatal exposure may cause persistent behavioral effects into adulthood. These data provide support for the susceptibility of the developing mammalian brain to chlorpyrifos exposure. The literature was summarized and a preliminary review was provided by the Agency for the 2008 SAP. At that time, the Panel agreed that exposure to doses of 1 mg/kg/d and greater, during some developmental period, produced significant and long-term effects on behavior. While behavioral changes were consistently reported, they were somewhat inconsistent, most likely due to experimental design differences. Such factors include route of exposure, developmental period of exposure, test methods, choice of dependent variable, statistical analyses, and so on, which are critical features of all developmental neurotoxicity studies.

In this section the Agency reviews new literature, *i.e.*, papers published since 2008, to build on the earlier SAP review. The new information provided is evaluated in terms of concordance with, or divergence from, the previous, pre-2008 findings. Overall, these new studies serve to strengthen the findings on which the 2008 SAP made their conclusions and further documents the scope of long-term behavioral effects resulting from chlorpyrifos exposure during development.

Papers considered herein by the Agency as addressing long-term outcomes from developmental exposure include only those where chlorpyrifos is administered during the preweaning period (either gestational and/or postnatal) and the offspring are examined at some time after weaning. That is, papers reporting evaluations shortly after birth or during the pre-weaning period do not reflect long-term consequences and may also be confounded by AChE/ChE inhibition during concurrent or recent exposure; thus, those data are not discussed further here. In addition, the Agency focused its efforts on studies using relatively low doses, and that did not (or would not be expected to) produce a considerable degree of brain AChE inhibition and resultant cholinergic toxicity. These constraints aid in the unencumbered evaluation of longer-term effects compared to acute impacts of AChE inhibition, but it does limit the use of some papers.

To date, 25 papers are identified and evaluated by the Agency; nine have been published from 2008 to the present (Abou-Donia et al., 2006; Aldridge, Levin, et al., 2005; Billauer-Haimovitch et al., 2009; Braquenier, Quertemont, Tirelli, & Plumier, 2010; Carr et al., 2001; Chakraborti, Farrar, & Pope, 1993; Chanda & Pope, 1996; Dam, Seidler, & Slotkin, 2000; Haviland, Butz, & Porter, 2010; Icenogle et al., 2004; D. A. Jett, Navoa, Beckles, & McLemore, 2001; Johnson, Chambers, Nail, Givaruangsawat, & Carr, 2009; Laviola, Adriani, Gaudino, Marino, & Keller, 2006; Levin et al., 2002; Levin et al., 2001; Maurissen, et al., 2000; Muto, Lobelle, Bidanset, & Wurpel, 1992; Ricceri et al., 2003; Ricceri et al., 2006; Venerosi,

Calamandrei, & Ricceri, 2006; Venerosi et al., 2008; Venerosi, et al., 2010; Venerosi, Ricceri, Scattoni, & Calamandrei, 2009; Zhang et al., 2011). It is important to appreciate the various experimental designs, encompassing species, gender, exposure scenarios, ages of assessment, and test apparatuses. Of these 25 papers, four papers and one study from a fifth paper did not meet the criteria stated above, and have been excluded from further consideration herein.

Specifically, these excluded studies are:

1) Muto et al. (1992) used a chlorpyrifos product that was formulated in xylene, a known developmental and reproductive toxicant, yet the vehicle control group received saline, not xylene. Thus, the findings of that study cannot be attributed to chlorpyrifos alone. Furthermore, all testing took place before weaning, no later than PND16;

2) A gestational-exposure study (GD12-19) used a dose (25 mg/kg/d sc) that produced 90% brain inhibition in the dams at the end of dosing, and ~20% brain inhibition in offspring on PND3 (Chanda & Pope, 1996). The reported motor effects are confounded with the concurrent cholinesterase inhibition on PND1 and 3, and no testing was conducted at later ages;

3) A postnatal study (Chakraborti, et al., 1993) used a very high dose (40 mg/kg/d sc) given directly to the pups postnatally (PND7, 11, 15, 19), with up to 60% brain inhibition evident four days after dosing;

4) A gestational study (Venerosi, et al., 2009) only tested pups up to PND12, reporting altered motor ontogeny and changes in ultrasonic vocalizations; and

5) The "postnatal" study described by (D. A. Jett, et al., 2001) involved dosing after weaning, while the rats were being tested in the Morris water maze. Note, part of this paper remains in consideration, since the "preweaning" study from that paper met the criteria for inclusion.

3.2.2.1 Summary of Existing Database

Of the 21 papers in the Agency's review, several trends are apparent. The numbers of rat and mouse studies are similar (11 rat, 10 mice). As seen in Table 5 below, this research now spans over a decade, with the first publications in 2000. These studies have been generated in ten different laboratories, four of which have been added since SAP literature review in 2008. Of the nine studies published in 2008 and later, seven evaluated mice and the remaining two used rats; however, one of the mouse studies was excluded from further discussion since there were no assessments conducted after weaning (Venerosi, et al., 2009). Most, but not all, of the studies include testing in both males and females. Two laboratories have generated the most research: Istituto Superiore di Sanita (ISS; Rome, six studies), and Duke University Medical Center (NC, USA, six studies). The majority of studies (14) used subcutaneous administration, eight used oral gavage, one employed an osmotic minipump, and one used a dermal route. Of the two major contributing laboratories, Duke University always uses subcutaneous administration of chlorpyrifos in DMSO vehicle, whereas the ISS laboratory uses oral dosing of the dams with peanut oil vehicle during gestation, and subcutaneous dosing of the pups postnatally with either peanut oil or DMSO as vehicle. Eight studies involved only gestational exposures, nine used only direct postnatal exposures to the pups, and two spanned gestational and lactational (LD) periods. Note that two studies actually included both gestational and direct postnatal exposures in the same subjects, and

this more complicated experimental design resulted in all possible combinations of doses both pre- and postnatally. For the discussion below, only treatments that included chlorpyrifos at one or the other dosing period, but not both, are included (*i.e.*, vehicle followed by chlorpyrifos, or chlorpyrifos followed by vehicle); however, these data are listed in Appendix 3. All studies used chlorpyrifos except one (Laviola, et al., 2006) who used the oxon metabolite. One study is in Chinese, with only the abstract available in English, and the route is not specified. Only two of the new papers include any measures of AChE inhibition, whereas a few others refer to earlier papers from the laboratory with such measurements.

Table 5 Summary of Developmental Chlorpyrifos Studies by Laboratory

Laboratory	Principal Investigator(s)	Number of Studies	Year(s) of publication
Duke University, NC, USA	Slotkin, Levin; Abou-Donia	6	2000, 2001, 2002, 2004, 2005, 2006
Mississippi State University, MS, USA	Carr	2	2001, 2009
Dow, MI, USA	Maurissen	1	2000
Johns Hopkins University, MD, USA	Jett	1	2001
University of Wisconsin Madison, WI, USA	Haviland	1	2010
Istituto Superiore di Sanita, Rome, Italy	Calamandrei; Laviola	6	2003, 2006, 2006, 2006, 2008, 2010
Hebrew University, Jerusalem, Israel	Yanai	2	2011
University of Liége, Belgium	Plumier	1	2010
Central South University, Changsha, China	Zhao	1	2011

In addition to differences in species, gender, and route and timing of exposure, evaluation of the existing database shows a lack of consistency for the specific methods, equipment, variables, and analyses of the various tests. For example, locomotor activity has been measured in various chambers of differing size and configuration, data are collected differently (*e.g.* automated or visual scoring), and measures may vary (*e.g.*, latency for a response or area of exploration). While conclusions are based on statistical tests of significance, differences across the papers range from restricting the alpha level (*e.g.*, $p<0.02$) to prevent type I errors to accepting interaction terms of higher probabilities (*e.g.*, $p<0.10$) to continue with post-hoc determinations. Despite these differences, the discussion below is based on the authors' conclusions and interpretations. While this situation is not unusual in the developmental neurotoxicity literature, it does render direct comparisons across studies more difficult (Cory-Slechta et al., 2001). For this reason, we and others (*e.g.*, (Prueitt, Goodman, Bailey, & Rhomberg, 2011) have focused more on evaluating outcomes in terms of overall domains of neurological function rather than the specific endpoints used to measure that function.

3.2.2.2 Developmental Impacts on Neurological Domains

Because many of these papers report a number of positive as well as negative findings, the Agency had previously taken the approach of comparing responses that were observed following various exposures to a common dose, 1 mg/kg/d ((FIFRA Scientific Advisory Panel (SAP), 2008a; U.S. Environmental Protection Agency, 2011). A more robust approach is taken here, to include important factors such as dose-response and differences in exposure scenarios. In order to evaluate effects on domains of neurological functions, broken down by exposure period, the papers have been summarized as such in Appendix 3. It is well-known that exposures during different critical periods of development can result in very different outcomes (Adams et al., 2000; Rice & Barone, 2000). Unfortunately, development of the nervous system does not occur in distinct non-overlapping temporal periods with which to attribute the apical behavioral changes observed. In the text below, the more recent papers (2008 to present) are described and evaluated in terms of their contribution to and concordance with, previous papers using similar measures. The Agency's evaluation follows several Bradford Hill criteria including consistency of findings across domains and dose response. The latter was considered as either graded magnitude of effects across doses or significant findings at a higher but not lower dose; unfortunately, many of the chlorpyrifos studies have evaluated only one dose.

The new studies include measures that evaluate the following neurobehavioral domains: cognitive function, anxiety and emotion, social behavior and maternal interactions, and motor activity. These are discussed in greater detail below; overall, the newer data were in general agreement with previous studies. Although a few earlier studies addressed neuromotor development (Abou-Donia, et al., 2006; Maurissen, et al., 2000), this domain has not been evaluated more recently; with no new information, earlier reviews have not changed.

3.2.2.2.1 Cognitive Function

A few of the new studies evaluated effects on cognitive function, using some of the same procedures used in the earlier studies (radial arm maze, Morris water maze). The earlier findings are summarized below, along with comparisons to the newer studies.

Radial Arm Maze

The radial arm maze is a spatial task requiring the subject to enter different arms for food reinforcer, which is located in only certain arms. A widely used test of learning and memory, both working errors (re-entering baited arms after taking the food) and reference errors (entering arms that are never baited) are recorded over days of training. From 2001-2005, a series of behavioral studies from a single laboratory (Duke University) described effects on radial arm maze performance in adult rats that were treated with chlorpyrifos during gestational or postnatal development. Both working and reference memory are altered in rats treated during gestation or early postnatally, and gender differences were described in all but one study. Across these studies, significant differences were detected in one or the other type of error, or both, and error selectivity was not fully consistent across studies. Following early gestational exposure (GD9-12), high-dose (5 mg/kg/d) rats of both sexes showed increased working and reference memory errors early in training (Icenogle, et al., 2004). With later gestational exposure (GD17-20), only females showed more errors; however, this was observed only at the low dose (1 mg/kg/d, not 5 mg/kg/d) (Levin, et al., 2002). With early postnatal exposure (PND1-4, 1 mg/kg/d), two separate studies showed similar results in that error rate was increased in males (worse performance) but decreased in females (better performance) (Aldridge, Levin, et al., 2005; Levin, et al., 2001). Only

the late postnatal exposure (PND11-14, 5 mg/kg/d) was without effect on this measure (Levin, et al., 2001).

Several new studies have confirmed chlorpyrifos impacts on learning and memory. In a new study using the radial arm maze and longer postnatal exposures (PND1-21), male, but not female, rats showed more working memory errors at all doses (lowest dose, 1 mg/kg/d) (Johnson, et al., 2009). This was evident later in training, indicating that the task was not learned as well as in controls. Reference memory errors showed the same gender-specific pattern as seen in two Duke studies (Aldridge, Levin, et al., 2005; Levin, et al., 2001), in that chlorpyrifos-treated females showed fewer errors during training, and males showed more. The altered reference memory pattern was only seen in the mid and high dose groups, which had incrementally increasing doses (up to 4 or 6 mg/kg/d) during the exposure period. In this study, the low-dose group had experienced about 14% brain AChE inhibition on PND20, with no inhibition evident when radial arm maze training began (PND36). On the other hand, the mid and high-dose rats showed some residual AChE inhibition at that time, making it challenging to separate the memory deficits from concurrent AChE holinesterase inhibition.

A study using mice exposed gestationally (GD17-20, 1 or 5 mg/kg/d) also used a radial arm maze, although comparisons are difficult due to differences in length of training and data presentation (Haviland, et al., 2010). Reference memory errors showed some differences across trial, dose, and sex; these are not interpreted by the authors as a meaningful effect, and may indeed be spurious. The authors used these data to compare with their novel foraging task, which used a modification of a radial arm maze and examined the rate of learning that a reward (food) is present (recognition) as well as its location (positional learning). Both of these parameters were altered by chlorpyrifos exposure. The low dose (1 mg/kg/d) females were delayed in learning to recognize the reward, but there were no statistically significant differences at that dose in positional learning (despite the authors' claim of an effect). On the other hand, low-dose males showed accelerated food recognition, and increased positional learning that was evident during only two sessions. The high-dose groups (both sexes) showed the same pattern of changes, with somewhat greater magnitude of differences, indicating dose-response. Thus, effects were observed on a spatial learning task, albeit a different apparatus and procedure. Note that the gender effect was reversed from that reported above for rats in the radial arm maze – these differences could be due to species or testing apparatus.

Morris Water Maze

The Morris water maze is a different type of spatial learning task, which can be varied to assess different types of learning and memory. In this test, the animals are trained over days to swim to the location of a submerged platform to escape from the water, and learning is evident by faster latencies and other measures of memory for the platform position. A recent paper using gestational exposure (GD9-18) in mice resulted in slower learning in the offspring, although a clear dose-response was not evident (Billauer-Haimovitch, et al., 2009). The authors report an overall effect of chlorpyrifos treatment, but state that only the lower doses (1, 3 mg/kg/d, but not 5, 10 mg/kg/d) were individually significant. While the magnitude of effect does not appear to be pronounced in the first study (visual inspection of Figure 3 in the paper), the finding was repeated, and was more obvious, in two additional studies using a single dose (3 mg/kg/d). Another recent paper from the same laboratory again reported slower learning in mice exposed during gestation (3 mg/kg/d; GD9-18) (Turgeman et al., 2011). Thus, there are internal replications of this finding across several studies, albeit in the same laboratory.

These recent findings, plus another study available only in Chinese[17], extend an earlier report of rats that showed deficits in the Morris water maze (beginning on PND23) following exposure on PND7, 11, and 15 (0.3, 7 mg/kg/d) (D. A. Jett, et al., 2001). The 2008 SAP, however, was critical of this paper, for reasons described below.

Summary

Taken together, these studies in rats and mice show altered cognitive function using well-accepted tests of spatial learning and memory (radial arm maze, Morris water maze). The direction of change may be sex-specific and dependent on the timing of exposure. Often these changes suggest impaired learning and/or memory. While enhanced function is also apparent in some studies, such changes are evidence of alterations in memory processes nonetheless. Several of these findings have recently been replicated across studies and laboratories. Effects were also reported in a spatial foraging task (Haviland, et al., 2010), but direct comparison between that and the radial arm maze is difficult. Earlier papers have reported that other cognitive tasks (spontaneous or delayed alternation, passive avoidance) are not altered, but there are no new studies using these other tasks. These outcomes are summarized in Table 6. In this and the following tables, details including dose, route of exposure and age at testing are not included but are provided in Appendix 3.

[17] Another new study also evaluated Morris water maze learning, using rats exposed postnatally (PND11-14, 5 mg/kg/d, route uncertain) (Zhang et al., 2011). The abstract states that learning and memory impairments were observed, but the rest of the paper cannot be critically evaluated at this time (article is in Chinese) and so cannot be combined with other findings.

Table 6 Summary of Outcomes on Cognitive Tests in Male (M) and Female (F) Rodents

	Early gestation GD9-12; 9-18	Late gestation GD 17-20	Perinatal GD6-LD10	Early postnatal PND1-4	Late postnatal PND11-14; 7,11,15	Postnatal PND1-21
Radial Arm Maze	Cognitive deficit - rat, M&F, dose-response[6]	Cognitive deficit - rat, F not M, no dose-response[4]		Cognitive deficit in M, improved function in F - rat[3] Cognitive deficit in M, improved function in F - rat[7]	No effect – rat, M&F[3]	Cognitive deficit in M, improved function in F - rat, dose-response[9]
Morris Water Maze	Cognitive deficit - mouse, M&F, no dose-response[8] Cognitive deficit – mouse, M&F[11]				Cognitive deficit - rat, M&F, dose-response[2]	
Foraging Maze		Cognitive deficit in F, improved function in M – mouse, dose-response in F not M[10]				
T-maze Spontaneous Alternation	No effect – rat, M&F[6]	No effect – rat, M&F[4]		No effect – rat, M&F[3]	No effect – rat, M&F[3]	
Delayed Spatial Alternation			No effect – rat, M&F[1]			
Passive Avoidance				No effect – mouse, only M tested[5]	No effect – mouse, only M tested[5]	

[1] Maurissen et al., 2000
[2] Jett et al., 2001
[3] Levin et al., 2001
[4] Levin et al., 2002
[5] Ricceri et al., 2003
[6] Icenogle et al., 2004
[7] Aldridge et al., 2005
[8] Billauer-Haimovitch et al., 2009
[9] Johnson et al., 2009
[10] Haviland et al., 2010
[11] Turgeman et al., 2011

3.2.2.2.2 Anxiety/Emotion

Anxiety and despair/affect have been modeled in animals using a number of different tests, including those that measure avoidance of aversive areas, investigation of novel areas, response to forced activity, and response to preferred substances. Several new studies assessed anxiety, despair, and affect. While different procedures were used in most cases, there was some concordance in outcomes. These are described below.

Tests of Anxiety

Several studies have suggested that chlorpyrifos alters measures of anxiety. The early studies came from the laboratories of Duke University and ISS (Italy). Three newer studies, all in mice, add to the findings in this area: two were from the ISS laboratory and one came from a third laboratory (Belgium). Earlier studies employed an elevated plus maze, which has open and enclosed areas: being fearful of bright open areas, rodents tend to stay in the enclosed arms. Decreased anxiety is inferred from changes such as increased time in the open arms, decreased head dipping, and other measures. Using this paradigm, lower anxiety was reported in rats (males, not females) exposed postnatally to 1 mg/kg/d (PND1-4)(Aldridge, Levin, et al., 2005; Ricceri, et al., 2003) and in mice (females, not males) exposure postnatally to 3 mg/kg/d (PND11-14) (Ricceri, et al., 2006). On the other hand, increased anxiety was measured in mice exposed gestationally (GD15-18), but only in the low-dose (3 mg/kg/d, not 6 mg/kg/d) males (not females) (Ricceri, et al., 2006). There were no changes in anxiety behaviors in rats exposed gestationally (GD9-12, 1 or 5 mg/kg/d) (Icenogle, et al., 2004).

Three new studies have used a different apparatus, the light-dark box, to measure anxiety, but the principle is the same in that rodents typically prefer the dark chamber. In one study (Braquenier, et al., 2010), both the elevated plus maze and the light-dark box were used, providing direct comparisons between these tests. Mice were exposed both gestationally and lactationally (GD15-LD14, 0, 0.2, 1, or 5 mg/kg/d), and only female offspring were tested. When tested in the light-dark box, the middle dose group only (1 mg/kg/d) showed decreased time in the center of the light side, which was considered to reflect increased anxiety. The same dose group moved back and forth between the sides less often, which could reflect general activity levels but was also interpreted by the authors as greater anxiety. The time in the dark and light sides, however, did not differ. Littermates were tested in the elevated plus maze, and again the middle dose group showed less time spent in the open arms and fewer open arm entries, supporting a conclusion of increased anxiety, but only at the middle dose. The lack of dose-response was not addressed by the authors.

A study in which mice were treated gestationally (GD14-17, 6 mg/kg/d) (Venerosi, et al., 2010) showed no difference in time spent on either side of the light-dark box, but females (not males) spent more time in the tunnel connecting the sides. This finding, along with a few other measures that did not reach statistical significance, was interpreted by the authors as increased anxiety. Another study by this group (Venerosi, et al., 2008) exposed mice postnatally (PND11-14, 3 mg/kg/d), then bred the female offspring as adults and allowed them to deliver normally. On postpartum day 2, the dams were removed from the pups and tested in the light-dark box. They reported a decreased latency and higher proportion of mice entering the light side, indicating decreased anxiety. Time in the dark or light sides was not reported.

Despair/Affect

Only a few studies have also included measures of despair and affect, and the results are not completely consistent. Most recently, behavioral despair was measured using a forced swimming procedure (Venerosi, et al., 2010). In that study, gestational exposure to chlorpyrifos did not alter any baseline responses, and there are no other studies using similar measures with which to compare. There have been no new studies that could add to, and aid in the interpretation of, the findings reported by others: decreased preference for chocolate milk in rats, or no effect on novelty exploration in mice (Aldridge, Levin, et al., 2005; Ricceri, et al., 2003).

Summary

Taken together, these assessments suggest that, in both rats and mice, changes in anxiety are dependent on exposure period. Specifically, postnatal exposure decreases anxiety, whereas increased anxiety is observed following late gestational exposure or a longer gestational plus postnatal exposure. Some inconsistencies are evident, such as lack of dose-response in a few studies and effects on one sex or the other (same laboratory, similar dosing regimen). Thus, while the data are not fully consistent, overall there is evidence for long-term changes in anxiety behavior following chlorpyrifos exposure, as shown in Table 7.

Table 7 Summary of Anxiety/Emotion Outcomes in Male (M) and Female (F) Rodents

	Early gestation GD9-12	Late gestation GD14-17; GD15-18	Early postnatal PND1-4	Late postnatal PND11-14	Perinatal GD15-LD14
Elevated Plus Maze (anxiety)	No effect – rat, M&F[1]	Decreased anxiety - mouse, M not F, no dose-response[5]	Decreased anxiety - rat, M not F[2]	Decreased anxiety - mouse, F not M, dose-response[5]	Increased anxiety - mouse, only F tested, no dose-response[3]
Light-Dark box (anxiety)		Increased anxiety - mouse, F not M[7]		Decreased anxiety - mouse, dams[6]	Increased anxiety - mouse, only F tested, no dose-response[3]
Despair		No effect – mouse, M&F[7]			
Novelty/preference			Decreased anxiety - rat, M&F[2] No effect – mouse, M&F[4]	No effect – mouse, M&F[4]	

[1] Icenogle et al., 2004
[2] Aldridge et al., 2005
[3] Braquenier et al., 2010
[4] Ricceri et al., 2003
[5] Ricceri et al., 2006
[6] Venerosi et al., 2008
[7] Venerosi et al., 2010

3.2.2.2.3 Social Behaviors/Interactions

Conspecific behaviors are not typically studied in the context of developmental neurotoxicity studies, and methods for such assessments are not well-standardized (Cory-Slechta, et al., 2001). Aggressive, social, and parental behaviors have been studied following developmental exposure to chlorpyrifos, all of which were conducted in the ISS laboratory using mice. Even within this one laboratory, however, various testing methods have been used. These include: 1) social investigation of nulliparous female:female pairs; 2) social investigation and agonistic behaviors in male:male pairs; 3) maternal behaviors towards pups, induced by placing nulliparous females with foster litters; 4) maternal behaviors in lactating dams towards their own pups, and 5) agonistic behaviors in lactating dam:male pairs. In most studies, measures include social (interactive) as well as nonsocial behaviors (*e.g.*, grooming, exploration). Within each grouping, there are often many measures taken, and treatment effects have sometimes been reported on just a few, suggesting that the changes are subtle. There are few systematic comparisons of chlorpyrifos effects across these varied behaviors using similar dosing regimens.

Maternal Behavior

Two recent papers focused on maternal behavior, measuring actions of the lactating dam towards her pups, nesting activity, and agonistic behaviors towards an intruder male. In one study (Venerosi, et al., 2008), mice were exposed postnatally (PND11-14, 3 mg/kg/d), mated as adults, and the dams were tested. They reported that the treated dams showed delayed start of nesting, decreased latency to lick pups, as well as fewer defensive postures and more social investigation of an intruder male. This latter effect was replicated in that decreased aggressive behaviors were also reported for lactating dams that had been exposed in their own fetal period (GD14-17, 6 mg/kg/d) (Venerosi, et al., 2010). These findings were interpreted by the authors as impaired maternalistic behaviors.

Social/Agonistic Behavior

Evaluations of male or female same-sex social behaviors generally show no effect of postnatal (PND1-4 or PND11-14) chlorpyrifos exposures (Ricceri, et al., 2003; Venerosi, et al., 2006), a finding that was recently confirmed (Venerosi, et al., 2008; Venerosi, et al., 2010). On the other hand, increased investigation of the stranger mouse was reported following gestation exposure (GD15-18)(Venerosi, et al., 2006), and increased solicitation behaviors were observed following late postnatal exposure (PND11-14) (Ricceri, et al., 2003). Thus, there are contradictory reports of changes in female social behavior, being either increased or not altered. The only study of male social behavior showed no effect. Earlier studies reported that male agonistic behaviors were consistently increased in mice exposed either postnatally (PND1-4, PND11-14) or gestationally (GD15-18); the lowest effective dose was 1 mg/kg/d administered during either postnatal period (Ricceri, et al., 2003; Ricceri, et al., 2006). None of the newer studies addressed male behaviors. These findings are summarized below (Table 8).

Summary

Overall, decreased aggressive behaviors of dams that had been exposed during their development are a common finding across several exposure periods, as are increased male agonistic behaviors. Social and/or maternal behaviors in females are less consistently or convincingly altered.

Table 8 Summary of Social/Interactive Behavior Outcomes in Male (M) and Female (F) Mice

Exposure/effect	Late gestation GD14-17, 15-18	Early postnatal PND1-4	Late postnatal PND11-14
Female:female social	Increased investigation, dose-response[3]	No effect[1]	Increased solicitation, dose-response[1] No effect[3] No effect[4]
Male:male social	Increased aggressive postures, dose-response[2]	Increased agonistic behavior, differs across time-course, dose-response[1]	Increased solicitation, dose-response[1] Increased agonistic behavior, over time, no dose-response[1] Increased attack behaviors, dose-response[2] No effect social[4,a]
Induced maternal	No effect[2]		Increased maternalism, dose-response[2]
Natural maternal			Decreased maternalism[4]
Natural maternal:male	Decreased aggression[5]		Decreased defensiveness[4]

DR dose-response; if not mentioned, only one dose tested
[a] apparently only social, no agonistic, behaviors measured
[1] Ricceri et al., 2003
[2] Ricceri et al., 2006
[3] Venerosi et al., 2006
[4] Venerosi et al., 2008
[5] Venerosi et al., 2010

3.2.2.2.4 Motor activity

Most of the developmental studies of chlorpyrifos have evaluated motor activity of some sort; these were presented and summarized in detail in the 2008 SAP report. In general, activity levels have been recorded in terms of: 1) locomotion; 2) response latencies or other activity measures in the course of testing in various apparatuses (t-maze, radial arm maze, etc); and 3) habituation of activity over the session in activity chambers. In general, the earlier studies reported that activity is increased, decreased, or not altered, in either both or just one sex. Recent studies add to this literature, but none provide information to better explain these varied and contradictory outcomes.

Activity Devices

Evaluations of locomotor activity in open fields or other activity devices have been made in numerous studies across quite a few laboratories. Looking across developmental period and gender, there have been similar reports of increased, decreased, or no change in various measures of activity (*e.g.*, exploration, rearing, etc). Generalizations of effect could not be made in the 2008 preliminary review.

Two recent studies have used an open field to measure activity. Braquenier et al. (2010) reported no change of activity levels in female mice that had been exposed from late gestation through late lactation (GD15-LD14). While Zhang et al. (Zhang, et al., 2011) reported decreased activity in rats, those data cannot be evaluated at this time due to the language barrier.

Ancillary Activity Measures

As with activity measured specifically in activity devices, various measures of activity during other tests have shown increases, decreases, or no effect, in both or only one sex, with no obvious association to exposure period. It is important to consider, for example, that the time it takes to visit the arms of a radial arm maze is a somewhat different behavior than exploratory movement in a novel chamber. Thus, these activity measures may not be fully comparable, and the evaluations described here should be interpreted with caution.

Three recent papers reported assessments of activity during cognitive and social behavior testing. There was no change noted in postnatally exposed rats during radial arm maze testing (PND1-21) (Johnson, et al., 2009), or in gestationally exposed mice during foraging testing (GD17-20) (Haviland, et al., 2010). Mice exposed postnatally (PND11-14) showed decreased exploratory behaviors during the acclimatization phase of social testing (*i.e.*, before being exposed to another mouse); however, this was only significant in the first block of the test session (Venerosi, et al., 2008).

Habituation

Habituation, or decrease in activity level during the course of a test session, has only been specifically evaluated in the a few laboratories. As with other activity outcomes, the data are specific to exposure period and gender. Within the same laboratory, habituation is either faster (early gestation, both sexes) (Icenogle, et al., 2004), slower (late gestation, females only; late postnatal, both sexes) (Levin, et al., 2002; Levin, et al., 2001), or not altered (early postnatal) (Levin, et al., 2001). A perinatal study (GD6-LD10) also reported no change in habituation; however, the data were not statistically analyzed in the

same way, so results are not comparable (Maurissen et al., 2000). This measure has not been addressed in more recent studies.

Summary

As seen in Table 9, there remains to be inconsistencies in the various measures of motor activity, but it is important to note the numerous differences in procedures and apparatuses in which activity was measured. Given this, generalizable conclusions and summaries cannot be made at this time.

Table 9 Summary of Motor Activity Outcomes in Male (M) and Female (F) Rodents

	Early gestation GD9-12	Late gestation GD14-16; 14-17; 15-18; 17-20	Perinatal GD6-LD10; GD15-LD14	Early postnatal PND 1-4	Late postnatal PND11-14	Postnatal PND1-21
Activity chambers	No effect – rat, M&F[6]	Increased activity - mouse, only M tested, dose-response[13] No effect - mouse[8] No effect – rat, M&F[10]	No effect – rat, M&F[11] No effect – mouse, only F tested[2]	No effect - mouse, M&F[12] Decreased activity - rat, M not F[4] No effect – rat, M&F[9]	Increased activity - mouse, only M tested, dose-response[13] Increased activity - mouse, M&F, dose-response[12] Increased activity - rat, M&F[4] No effect – rat, M&F[9]	Decreased activity - rat, M&F, dose-response[3]
Activity measures	Increased activity - rat, M&F, dose-response[6]	No effect - mouse, only F tested[14] No effect - mouse, M&F[5] Increased activity - rat, M&F, DR[10] No effect - mouse, M&F[13]	No effect – mouse, only F tested[2]	Increased activity - rat, M not F[1] Increased activity - mouse, M&F[12] Decreased activity - rat, M not F[9]	Decreased activity - mouse, M&F[14] Decreased activity - rat, M not F[9] Increased activity - mouse, M&F[12] No effect – mouse, M&F[13]	No effect – rat, M&F[7]
Habituation rate	Increased habituation - rat, M&F, dose-response[6]	Decreased habituation - rat, F not M, dose-response[10] No effect – mouse[8]	No effect – rat, M&F[11]	No effect – rat, M&F[9]	Decreased habituation - rat, M&F[9]	

[1] Aldridge et al., 2005
[2] Braquenier et al., 2010
[3] Carr et al., 2001
[4] Dam et al., 2000
[5] Haviland et al., 2010
[6] Icenogle et al., 2004
[7] Johnson et al., 2009
[8] Laviola et al., 2006
[9] Levin et al., 2001
[10] Levin et al., 2002
[11] Maurissen et al., 2000
[12] Ricceri et al., 2003
[13] Ricceri et al., 2006
[14] Venerosi et al., 2008

3.2.2.3 Dose-Response Considerations

One of the key issues for the Agency is the degree to which points of departure based on cholinesterase inhibition are protective for the neurobehavioral outcomes reviewed in this chapter following developmental exposure. Few of these papers assess AChE inhibition at all, much less at time points potentially critical to the ultimate outcomes, *e.g.*, within hours after dosing or after the last of several repeated doses. The Agency and the 2008 SAP agreed that doses ≥1 mg/kg/d produce significant neurobehavioral changes following gestational and/or postnatal exposure(s) (FIFRA Scientific Advisory Panel (SAP), 2008a; U.S. Environmental Protection Agency, 2011). A dose of 1 mg/kg/d has been often used across studies and laboratories. Half of the papers published from 2008 on also used this dose, reporting significant effects on several different measures; this confirms the earlier conclusions.

A few studies have reported AChE inhibition when 1 mg/kg/d is administered directly to the pup postnatally (Dam, et al., 2000; Johnson, et al., 2009; Ricceri, et al., 2003). However, none of the neurobehavioral studies described here tested for fetal AChE inhibition when 1 mg/kg/d is given during gestation. A companion study to Maurissen et al. (2000) reported no cholinesterase inhibition in fetuses taken 4 hr after dosing to the dam when 1 mg/kg/d had been administered daily since GD6 (Mattsson, et al., 2000). Qiao et al. (2002) also reported no brain AChE inhibition in fetuses 24 hr after the last dose of 1 mg/kg/d on GD17-20. No other time points or days were assessed in either study. This suggests, but does not confirm, that the fetus would not experience AChE inhibition at 1 mg/kg/d, further suggesting that the behavioral effects reported in those studies were not due to AChE inhibition.

At the time of the 2008 SAP, there were only two studies in the literature (both in rats) that included a dose less than 1 mg/kg/d (D. A. Jett, et al., 2001; Maurissen, et al., 2000); a third study (in mice) has since been added (Braquenier, et al., 2010). In Maurissen et al. 2000, exposures of 0.3 mg/kg/d from GD6-LD10 produced no significant changes in the offspring across the numerous measurements (motor activity, startle response, delayed spatial alternation), whereas the highest dose (5 mg/kg/d) produced delayed growth and maturation. Female adults at both 1 and 5 mg/kg/d dose presented decreased parietal cortex thickness, yet this measure was not evaluated in the lowest dose group; therefore, conclusions about low-dose effects on that outcome cannot be made.

A different dosing paradigm was used by Jett and colleagues (2001), who dosed pups directly with 0.3 or 7 mg/k/g/d on PND7, 11, and 15. Rats were tested in the Morris water maze, starting on PND24, and the data suggested an effect in the 0.3 mg/kg/d dose group, in that latencies to find the platform were longer on the first day of training. With continued training, they were no longer different from control. While the authors state in the results that these latencies were “significantly greater” with p=0.05, the subsequent discussion describes this as a trend and that the group was “not significantly different” from control; thus, even the authors were not consistent in their conclusion. Figure 2 of Jett et al. (2001), however, shows that the means and standard errors for these groups do not overlap, suggesting that this may be a transient but real effect. The 2008 Panel was critical of this study with regards to the postweaning dosing regimen (not discussed here; see Section 3.2.2 for explanation) and the poor asymptotic performance of controls. It is critical to note, however, that these rats received a total of only six training trials, whereas at least twice that many trials are typically used for control animals to reach asymptotic performance. While the authors measured AChE inhibition, there were no changes even at the higher dose, 7 mg/kg/d. These findings are not in agreement with the 30% brain inhibition measured after a lower dose (5 mg/kg/d sc) on PND11 (Dam, et al., 2000), or with the number of studies

reporting brain inhibition at lower doses following acute oral dosing. Thus, uncertainty in the data for the high dose raises question about the AChE data altogether.

Braquenier et al. (2010) dosed mice at 0.2, 1, or 5 mg/kg/d from GD15-PND14. The only significant findings were increased anxiety in the 1 mg/kg/d dose group only. This lack of dose-response was seen using two different apparatuses, and testing different mice (littermates). The authors do not provide a discussion or explanation for this aberrant finding. The authors report ~14% brain AChE inhibition in the high dose group (only measured on PND1), and imply that the lower doses (not measured) would not produce any brain inhibition. Given uncertainty of no effects at 0.3 mg/kg/d (in rat), these new data suggest that 0.2 mg/kg/d, administered to mice for several weeks covering late gestation and early postnatal development, is a no-effect level. Notably, this no-effect level is similar to the acute point of departure and is approximately 10-fold higher than the repeated dosing point of departure.

3.2.2.4 Conclusions

These studies report a range of neurobehavioral changes in rats and mice following developmental exposure to chlorpyrifos. In this ongoing area of research, more studies are being generated in even more laboratories, all of which report some form of neurobehavioral alteration. Obvious species differences have not emerged, and effective doses are similar (1-6 mg/kg/d). Changes in various aspects of cognitive tests indicate perturbations of learning and/or memory, even though in some cases these may be manifest as improved function. Likewise, alterations in domains such as anxiety and social interactions may differ in direction of change, but are still suggestive of impacts on normal neuronal processing. There is replication of some effects across studies, and with some of the newer papers, across laboratories as well. Activity measures, on the other hand, still provide results as varied as the different measures of assessment. Taken together, these data do not provide evidence for a specific profile of effects but instead suggest more global alterations in neurobehavioral function.

All testing reported herein was conducted after weaning, and there is a presumption that the effects are permanent; however, no study has directly addressed this issue, and there is a range in test ages. Dose-response is not always evident, since many studies only use one dose, and of those using two or more doses, there is not always a monotonic response. Furthermore, the summary presented herein combines studies of different dosing regimens. While there are demonstrated differences in uptake and persistence of chlorpyrifos given subcutaneously vs. oral, with different oils or DMSO as vehicle, the developmental literature does not provide obvious differences in outcome based on this. Likewise, the literature has not shown that any specific developmental period is critical overall to the long-term outcomes, since similar effects are shown with different exposure periods. For example, cognitive changes in the radial arm maze were observed following gestational and early postnatal (PND1-4), but not late (PND11-14), exposure (Aldridge, Levin, et al., 2005; Icenogle, et al., 2004; Levin, et al., 2002; Levin, et al., 2001). However, cognitive deficits were reported with the Morris water maze following both gestational and late postnatal exposures (Billauer-Haimovitch, et al., 2009; D. A. Jett, et al., 2001; Turgeman, et al., 2011). Likewise, some changes in anxiety and social behaviors were reported at both gestational and postnatal exposure periods. Overall, these data do not clearly show specific critical periods of exposure, or definitive sensitive behavioral outcomes. Unfortunately, no laboratory has provided systematic comparisons across exposure period, dosing regimen, and age of testing; such studies would improve understanding of the impact of these critical factors.

These studies have almost exclusively focused on doses that could produce some degree, however minimal, of AChE inhibition. Thus it is not possible to know whether effects would be present at lower

doses, since they have not been adequately studied; thus far, only one study (Braquenier, et al., 2010) has tested a dose lower than the point of departure. The broad profile of neurological effects that have been reported do not aid in the development of a specific AOP, and as described in section 3.2.1., existing experimental studies have not been designed to examine and track possible mechanisms from early initiating events to the final neurological outcome. Such studies represent longer term research efforts by the different laboratories.

4.0 Review of Chlorpyrifos Epidemiology Regarding Children's Health

4.1 Scope and Purpose

In September 2008, EPA presented to the FIFRA SAP its preliminary review of available epidemiologic investigations of prenatal exposure to chlorpyrifos in association with measures of fetal growth and adverse neurodevelopmental effects in three major prospective children's health cohorts in the U.S.[18] These are: 1) The Mother's and Newborn Study of North Manhattan and South Bronx performed by Columbia University researchers referred in this document as "Columbia Mother's and Newborn Study;" 2) the Mt. Sinai Inner-City Toxicants, Child Growth and Development Study or the "Mt. Sinai child growth and development study;" and, 3) Center for Health Assessment of Mother's and Children of Salinas Valley (CHAMACOS) conducted by researchers at University of California Berkeley, the "CHAMACOS study." In this meeting, EPA updates and expands its targeted evaluation of this important line of evidence regarding chlorpyrifos developmental neurotoxicity.

At the previous meeting, the Panel agreed with EPA's conclusions that "chlorpyrifos likely played a role in the birth and developmental outcomes noted in the three cohort studies" (pp. 37 Meeting Minutes). In support of this statement, the Panel offered that investigations performed within these three epidemiological cohorts utilized a similarly strong study design (prospective cohort); measured exposure using several different methods including specific and non-specific biomarkers of chlorpyrifos; ascertained developmental outcomes using validated assessment tools common to both clinical and research settings; and, analyzed, selected and statistically adjusted for potentially confounding variables using reasonable and appropriate methods. Overall, the Panel noted that the epidemiological database at that time presented an informative body of evidence with some notable consistencies across studies. Areas of inconsistency were also observed, and judged, in part, to be due to differing methods of measurement and evaluation, as well as dissimilar exposure profiles (*i.e.*, residential versus direct and indirect occupational (farm laborer) exposure). Importantly, the Panel at the 2008 FIFRA SAP meeting also stated that it could not conclude that chlorpyrifos was the sole contributor to these outcomes, as co-exposure to other organophosphate pesticides and mixtures of environmental exposures may also have played a part in these outcomes.

For the purpose of informing the chlorpyrifos risk assessment, the Panel concurred with the Agency with respect to the primacy of the Columbia Mother's and Newborn Study among the three birth cohort studies, although the Panel also encouraged the Agency to comprehensively consider the results of the three children's health cohorts. While the Columbia Mother's and Newborn Study researchers measured the parent compound chlorpyrifos, as opposed to non-specific organophosphate metabolites, the other cohorts examined health outcomes (the Brazelton index of neonatal development) and performed supplemental analyses (*e.g.,* effect modification by PON1 status) not reflected in the Columbia Mother's and Newborn Study database. Therefore, the Panel expected EPA could strengthen its understanding of the potential developmental neurotoxicity of chlorpyrifos by considering the three children's health cohort studies together. Additionally, the Panel in 2008 suggested supplemental statistical analyses to enhance understanding of epidemiological study results in the risk assessment context (See Appendix 4). The Panel also generally noted both strengths and limitations of these studies, and offered that random

[18] See Meeting Minutes at:
http://www.epa.gov/scipoly/sap/meetings/2008/september/sap0908report.pdf.

or systematic errors in the design, conduct or analysis of these studies were unlikely to fully explain observed associations. However, the Panel also noted that absent the available toxicological and epidemiological databases was a well understood and defined mode of action as to the role of chlorpyrifos exposure in the etiology of adverse infant and child neurodevelopmental outcomes.

At this time, EPA expands and updates its review of the available epidemiologic data concerning the effect of chlorpyrifos exposure on children's environmental health in conjunction with a review of recent experimental studies and hypothesized adverse outcome pathways (AOP) (See Section 2.0). Observational studies published subsequent to the September 2008 FIFRA SAP evaluation extend the knowledge base of potential long-term sequalae of prenatal chlorpyrifos exposure. Specifically, in April 2011, researchers with each of the three prospective children's health cohort studies concurrently published results of their respective investigations of prenatal chlorpyrifos exposure and measures of intelligence among school aged children approximately 7-years (Bouchard et al., 2011; Engel et al., 2011; V. Rauh et al., 2011). Additionally, researchers with the Mt. Sinai Child Growth and Development study contributed their evaluation of organophosphate exposure and mental and psychomotor development, and authors with the CHAMACOS study published an evaluation of chlorpyrifos exposure and both fetal growth and neurodevelopment measures in young children as modified by paraoxonase-1 (*PON1*) genotype and phenotype (Eskenazi et al., 2010; Harley et al., 2011).

Researchers have also performed epidemiologic methods research which in many ways reduces uncertainties related to key measures within these studies. Within the Columbia Mothers and Newborn study, investigators published results of analyses evaluating the validity of prenatal chlorpyrifos exposure measures in the time periods immediately after the voluntary cancellation of chlorpyrifos for residential use (Whyatt et al., 2009; Whyatt et al., 2007), as well as employed innovative statistical techniques to further assess the potential confounding bias of socio-economic status (SES) in the relation between prenatal chlorpyrifos exposure and adverse neurodevelopmental health outcomes (Lovasi et al., 2011). Overall, these additions to the epidemiologic database concerning children's health effects in combination with other lines of evidence as discussed within this document (see sections 2, 3, and 5), add to the body of knowledge available to inform the ways in which chlorpyrifos exposure may be related to adverse neurodevelopment outcomes in children.

4.2 Summary of Epidemiology Findings

In this section, EPA summarizes and critically reviews epidemiologic studies of prenatal chlorpyrifos exposure and subsequent fetal growth and child development evaluated within the three prospective children's health cohorts described above. Specifically, in this section the design, conduct and methods of analysis of each cohort study are briefly presented; individual study results are summarized by type of health outcome investigated; and strengths and limitations of these investigations are discussed. Appendix 5 includes evidence tables summarizing details of each investigation, and Appendix 6 includes detailed study reviews and critical analysis of each investigation. The following section 4.3 reflects EPA's synthesis and evaluation of the current chlorpyrifos epidemiology database. In accordance with the draft "Framework for Incorporating Epidemiology in Risk Assessment,"[19] this analysis considers the strengths and limitations reflected in each cohort and research study as well as modified Bradford Hill considerations for the synthesis of these data.

[19] http://www.epa.gov/scipoly/sap/meetings/2010/020210meeting.html#materials

4.2.1 Overview of Design and Methods of Children's Health Studies

These cohorts were recruited for the purpose of studying the potential health effects of environmental exposures during pregnancy on subsequent child development: The Columbia University's Mother's and Newborn Cohort (Lovasi et al., 2011; Rauh et al., 2011; Rauh et al., 2006; Whyatt et al., 2009; Whyatt et al., 2007; Whyatt et al., 2004); The Mount Sinai Hospital Children's Environmental Health Cohort (Berkowitz et al., 2004; Engel et al., 2007; Engel et al., 2011); and UC Berkeley's the Center for the Health Assessment of Mothers and Children of Salinas (CHAMACOS) Cohort (Bouchard et al., 2011; Eskenazi et al., 2004; Eskenazi et al., 2010; Eskenazi et al., 2007; Harley et al., 2011; Marks et al., 2010; Young et al., 2005). These studies enrolled pregnant women at baseline and prospectively assessed associations in their newborns and young children. Multiple reports have been published based on the findings in each cohort.

The three study populations reflect different exposure profiles. The Columbia Mother's and Newborn study and the Mt. Sinai Child Growth and Development study participants were likely exposed to chlorpyrifos through residential use of the pesticide for indoor pest control. In the residential setting, chlorpyrifos was among the most widely used household pesticides in the US during the time period of these epidemiologic investigations. However, pesticide companies voluntary cancelled indoor residential uses of chlorpyrifos-containing pesticide products on December 31, 2000, during the time period of these cohort investigations. In the agricultural setting, chlorpyrifos was registered for use on commonly consumed crops such as corn, almonds, apples and oranges. Therefore, these study populations were most likely additionally exposed to chlorpyrifos via the oral route through ingesting residues in the diet and from hand-to-mouth contact with in-home chlorpyrifos-contaminated surfaces, as well as possible dermal exposure through contact with treated areas in the home environment (Whyatt et al., 2003; Whyatt et al., 2009; Whyatt et al., 2007). In contrast, CHAMACOS cohort participants were employed as farm laborers or were residing in homes with farm laborers. These participants likely experienced either occupational exposure through the inhalation and dermal routes, as well as probable indirect exposure through drinking water and take-home exposures (Bradman et al., 2007). In each of the children's health cohorts, the prevalence of pesticide exposure was high; however, reported use of chlorpyrifos in the CHAMACOS region was modest (10% of total pesticide use) (Eskenazi et al., 2004).

4.2.1.1 The Mother's and Newborns Study of North Manhattan and South Bronx (Columbia University)

Researchers with the Columbia Mother's and Newborn Study evaluated the association between prenatal exposure to pesticides including chlorpyrifos and developmental outcomes in children through age 7 years. In this birth cohort study, participants were recruited during early pregnancy (≤20th week) among African-American and Dominican women age 18-35 years, and registered for prenatal care and delivery at either New York Presbyterian Medical Center or Harlem City hospitals. Women who smoked, had a history of drug abuse, diabetes, hypertension, or HIV infection were excluded from participation in the study, as were women who resided in New York City for less than 1 year. The study samples represented in the reports reviewed were recruited between 1998 and 2004, a period which overlaps the voluntary cancellation of chlorpyrifos use in the residential environment.

In this cohort, authors measured chlorpyrifos exposure in several different biological and environmental matrices. These include chlorpyrifos parent compound in cord blood; the chlorpyrifos metabolite 3,5,6-TCPy in maternal and infant urine and meconium; and chlorpyrifos in personal and stationary air monitoring samples. In epidemiologic analyses, investigators consistently utilized cord blood measures

of chlorpyrifos as the measure of prenatal exposure. Chlorpyrifos levels in umbilical cord blood samples were sampled as close to the time of delivery as possible, and within 2 days post-partum. Cord blood plasma chlorpyrifos levels were imputed from maternal blood levels for newborns for whom no cord blood sample was obtained because correlation was high (>80%). Quantification of chlorpyrifos levels in plasma were conducted by the Centers for Disease Control and Prevention (CDC). Information regarding basic demographics, socio-economic status, and pregnancy related measures, among other factors was collected through self-report questionnaire at the time of enrollment.

4.2.1.2 Inner-City Toxicants, Child Growth and Development Study (Mt. Sinai Hospital)

Researchers with the Mt. Sinai Children's Health and Development study conducted a prospective birth cohort study in which they enrolled primiparous women presenting for prenatal care with singleton pregnancies at the Mount Sinai prenatal clinic and two private practices and who delivered their infants at Mount Sinai Hospital in New York City between May 1998 and July 2001. Mothers were excluded if they had any of the following characteristics: an initial prenatal visit after 26 weeks of gestation, serious chronic diseases, a serious pregnancy complication that could affect fetal growth and development. Additional, participants were excluded for risky health behaviors including alcohol consumed greater than two alcoholic beverages per day or illicit drug use. Mothers and infants were also excluded if the child was born with a congenital malformation or severe prematurity.

To measure prenatal pesticide exposure, researchers implemented a self-report questionnaire to solicit information regarding pesticide usage, in-home pest pressure and other exposure characteristics. In addition, in the early third trimester, participants were asked to provide a urine sample at the time of a routine clinical blood draw. Using this biological sample, authors measured urinary concentration of pesticide metabolites including both TCPy (Berkowitz et al., 2004), and also non-specific measures of organophosphate exposure, DAPs (Engel et al., 2007; Engel et al., 2011). Authors adapted analytical methods to conduct laboratory measurement of TPCy; DAPs were measured by CDC using published methods (Barr et al., 2002). Using maternal and infant (cord blood) blood sample, researchers measured PON1 enzymatic activity levels, and conducted genotyping analysis to determine prevalence of *PON1* variant alleles.

Potentially confounding variables were measured through self-report questionnaire and included in final statistical models if variables were known to be associated with either pesticide exposure or fetal growth. Authors also measured concentration of other pesticides in biological matrices including metabolites of pyrethroid exposure (PBA), pentachlorophenol (Berkowitz et al., 2004), as well as other organophosphates (malathion) and organochlorines compounds including polychlorinated biphenyls (PCBs) (Engel et al., 2007). However, authors did not present analyses concerning potential confounding effect of co-exposure to other environmental chemicals.

4.2.1.3 Center for Health Assessment of Mother's and Children of Salinas Valley, CHAMCOS (University of California/Berkley)

The Center for the Health Assessment of Mothers and Children of Salinas (CHAMACOS) cohort is comprised by participants who live and work in the Salinas Valley, CA. The Salinas Valley is a major center of agricultural production in the United States with approximately 500,000 pounds of organophosphate pesticides applied annually (California EPA. Pesticide Use Reporting 2001 Summary Data, 2002. (www.cdpr.ca.gov/docs/pur/pur01rep/01_pur.htm)), chlorpyrifos, however, was not

frequently used in agriculture in this area during the time period of this study. This cohort is comprised of low-income, predominantly Mexican-American (or Mexican immigrant) women employed as farm laborers or living with someone employed as a farm laborer. Enrollment of the Center for the Health Assessment of Mothers and Children of Salinas (CHAMACOS) cohort took place at regional community clinics. Women considered eligible for the study were less than 20 weeks gestation, aged 18 years or older, Medi-Cal eligible, fluent in English and/or Spanish, and planning to deliver at Natividad Medical Center. Excluded from analyses were women with gestational or preexisting diabetes, hypertension, twin births, or stillbirths.

Data collection was performed using several different tools. Authors administered self-report questionnaires to study participants to obtain information regarding demographic characteristics, work history and health behaviors. To assess exposure, participants were asked to provide biological samples (urine, blood). Specifically, maternal and fetal exposure to organophosphate pesticides was assessed by measurement of organophosphate DAPs and seven different pesticide-specific metabolites, including TCPy, in maternal urine during two periods in the pregnancy. Maternal urine samples collected between 5 and 27 weeks gestation and again between 18 and 39 weeks. The post-delivery urines were collected within 1 week of delivery for 73% of the sample, with the remainder obtained up to 176 days afterwards. Total DAP, including DMP, and DEP levels were determined for each participant for each of the two pregnancy urine samples, and because the measures did not differ substantially, these values were averaged in epidemiologic analyses to estimate prenatal pesticide exposure. Quantification of organophosphate metabolites was conducted by the Centers for Disease Control and Prevention (CDC) labs (Barr et al., 2002).

In this cohort, authors also measured ChE in whole blood and BuChE in plasma as a surrogate for OP exposure. PON1 was also measured in blood. Researchers also measured concentration of other environmental chemicals including PCBs, lead, DDT/DDE, HCB, and PBDEs; however, these compounds were not included in final models as potential confounding variables.

4.2.2 Summary of Research Results

Across these three children's health cohorts, authors have assessed the relation between measures of prenatal chlorpyrifos exposure and various measures of fetal growth and infant and child neurodevelopment. In this section, research results are briefly summarized by health outcome category. An evidence table summarizing key study features is presented in Appendix 5. A more detailed description of the study design, methods and analysis, as well as research results and a critique of individual study strengths and limitations are presented in Appendix 6. Table 10 briefly summarizes these results.

4.2.2.1 Measures of Fetal Growth

Authors with each of the three respective children's health cohorts measured prenatal chlorpyrifos exposure in association with fetal growth including birth weight, birth length, head circumference, gestational age and Ponderal index. To ascertain birth characteristics, authors linked with medical records at respective participating hospitals. Across these children's health cohorts, authors observed inconsistent evidence of an association; however, differing exposure profiles as well as dissimilar methods of prenatal chlorpyrifos exposure assessment likely played a role in this observation (Needham, 2005).

Table 10 Summary of Findings in the Columbia Mothers and Newborn Cohort, the Mount Sinai Hospital Children's Environmental Health Cohort, and the UC Berkeley Center for the Health Assessment of Mothers and Children of Salinas Studies of Prenatal Organophosphate Pesticide Exposure and Child Development.

	Columbia Cohort	Mount Sinai Cohort		UC Berkeley Cohort	
Markers of Exposure:	Chlorpyrifos	TCPy (fetal growth)	DAPS (BNBAS, Bayley, Wechsler)	TCPy (fetal growth, Bayley);	DAPS (BNBAS, Bayley, Wechsler)
Birth Length	Inverse (Null post 2000)	Null		Positive (NS)	
Birth Weight	Inverse (Null post 2000)	Null		Positive (NS)	Positive*
Head Circumference at Birth	Null	Null Inverse*		Null	Positive*
Gestational Age	--	Null		Inverse*	
BNBAS Newborn (Abnormal reflexes)	--		Positive*	Positive	
BNBAS Newborn (Neurodevelopment)	--		Null	Null	
Bayley Scores 6 months (MDI/PDI)	--		--	Null/Inverse (NS)	Inverse (NS)/Inverse (NS)
Bayley Scores 12 months (MDI/PDI)	Null / Null		Positive** (among blacks and Hispanics) / Positive (NS)		Null / Null
Bayley Scores 24 months (MDI/PDI)	Null / Null		Inverse* /Null*	Null / Null	Inverse / Null
Bayley Scores 36 months (MDI/PDI)	Inverse/ Inverse		--		--
Pervasive Development Disorder (PDD)	Positive (36 mo.)		--		Positive (24 mo.)
Mental Development (WPPSI-III, age 6 years)	--		Null		--
Mental Development (WISC-IV, age 7-9 years)	Inverse (Full-scale IQ and Working Memory);Null (Others)		Inverse (NS) (Full scale IQ, perceptual reasoning, verbal comprehension, working memory and processing speed)		Inverse
Odds of ADHD/ attention and behavior problems at age 7 years.	--		--	Positive (NS 3 years) Positive (5 years)	

*Interaction observed between pesticide markers and PON1 activity or genotype
**Interaction observed between pesticide markers and race
NS=Not statistically significant
Inverse= Higher levels of exposure associated with adverse health outcomes (measurement value or score decreased)
Positive = Higher levels of exposure associated with adverse health outcome (measurement value or score increased)
Null = No association observed
MDI= Mental Development Index
PDI = Psychomotor Development Index

Researchers with the Columbia Mothers and Newborn study observed an association between decreased fetal growth and prenatal chlorpyrifos measures among 314 mother-infant pairs selected for this study (Whyatt et al., 2004). Controlling for potential confounders, for each log unit increase in cord plasma chlorpyrifos levels, birth weight decreased by 42.6 g (95% CI: –81.8 to –3.8) and birth length decreased by 0.24 cm (95% CI: –0.47 to –0.01). Whyatt et al. (2004) did not report an association with head circumference. Combined exposure to both chlorpyrifos and diazinon (adjusted for relative potency using US EPA cumulative risk assessment methods) were also significantly inversely associated with birth weight and length ($p < 0.05$). When births were stratified by time period prior to or after the voluntary cancellation of chlorpyrifos for residential use, researchers no longer observed evidence of an association ($p > 0.8$) among births which took place in the cancellation period (n=77). In supplementary analyses, authors replicated this analysis with additional post-cancellation era births (total n=193) and similarly did not observe evidence of an association with chlorpyrifos in the later time period (Whyatt & Rauh, 2011) (See Appendix 4). Overall, authors suggest that prenatal chlorpyrifos exposures may have impaired fetal growth among this inner-city, low income cohort. While suggestive of a conclusion that cessation of chlorpyrifos exposure explains the different associations with fetal growth before and after the period of the voluntary cancellation of chlorpyrifos, EPA notes that this finding may also be suggestive of a possible threshold effect, or it could also be due to a lack of statistical power to assess associations by time period.

Within the Mt. Sinai Child Growth and Development study, Berkowitz et al. (2004) assessed the association between prenatal chlorpyrifos exposure measured as urinary TCPy and subsequent risk of impaired fetal growth (Berkowitz et al., 2004). Authors also evaluated potential effect modification by *PON1* genotype and phenotype in the relation of interest. Among 404 births which took place between 1998 and 2002, authors found no statistically significant associations between fetal growth including birth weight, or birth length or head circumference and chlorpyrifos (estimated as TCPy) exposure. However, Berkowitz et al. (2004) did observe evidence of heterogeneity of effect by PON1 activity level. Specifically, researchers observed a small, but statistically significant reduction in head circumference among children of mothers with levels of chlorpyrifos above the limit of detection and also in the lowest tertile of PON1 activity (least able to metabolize exogenous exposures such as OP pesticides). In the subgroup of infants whose mothers had TCPy levels greater than the level of detection, those with low maternal PON1 had an average (SD) head circumference of 33.3cm (1.5cm) which was significantly smaller than those with medium (34.0cm (1.5cm)) and high (34.1cm (1.6cm)) maternal PON1 activity after adjusting for race/ethnicity, infant sex, and gestational age ($p = 0.014$), although the statistical interaction was not significant. Authors did not observe evidence of heterogeneity of effect by PON1 in the relation between other measures of birth outcomes including birth weight and birth length and pesticide exposure. Neither maternal *PON1* genetic polymorphisms nor infant paraoxonase levels were associated with reduced head size.

Within the CHAMACOS cohort, among 488 participants enrolled between 1999-2000 Eskenazi et al. (2004) did not observe a significant adverse relationship between fetal growth and any measure of *in utero* organophosphate pesticide exposure (Eskenazi et al., 2004). On the contrary, investigators reported positive associations between birth length and head circumference associated with non-specific organophosphate exposure measures (DAPs). Researchers observed decreases in gestational age associated with measures of *in utero* pesticide exposure: urinary DMP metabolites ($\beta = –0.41$ weeks per log10 unit increase; 95% CI: –0.75—0.02; $p = 0.02$), which reflects exposure to DMP organophosphate compounds such as malathion, but not chlorpyrifos. Authors did not observe an association using TCPy as a measure of chlorpyrifos exposure; however they did report an increased risk

of preterm delivery with decreasing cholinesterase concentrations, a third biomarker of organophosphate exposure. To identify "critical windows" of fetal development when exposure may have a greater impact, the authors analyzed the associations of outcomes and metabolite levels measured during moving 6-week windows of pregnancy (*e.g.*, 5–10 weeks, 6–11 weeks, 7–12 weeks) using a series of multiple regression analyses. No period of greater impact was observed in this largely null study.

In a follow-up analysis within this cohort, Harley et al. (2011) performed the same analysis however evaluating the potential effect modifying role of *PON1* genotype and phenotype in the relation between prenatal pesticide and chlorpyrifos exposure (DAP, TCPy, ChE) and fetal growth (Harley et al., 2011). In this study, infants' (but not mother's) *PON1* genotype and PON1 activity modified the association between gestational age and head circumference. Infants with the susceptible $PON1_{-108}$ TT genotype had shorter gestational age (beta =-0.5 weeks, 95%CI: -0.9, 0.0) and smaller head circumference (beta=-0.4 cm, 95% CI: -0.7, 0.0) than those without the susceptible genotype ($PON1_{-108}$ CC genotype). Infants' arylesterase and paraoxonase activity were positively associated with gestational age. Maternal DAP concentrations were associated with shorter gestational age among infants of the susceptible $PON1_{-108}$ TT genotype, although only the interaction between $PON1_{-108}$ genotype and DEP metabolite concentrations was statistically significant (p-value for interaction = 0.09). However, maternal DAP concentrations were associated with larger birth weight (p-value for interaction = 0.06) and head circumference (p-value for interaction =0.01) in infants with non-susceptible genotypes. The authors conclude that infants with certain *PON1* genotypes (*e.g.*, $PON1_{-108}$ TT) may be more susceptible to effects of *in utero* organophosphate pesticide exposure.

4.2.2.2 Brazelton Neonatal Neurological Functioning

Researchers with both the Mt. Sinai Child Growth and Development study and the CHAMACOS cohort evaluated neonatal neurological functioning in association with prenatal chlorpyrifos exposure. To measure indices of abnormal neonatal behavior and/or neurological integrity authors used outcome measures derived from the Brazelton Neonatal Behavioral Assessment Scale (BNBAS). The BNBAS includes 28 behavioral items and 18 primitive reflexes which assesses the infant across several different developmental areas. This tool was administered to infants within days after birth and before they left the hospital (2-5 days post-partum). Examinations were conducted by trained neonatologists in the hospital setting using similar environmental conditions. The Mt. Sinai Child Development study and the CHAMACOS cohort evaluated this outcome measure; researchers with the Columbia Mother's and Newborn study did not measure the relation.

Among the 438 infants eligible to participate in the Mt. Sinai evaluation, Engel et al. 2007 observed an association between generic (DAPS) biomarkers of prenatal organophosphate pesticides and an increased number of abnormal primitive reflexes which are considered a critical marker of neurologic integrity. Controlling for confounding factors, subjects with prenatal total DEP levels above the median (24.7 nmol/L) delivered infants who were 2.3 times more likely to have at least two abnormal reflexes (95% CI: 1.1, 5.0). Associations with other DAPs were also reported. Notably, no statistically significant adverse associations were found for any of the DAP metabolites and other domains of the Brazelton assessment tool in the author's primary analyses, *e.g.*, habituation, range of state, orientation. Authors also observed evidence of potential effect modification by PON1 activity level in the relation between DAPs and neonatal neurodevelopment. Specifically, in the first tertile of paraoxonase 1 expression (slowest metabolizers), the relative risks (RR) of having abnormal reflexes were significantly related to DAP and DMP levels; the risk estimate for DEP and the number of abnormal reflexes within the lowest

PON1 level was not significant. In the CHAMACOS cohort, Young et al. (2005) observed a statistically significant association between organophosphate pesticide exposure and the reflex cluster of the BNBAS (Young et al., 2005). The proportion of infants with more than three abnormal reflexes was 3-fold increased among those with prenatal DEP exposure (DEP: OR = 3.4, 95% CI = 1.2, 9.9). Similar to Engel et al. (2007), no other associations between prenatal DAPs and other aspects of the Brazelton assessment were noted (other neurodevelopmental domains) (Engel et al., 2007). No adverse associations were found between postnatal urinary metabolite levels and any of the developmental outcomes.

4.2.2.3 Bayley Scale of Mental and Psychomotor Development

Researchers across the three children's health cohorts utilized the Bayley Scales of Infant Development II (BSID-II) to generate a Mental Development Index (MDI) and a Psychomotor Development Index (PDI) to assess neurodevelopment in early childhood. As a complement to this measure of mental development and behavior, authors also used the 99-item Child Behavior Checklist (CBCL). In addition, because the quality of the home environment is a key determinant of child development, authors also used the Home Observation for Measurement of the Environment (HOME) instrument. This instrument collects data regarding the physical and interactive qualities of the child's home as a measure of mental stimulation and interactions. Results from each of these measurement tools were used to model the relation between prenatal chlorpyrifos exposure and adverse neurodevelopmental outcomes in infancy and toddlerhood (6-36 months of age).

Rauh et al. 2006 investigated Mental Development Index (MDI) and a Psychomotor Development Index (PDI) at 12, 24, and 36 months of age within the Columbia Mother's and Newborn Study. Children were categorized as having either high (>6.17pg/g) or low (≤6.17pg/g) prenatal exposure, using categories informed by results of the previous study on birth characteristics (Whyatt et al., 2004). Authors reported that the difference in MDI scores was "marginally significant" (p = .06) with the exposed group scoring an average of 3.3 points lower. When the same multivariate regression models were calculated regarding the PDI scores, none of the 12 or 24 month PDI scores showed significant effects, but the 36 month score was significantly related to chlorpyrifos exposure. Investigators also calculated estimates of adjusted risk for developmental delays (MDI and PDI) related to chlorpyrifos exposure and illustrated that before 36 months of age, delays were no more likely in the highly exposed group, but, at the 36 month milestone, the likelihood of highly exposed children developing mental delays were 2.4 times greater (95% CI: 1.12-5.08, p = .02) and motor delays were 4.9 times greater (95% CI: 1.78-13.72; p = .002) than those with lower prenatal exposure. Using general linear modeling (GLM), authors analyzed developmental trajectories of the effects and results were consistent with the 12, 24, and 36 month milestone analyses indicating that the age effects most significantly occurred at the later phase of the 3 year period. In supplemental analyses suggested by the 2008 FIFRA SAP, authors illustrated that diazinon was a strong confounding variable in this association (correlation with chlorpyrifos 0.63), increasing the magnitude chlorpyrifos risk estimate for MDI and PDI 50-200% in the same direction (away from the null) (Whyatt & Rauh, 2011).

Within the Mt. Sinai Children's Environmental Health study, authors administered the BSID-II to participating children at 12 and 24 months. Among 404 women originally enrolled between May, 1998 and July, 2001 (n = 404), children of these mothers returned for neurodevelopment assessments at ages 12 months (n = 200), 24 months (n = 276) of age. Using generalized linear models, authors found that prenatal total DAP metabolite level was associated with a decrement in mental development at 12 months among blacks and Hispanics children. The associations appeared to be strongest among children of mothers who carried the *PON1* Q192R QR/RR genotype. The authors concluded that their findings are

suggestive of an association between prenatal exposure to organophosphates and decrements in cognitive development, particularly perceptual reasoning, with evidence of effects beginning at 12 months and continuing through early childhood, with *PON1* being a potentially important susceptibility factor for these deleterious effects. The authors did not observe any effect modification by PON1 enzyme activity level. In general, associations observed during the 12-month follow-up were either attenuated or non-existent at the 24-month visit.

In the CHAMACOS cohort, Eskenazi et al 2007, the authors report on the relationship between prenatal and child urinary organophosphate metabolite levels and child neurodevelopment at ages 6, 12, and 24 months of age. Controlling for indicators such as the psychometrician conducting the assessment and the location of assessment, age at assessment, sex, duration of breast-feeding, HOME score, and household income, parity and indicator of maternal intelligence (PPVT score), authors observed that prenatal DAP levels were adversely associated with MDI, while early life DAP levels were positively associated with MDI. At 24 months of age, these associations reached statistical significance (per 10-fold increase in prenatal DAPs: β = –3.5 points; 95% CI: –6.6 to –0.5; child DAPs: β = 2.4 points; 95% CI: 0.5 to 4.2). In a subsequent study, investigators did not observe evidence of effect modification by *PON1* status in the relation between prenatal DAPs and child neurodevelopment as measured by the Bayley Scale (Eskenazi et al., 2010). Neither prenatal nor child DAPs were associated with PDI or CBCL attention problems. Both prenatal and postnatal DAPs were associated with risk of pervasive developmental disorder (per 10-fold increase in prenatal DAPs: OR = 2.3, p = 0.05; child DAPs OR = 1.7, p = 0.04). TCPy as a biomarker of chlorpyrifos exposure was not associated with any neurodevelopment outcome in this study.

4.2.2.4 Attention Problems

Also within the CHAMACOS cohort, Marks et al (2010) conducted a study to investigate the association between urinary DAP metabolites in pregnant women and their children, as a marker of organophosphate exposure, and attention-related outcomes among 348 children who had available data at 3.5 and/or 5 years and met inclusion criteria (Marks et al., 2010). Attention-related health outcomes were measured through maternal report of child behavior at 3.5 and 5 years of age; direct assessment of the child at 3.5 and 5 years; and by a psychometrician's report of the behavior of the child during testing at 5 years. To identify children whose behaviors were most suggestive of possible ADHD within the cohort, a composite ADHD variable was defined that combined the results of the maternal report (CBCL), child testing (K-CPT), and the psychometrician report (Hillside). In this study population, higher concentrations of organophosphate metabolites in the urine of pregnant women were associated with increased odds of attention problems and poorer attention scores in their children at age 5 years. Prenatal DAPs were non-significantly associated with maternal report of attention problems and ADHD at age 3.5 years but were significantly related at age 5 years (CBCL attention problems: β = 0.7 points; 95% CI: 0.2–1.2; ADHD: β = 1.3; 95% CI: 0.4–2.1). Prenatal DAPs were associated with scores on the K-CPT ADHD Confidence Index > 70th percentile (OR = 5.1; 95% CI: 1.7–15.7 and with a composite ADHD indicator of the various measures (OR = 3.5; 95% CI: 1.1–10.7). Some outcomes exhibited evidence of effect modification by sex, with associations found only among boys. Children's concurrent total DAP and DMP metabolite levels at 3.5 years and 5 years were unrelated to attention outcomes, and but child DEP concentrations at 5 years were adversely associated with the composite measure of attention (OR = 2.0; 95% CI:1.1–3.6). The results of this investigation by Marks et al. (2010) in the CHAMACOS cohort are suggestive of a detrimental association between prenatal organophosphate exposure, as measured by maternal urinary metabolite levels, and attentional difficulties at age 5 years using three different measures of this neurodevelopmental outcome.

4.2.2.5 Intelligence Measures

To measure intelligence among school aged children, authors from each of the three children's health cohorts used the Wechsler Intelligence Scale for Children, 4th edition (WISC-IV). The instrument measures four areas of mental functioning: the Verbal Comprehension Index, the Perceptual Reasoning Index, the Working Memory Index, and the Processing Speed Index. These indices are associated with, but distinct from, overall intelligence quotient (IQ) and are sensitive to cognitive deficits related to learning and working memory. A Full-Scale IQ score combines the four composite indices. A General Ability Index score is a summary score of general intelligence, similar to Full-Scale IQ, but excludes contributions from both Working Memory Index and Processing Speed Index. WISC-IV scores are standardized against U.S. population-based norms for English and Spanish-speaking children.

Rauh et al. (2011) evaluated the relationship between prenatal chlorpyrifos exposure and neurodevelopment among 265 of the cohort participants who had reached the age of 7 years and had a complete set of data including prenatal maternal interview data, prenatal chlorpyrifos marker levels from maternal and/or cord blood samples at delivery, postnatal covariates, and neurodevelopmental outcome data (Rauh et al., 2011). While models were developed using continuous measures of both prenatal chlorpyrifos exposure and Wechsler scores, for ease of interpretation, investigators reported that for each standard deviation increase in exposure (4.61pg/g) there is a 1.4% reduction in Full-Scale IQ and a 2.8% reduction in Working Memory.

In the Mt. Sinai Child Growth and Development study, Engel et al. 2011 used generalized linear models to analyze the relationship between total DEPs, total DMPs, total DAP metabolites measured in maternal urine during the third trimester, and subsequent cognitive/intelligence development evaluated at age 7 years. In this study, prenatal maternal DEP urinary metabolite concentrations were associated with slight decrements in FSIQ, Perceptual Reasoning, and Working Memory between the ages of 6 and 9 years. Among children of mothers with the susceptible *PON1* genotype, DAP and DMP urinary metabolite concentrations were associated with poorer scores on Perceptual Reasoning and FSIQ.

In the CHAMACOS cohort, Bouchard et al. (2011) observed evidence of an association between prenatal exposures to organophosphate pesticides as measured by urinary DAP metabolites in women during pregnancy, and decreased cognitive functioning in children at age 7 (Bouchard et al., 2011). Authors observed this finding using total DAP, DMP, and DEP metabolites to estimate pesticide exposure. Children in the highest quintile of maternal DAP concentrations had an average deficit of 7.0 IQ points compared with those in the lowest quintile. Authors reported the associations were linear with no threshold. Urinary DAP concentrations in childhood (postnatal) were not associated with cognitive scores in this cohort of children. Of note, the following known or suspected neurotoxicants were measured: polybrominated diphenyl ethers (PBDEs), polychlorinated biphenyls (PCBs), p,p'-dichlorodiphenyltrichlorethane (DDT), p,p'- dichlorodiphenyltrichlorethylene (DDE), and lead. Lead was measured in maternal blood at 26 weeks gestation, in cord blood for a subset of participants, and children's blood a 2 years of age. These other environmental exposures including blood lead concentration did not change the magnitude and direction of the associations observed between DAP/DEP and IQ measures among school-aged children.

4.2.3 Strength and Limitations of Children's Health Cohort Studies

There are several strengths reflected in the design, conduct and analysis decisions made by investigators with each of the three children's health studies, as well as some limitations to consider. Strengths of these studies include the prospective nature of these investigations, extensive data collection including several different measures of prenatal chlorpyrifos exposure which have been validated, and use of neurodevelopmental outcome ascertainment tools which have been validated in different populations and commonly used in both clinical and research settings, among other strengths noted in this section. Key limitations reflect the difficulty in measuring multi-dimensional characteristics such as socio-economic status accurately, the challenge of conducting environmental epidemiology studies in populations exposed to several different compounds and mixtures, and the ability to assess effect modification (*e.g.*, genetic variability) with precision and accuracy. Considering both the strengths and limitations of the respective investigations, EPA puts forward epidemiologic inference at the conclusion of this section.

Study authors from each of the respective long-term studies hypothesized a role for environmental exposures, including but not limited to pesticides generally and chlorpyrifos specifically, in the etiology of adverse fetal growth and neurodevelopmental outcomes. Study authors stated their motivation was the knowledge that these populations were highly exposed to pesticides and other environmental contaminants, and that prior research had linked *in utero* environmental exposure to adverse neurodevelopmental outcomes. Cohorts were each funded through the EPA and National Institute of Environmental Health Science (NIEHS) Children's Environmental Health and Disease Prevention Research Centers[20]. Cohort studies were comprised of several hundred mother-infant pairs (range: n=102 to n=488) and were likely sufficiently statistically powered to detect hypothesized main effects, but studies may have lacked statistical power to perform stratified analyses, *e.g.,* effect modification by genetic variant. Similarly, statistical methods utilized across these investigations were appropriate to the data collected and research question hypothesized, however statistical analyses were discussed in varying degrees of detail by separate study groups. Statistical model selection was generally parsimonious in nature, and investigators performed multiple sensitivity analyses (not always shown) within each study to further explore and eliminate alternative explanations for observed results, *e.g.*, confounding by blood lead, variable transformations. Concerning the main effects hypothesized, both design and analysis decisions made by researchers enhanced the probability of identifying an association, if an association exists. For the association under study, measurement errors likely occurred due to the challenge of estimating exposure during the critical window of development.

Selection criteria were clearly defined and appropriate across the three cohorts. Investigators limited participants to individuals within a similar racial and ethnic group characteristics, income and education level, and geographic area, and who reported low prevalence of risky health behaviors (alcohol use, smoking during pregnancy), and who did not report major medical co-morbidities which may have adversely affected fetal growth and development (*e.g.*, gestational diabetes). Therefore, authors sought to control through design (restriction) variability in the sample population to isolate the effect of chlorpyrifos exposure on fetal outcomes. However, the many selection criteria applied may have affected the generalizability of study results. The selected study population is narrowly defined and dissimilar to the general U.S. population across many demographic and exposure related characteristics.

[20] http://epa.gov/ncer/childrenscenters/

However, as detailed in Section 5.0, comparison with biomonitoring levels in the U.S. general population, exposure among these cohorts was greater, but within a similar range. In addition, the external validity of the study findings may be limited if the organophosphate pesticide exposure-fetal development association is modified by factors that are more, or less, prevalent in the study populations relative to the population(s) to which inference is being made, or to populations with a substantially different exposure range.

All three studies are prospective cohort studies in which exposure measurements were obtained prenatally and/or at delivery, and fetal growth and neurodevelopmental outcomes were assessed subsequently. This study design eliminates or reduces several potential sources of bias. This design eliminates temporal bias, *i.e.*, uncertainty as to whether exposure precedes the adverse outcome, and, and also reduces the effect of differential exposure measurement error (a type of information bias) as it is unlikely exposure measurement would be differentially assessed by (unmeasured, future) health outcome.

Researchers utilized similar tools to measure adverse health outcomes across the studies. The use of similar outcome measures across studies aids comparability of studies and assessment of the consistency of results. Neurodevelopmental outcome assessment tools utilized were common to both clinical and research settings and most have been validated in both English and Spanish speaking populations. While measures of fetal growth are somewhat objective to measure, neurodevelopmental outcomes measured within these studies are more difficult to assess. While administered consistently within these investigations, tests of mental development, cognition, psychomotor development, and intelligence tools are somewhat subjective in nature, and may be affected by child's anxiety (Bayley Scales) and influenced by cultural factors (Wechsler). However, the homogeneity of the study population may reduce the potential influence of a cultural bias or differences in assessment tool response based on external factors, and these are among the best (gold standard) of tools to assess these types of developmental issues. Authors employed appropriate quality assurance and control measures such as training of test administrators, periodic evaluation of adjudicators, consistent environments in which tests were administered, and in many instances included an indicator variable for test administrator in statistical analysis to adjust for slight differences in examiner effect. With the exception of the child behavior checklist (CBCL), errors in the measurement of health outcomes were likely non-differential in nature, leading to an attenuation of the risk estimates. Overall, across these studies researchers utilized the best available outcome measurement tools for neurodevelopment health effects, and implemented the evaluations in a consistent, standardized manner with trained health professionals and/or study staff.

Across the three children's health cohorts, study authors measured parent chlorpyrifos, TCPy and DEP to estimate chlorpyrifos and/or organophosphate exposure. Chlorpyrifos metabolites (TCPy and DAPs) are likely more accurate and objective indicators of organophosphate pesticide exposure than other exposure ascertainment methods such as self-report, but uncertainty remains as to the extent measurement of non-specific metabolites reflects chlorpyrifos exposure. TCPy is a metabolite specific to chlorpyrifos, but can also be produced as a result of chlorpyrifos-methyl exposure or environmental exposure to TCPy itself (Morgan et al., 2005; Wilson, Chuang, Lyu, Menton, & Morgan, 2003). Urinary DAP metabolites of organophosphorus insecticides are markers of exposure to organophosphate pesticides generally; chlorpyrifos and diazinon are metabolized to DEP, while other organophosphates are metabolized to DMP. For risk assessment purposes, it is difficult to infer chlorpyrifos effects specifically from urinary DAPs and to a lesser extent the TCPy metabolite. Different exposure measurements may in part explain inconsistencies across study findings, as noted by others (Needham,

2005). The three birth cohorts recruited participants who likely have chlorpyrifos exposures that are higher than the general population, and the exposures in these cohorts may also be more consistent over time. Inner-city populations report frequent use of pesticides, most applied at least once per week, during the time period of these studies (Whyatt et al., 2003; Whyatt et al., 2002). In New York City, location of two of the three children's health cohort studies, housing authority reported common use of chlorpyrifos for indoor pest control during the time period of these studies, prior to the time period of the voluntary cancellation of these uses. In the CHAMACOS cohort, chlorpyrifos metabolite (TCPy) was detected in 77% of urine samples (Bradman et al., 2005; Eskenazi et al., 2004). Despite being considered to have greater opportunities for exposure relative to the general population, the average levels of chlorpyrifos biomarkers in the three cohorts were generally comparable to the U.S. general population, which may indicate a difference between actual and measured exposure. Nevertheless, it is not clear to what extent the use of one or two biomarker measurements conducted in the reported studies reflects exposure(s) over critical windows of development during pregnancy. This remains an uncertainty within these studies. As previously mentioned, chlorpyrifos exposure was assessed by quantification of biomarkers primarily in one- or two-samples of maternal urine taken during the third trimester, and also, in one cohort, in a single maternal blood or cord blood sample obtained at the time of delivery. If the exposure is chronic, a biomarker measured at a single time point may provide a representative dosimeter, even if the toxicant has a short half-life, as is the case for chlorpyrifos and organophosphate pesticides generally. However, if pesticide exposures are sporadic or otherwise vary over short time scales, the biomarker measurement may not be representative of "usual" exposure or of the exposure during critical periods of fetal development. In the UC Berkeley CHAMACOS cohort (Eskenazi et al., 2004), prenatal urine was collected between 5 and 27 weeks gestation and again between 18 and 39 weeks. The within-person standard deviation in the DAP metabolites was approximately three times larger than the between-person standard deviation, and concluded that a single biomarker assessment, as was conducted in the other two cohorts, "may not accurately reflect exposures over the entire pregnancy."

This potential limitation was also assessed in two validation studies conducted by Whyatt et al within the Columbia Mother's and Newborn Study. The first of these studies (Whyatt et al., 2007) assessed within- and between-home variability in indoor-air insecticides over the final 2 months of pregnancy among a subset of participants. Authors observed little within-home variability and no significant difference in air concentrations within homes over time ($p \geq 0.2$); between-home variability accounted for 92% of the variance in the indoor air levels of chlorpyrifos. Indoor and maternal personal air insecticide levels were highly correlated ($r = 0.7–0.9$, $p < 0.001$). While this study provides some evidence that assessment of chlorpyrifos exposure at a single time point may be reasonable, the study took place during a period of rapid decreasing indoor use of chlorpyrifos, and may not reflect variability in indoor air concentration prior to the period of cancellation. In addition to inhalational exposure, participants were likely also exposed by ingesting residues in the diet and from hand-to-mouth contact with surfaces contaminated with the pesticide. If true, then high correlation of chlorpyrifos air exposure over time may not indicate a true consistency in exposure levels.

The variability in exposure measures and the validity of different biomarkers of exposure was evaluated in a second study (Whyatt et al., 2009) which evaluated trends over time in multiple biomarkers of prenatal chlorpyrifos exposure. Authors measured TCPy levels in repeat prenatal urine samples and determined they were positively, but only moderately correlated ($r = 0.23–0.56$), and within-subject variability exceeded between-subject variability (intraclass correlation coefficient = 0.43). This indicates that variability in individual exposure over time may be considerable. Indoor air levels explained only 19% of the variance in prenatal urine TCPy ($p = 0.001$) in this study. However, these researchers also

demonstrated the presence of TCPy in meconium; meconium is fecal matter accumulated in intestines of developing fetus from week 13 of gestation and released immediately prior to or within a few days after delivery. This matrix is considered to be an integrated measure of exogenous exposures to the fetus during the gestational period. Importantly, investigators reported a moderate and significant correlation between maternal blood and urine collected in the later part of the pregnancy period, infant cord blood collected at delivery, and meconium (Whyatt et al., 2009). As noted in earlier sections, the critical windows of effect for these specific outcomes is unknown at this time, and may span the period of early pregnancy through early childhood (Rice & Barone, 2000; Rodier, 2004). The correlation among several biomarkers of chlorpyrifos exposure reflecting different windows of exposure from week 13 through delivery (*e.g.*, correlation between meconium and infant cord blood r=0.33, p=0.01, n=56), suggests a one-time measure of exposure may accurately rank participants with respect to prenatal exposures, at least within a 2- to 3-fold level of variation as suggested by authors (Whyatt et al., 2009). Additionally, the correlation between the analytic measure of exposure and a measure reflective of exposure during a large proportion of the gestational period suggests the exposure measures may also reflect exposure during the several likely critical windows of development in the mid- to latter period of gestation. However, to the extent participants experienced “peak” exposures at key periods of development, this information would not likely be reflected in the one-time measure, and likely lead to non-differential measurement error.

Bias due to confounding occurs primarily when risk factors for the outcome are unequally distributed among exposure groups, but are not themselves caused by the exposure, and these factors are not controlled in either the design or the analysis of the study. The greatest concern is for unmeasured confounders, those variables not measured within the study that may be related to both the exposure and disease of interest but are unknown to the investigator. Residual confounding can also arise due to errors in the measurement or categorization of confounders, even after apparent adjustment for these factors in the study, *i.e.*, these are factors not measured well enough. Among the major potential unmeasured or poorly measured confounding variables in the chlorpyrifos and neurodevelopmental association are: 1) difficult to measure aspects of the social environment (*e.g.*, chronic stress, socioeconomic conditions), and 2) environmental co-exposures that may be determinants of neurodevelopment (*e.g.*, lead exposure, mercury exposure, air pollution, tobacco smoke exposure, maternal alcohol intake, and exposure to other pesticides). The selection of cohorts that are homogenous with respect to many demographic and social factors significantly reduces the potential for residual or unmeasured confounding factors related to socio-economic status to have influenced these results as they have been controlled through restriction (another method of adjustment). For example, socioeconomic status cannot confound the main association in a cohort of individuals living under the same socioeconomic condition, *i.e.*, there is little variation in the study population on this factor. However, close examination of descriptive tables in these publications indicates some degree of variability remains. Therefore, authors appropriately evaluated and included in final models several individual-level markers for socio-economic status such as education, income, and race. Additionally, within the Columbia Mother’s and Newborn study, authors performed new analyses using hierarchal regression techniques to model variability due to SES in the relation between prenatal chlorpyrifos exposure and mental and motor development (the Bayley Scale) (Lovasi et al., 2011). These techniques utilize individual- as well as group-level variables to model SES. In this research, authors did not observe a significant difference in risk estimates, indicating that the role of SES as a major confounder in the Columbia cohort is appropriately adjusted. However, others argue that the multi-dimensional nature of a characteristic such as SES can never be fully captured through commonly used variables, and suggest within a term such as SES there could be components that act as both positive and negative confounding

variables (Bellinger, 2011). However, at this time, there is no compelling evidence that lack of control of the confounding influence of SES significantly biased the reported risk estimates.

Additionally, authors have measured several other environmental chemicals and in some instances considered these other exposures as potential confounding variables (not always shown). Researchers collected information from participants regarding biomarkers of other organophosphates, environmental tobacco smoke (ETS), blood lead, PAH, methylmercury (Whyatt & Rauh, 2011), pyrethroids/pyrethrins, organochlorines such as DDT/DDE and hexachlorobenzene, PBDE, and polychlorinated biphenyls. While authors evaluated these environmental exposures in relation to both chlorpyrifos and outcomes, none were included as confounders in final models due to lack of statistical evidence of confounding. Within the Columbia Mothers and Newborn study, authors performed additional analyses suggested by the 2008 FIFRA SAP to evaluate the role of other organophosphate exposures, specifically diazinon and propoxur, in the relation between chlorpyrifos and both birth characteristics and mental and motor delays (Bayley scores) (Whyatt & Rauh, 2011). Chlorpyrifos and diazinon biomarkers were highly correlated in this cohort (r=63%). While the effect sizes reported for the reported relation between both birth length and also birth weight remained unchanged from published reports when diazinon was added to the model, significant differences were observed in the relation with neurodevelopmental functions (mental (MDI) an PDI (motor) scores). Specifically, authors reported a 50-200% increase in effect sizes with MDI and PDI, respectively, *i.e.*, observed adverse effects became more pronounced with diazinon added to the model (See Appendix 4). Limited evidence of confounding by propoxur was also observed, but the correlation between chlorpyrifos and propoxur was moderate (23%), the confounding effect was not significant. In addition, neither pre- nor post-natal blood lead levels or methyl mercury levels measured in cord blood were significantly correlated with chlorpyrifos, and was therefore not considered a confounding variable in the association of interest within the Columbia cohort (Whyatt & Rauh, 2011; Rauh et al., 2006).

However, uncertainties remain as to the role of other (unmeasured) environmental compounds as potential confounding variables in this association. None of the authors evaluated the possible effect modifying role of other environmental chemicals in the relation between chlorpyrifos and neurodevelopment, nor were studies suited to examining the effect of mixtures (simultaneous combined exposures at potentially relevant critical windows of development). New investigations are needed to address these hypotheses, if relevant. While the possibility of unknown, unmeasured positive confounding bias can never be completely excluded, given the evidence available it is unlikely this potential bias may entirely explain observed associations.

Selection bias occurs when study participants are either selected or lost to observation as a result of a third, unmeasured factor that is associated with both the exposure and outcome of interest. In these prospective cohort studies, selection bias is most likely induced not by selection into the study, but by selection out of the study, *i.e.* due to attrition of study participants and missing data. Because data are often not obtained on those that are lost to follow-up or for whom data is otherwise missing, it is difficult to determine whether or not, and to what extent, the observed associations are biased due these factors. The remedy is simple, in theory – follow the entire cohort and obtain all relevant data – but often difficult in practice, particularly for long-term cohort studies such as those reviewed here. Across these studies, the amount of missing data varied but was great in some studies. To address missingness in data, authors imputed values in several instances, and performed sensitivity analyses with and without imputed data to illustrate comparability in reported results. In addition, , authors were able to illustrate differences between those included and excluded were comparable on several major characteristics, but some were not evaluated such as blood lead levels. Therefore, while it is difficult to

ascertain the degree to which selection bias due to missing data may have influenced these study results, authors employed appropriate analytic tools to address missingness in data to the extent possible.

Information bias arises due to misclassification or error in the measurement of either the exposure or outcome of interest, or the accurate measurement of confounding variables. Qualities of the outcome ascertainment method, the exposure assessment method and the measurement of confounding variables were reviewed above. In summary, regarding exposure measurement error the true relevant exposure in these studies is prenatal chlorpyrifos to the developing fetus. Sources of exposure measurement errors include 1) the single measurement chlorpyrifos or its metabolites during the third trimester, 2) error arising from differences between measured biomarker levels and actual chlorpyrifos or chlorpyrifos oxon exposure, even at the one time point, 3) unmeasured time- and space- dependent patterns of chlorpyrifos exposure, 4) uncertainty regarding the critical period for chlorpyrifos effects on development, 5) missing exposure data, 6) laboratory errors, and 7) imputation of missing exposure levels. Measurement errors in the ascertainment of chlorpyrifos exposure are likely to have occurred in the studies reviewed. However, because of the prospective study designs employed, the errors are unlikely to result in falsely positive findings, because the probability and magnitude of these errors are likely to be independent of the outcome status of participants. Measurement errors in the ascertainment of the health outcomes were also likely to have occurred, perhaps more for the neurocognitive outcomes, than for the indicators of fetal growth and development, and also are unlikely to have spuriously positive findings. These measurement errors are more likely to have resulted in false negative findings, if a causal association truly exists.

4.2.3.1 Epidemiologic Inference of Combined Children's Health Cohorts

As stated earlier, the three children's health cohorts considered herein have several strengths as well as limitations to consider in the interpretation of these studies. Within these studies, there are several factors that would tend to under-estimate the actual association (possibly leading to false negative association), as well as some characteristics that may lead to over-estimation (possibly leading to a false positive association). However, it must also be noted that methodological research and supplemental analyses (primarily within the Columbia Mothers and Newborn study) performed subsequent to the 2008 FIFRA SAP deliberations concerning these children's health cohort studies have reduced and further characterized important sources of uncertainty. As noted in the Introduction to this paper, currently scientists cannot determine with accuracy the critical window of exposure for these outcomes (early gestation through early childhood). Across the children's health cohorts, researchers used one or two measure(s) of exposure to estimate gestational exposure during the critical window of development, and investigators assessed the main exposure using non-specific biomarkers of chlorpyrifos. Undoubtedly, exposure measurement error occurred. However, exposure validation studies illustrate some degree of correlation across exposure measures made at different periods of gestation (*e.g.,* meconium, week 13 through delivery; maternal urine during last 8-weeks of pregnancy; and cord blood at delivery). Additionally, given the prospective nature of these studies, it is unlikely the measurement error was differential by outcome, *i.e.,* non-differential exposure misclassification leading to biased estimates toward the null is anticipated. Finally, as noted above the degree of missingness in some key variables across these studies may have resulted in a form of selection bias which may have also lead to an under-estimation of effects. However, given the data are missing, it is difficult to assess the magnitude and direction of this error on study results. Low sample size may have limited the ability of researcher to identify sources of effect modification or perform stratified analyses with accuracy.

Conversely, there may have been factors at play which led to an inflated estimate of the true association. Factor that may lead to false positive associations include unmeasured or poorly measured confounding factors which are positively associated with both chlorpyrifos use and adverse neurodevelopmental outcomes (positive confounding variables). The measurement of socio-economic factors and other environmental exposures experienced either pre- or post-natal environment are most likely among these potential factors, although others may exist. However, when additional adjustment was made for these factors using both individual- and group-level variables, the magnitude and direction of the associations remained stable (diazinon, propoxur, blood lead, methyl mercury, SES, post-natal exposure) (Lovasi et al., 2011; Rauh et al., 2006; Whyatt et al., 2004; Whyatt & Rauh, 2011). In the instances in which authors assessed the role of early childhood exposure to chlorpyrifos or other environmental contaminants as potentially confounding variables in the relation between prenatal exposure and neurodevelopmental outcomes, confounding bias was not observed. However, not all investigations evaluated the role of post-natal pesticide exposure, and authors note the limitation. Selection bias due to missing or drop outs from the study could also have influenced the observation of a false positive association, and this is again difficult to assess in the absence of data. However, researchers uniformly discussed and described the comparison of those included and excluded from individual analyses based in past of missing data using several important factors, and in many instances reported a level of comparability which is reassuring. The observation of positive associations as a result of multiple comparisons may also be a factor to consider; however the *a priori* identification of research questions of interest and the consistency of findings across several neurodevelopmental domains argues against a large role for multiple statistical testing to explain positive findings.

Overall, these are well performed studies which are shielded from several major sources of bias in the interpretation of results due to the strong design, conduct and analyses utilized in these investigations. While factors are present across these studies which may have led to either false positive or negative associations, it is notable that positive associations were observed as EPA believes the possibility of under-estimation of effect size is more likely than factors that would lead to over-estimation of effect size. Authors have taken significant steps to address major sources of uncertainty that may lead to over-estimation of effects, *i.e.*, positive confounding bias as a result of poorly measured factors related to socio-economic status and other environmental chemical exposures.

4.3 Chlorpyrifos Epidemiology Synthesis and Evaluation

In this chapter, EPA reviewed the results of epidemiological investigations of the association between prenatal chlorpyrifos exposure and adverse effects upon fetal growth and neonatal and early childhood neurodevelopmental outcomes across three major prospective children's health cohort studies in the U.S. In accordance with the OPP's draft "Framework for the Incorporation of Epidemiology into Regulatory Risk Assessment"[21], including the use of the modified Bradford Hill considerations in judging the potential causal nature of observed associations, in this section EPA considers the totality of the epidemiological evidence from these cohorts. To perform this analysis, EPA considered the strength of the associations observed and the presence of exposure-response trends, the temporality of the observed associations, and the degree to which alternative explanations have been considered and eliminated as explanatory factors, among other considerations. Issues of biological plausibility and specific mechanisms of action which may explain a causal role for chlorpyrifos in adverse

[21] http://www.epa.gov/scipoly/sap/meetings/2010/020210meeting.html#materials

neurodevelopmental outcomes, while broadly considered within the context of these investigations, are discussed in depth in accompanying chapters (See Sections 2, 3, 6).

As noted previously, each investigation is a prospective cohort study in which prenatal exposures occurred prior to the developmental of either fetal growth anomalies or neonatal or early childhood neurological delays. Therefore, in each study exposure preceded the health effect, risk may be directly calculated, and measurement error is more likely non-differential in nature, *i.e.*, likely under-estimation rather than over-estimation of risk effect. Table 10 summarizes the associations observed for chlorpyrifos markers and neurodevelopmental outcomes in the three cohorts. Within this database, moderately strong associations have been observed, and in many instances measured with precision. The relation between both neonatal neurological development as measured using the Brazelton index (number of abnormal reflexes) and also mental and psychomotor development among toddlers (Bayley Scale for 24-36 months) with prenatal chlorpyrifos exposure is elevated approximately 2- to 4- fold among those more highly exposed. This observation was consistent across cohorts with respect to the neonatal period, but not across measures using the Bayley Scale. Considering adverse effect in the 24-36 months period of development, the effects became more pronounced over time in one cohort (the Columbia Mother's and Newborn study), and became less pronounced over time in the Mt. Sinai Child Development study using non-specific biomarker of chlorpyrifos exposure (DAPs). However, it is notable the significant effects were seen when chlorpyrifos parent compound was measured directly; and, the effect size became significantly greater when diazinon was considered in the final statistical model. Notable decrements in intelligence measures were consistently observed across all three cohorts. While effect sizes measured as beta-coefficients are reflective of average change in intelligence score per unit change in chlorpyrifos exposure, and may appear modest in effect size, EPA notes that according to these models, the adverse influence at the extremes of the chlorpyrifos exposure distribution, if truly present in nature, would be more deleterious. Strong, consistent evidence of a positive association between prenatal chlorpyrifos and attentional problems, pervasive developmental disorder, and ADHD-like symptoms, although not measured with precision.

An association is consistent when a similar magnitude and direction of results are replicated in studies in different settings using different methods. If a relationship is causal, one may expect to observe it consistently in different studies and among different populations. Several consistencies were noted across these studies. Higher levels of the chlorpyrifos-specific metabolite TCPy were associated with prevalence of abnormal reflexes among newborns, as assessed using the Brazelton Neonatal Behavioral Assessment Scale (BNBAS), in both the Mount Sinai Hospital and UC Berkeley (CHAMACOS) Cohorts. The BNBAS assessment was not conducted in the Columbia Mothers and Newborns cohort. TCPy levels were consistently not associated with BNBAS indicators of newborn neurodevelopment such as behavioral domains other than those observed for number of abnormal reflexes. In addition, associations between pesticide markers and the Mental Development Index (MDI) portion of the Bayley Scores at 12, 24, and 36 months varied between small, largely non-statistically significant decrements, and null. However, in supplemental (post-publication) analyses, authors illustrated that adjustment for other organophosphate pesticides in the relation between prenatal chlorpyrifos and MDI strengthened the magnitude of the effect significantly at the 36 months time point (Whyatt & Rauh, 2011). Associations between early life organophosphate pesticide exposure biomarkers and decrements in mental development in childhood (age 7-9 years) were observed in all three studies, although the relationships were not consistently statistically significant.

Some notable inconsistencies were also observed. These may be explained by differing exposure biomarkers, timing of biomarker collection, or other limitations of study design and measurement, *e.g.,*

the magnitude of non-differential exposure misclassification. However, inconsistency in results may also argue against a true causal association. Associations between markers of prenatal pesticide exposure and fetal growth were not consistent across the three studies, as noted in the FIFRA SAP 2008 meeting. In the Mount Sinai Child Growth and Development cohort, no statistically significant associations were observed in primary analyses, although a modest decrease in average head circumference with increasing TCPy was noted among those with low PON1 activity. In the Columbia Mothers and Newborn Study (Whyatt et al., 2004), chlorpyrifos levels in maternal blood were modestly associated with decreased fetal growth. In contrast, among the UC Berkeley CHAMACOS study participants, modest increased birth weight and head circumference were associated with *in utero* DAP concentrations (Eskenazi et al., 2004; Harley et al., 2011). In fact, stratification by *PON1* status seemed to enhance these positive associations, such that in the non-susceptible groups (*i.e.*, $PON1_{-108}$ CC, $PON1_{192}$ RR, and high arylesterase activity), increasing DAP concentrations were significantly associated with increased birth weight and head circumference. The pesticide biomarkers differed between the two studies; cord blood chlorpyrifos levels were assessed in the Columbia Mothers and Newborn study, while DAPs were assessed in the UC Berkeley CHAMCOS cohort. It is possible that high urinary DAP concentrations may be an indication of rapid detoxification and excretion of organophosphate pesticides, rather than a marker of high exposure, particularly among the participants that are less potentially less susceptible to pesticide effects due to their *PON1* genotype or PON activity level, as suggested by study authors.

Although consistency of associations across studies set in different populations, and employing different methods has been used as a criterion for causality, it is not the only compelling factor. In fact, heterogeneity across studies is expected to occur between studies in the presence of effect modification, if the prevalence of the effect modifier varies between the populations being assessed. Apparent inconsistencies could also be due to chance differences, and due to the presence of biases operating in the studies under consideration.

There was evidence for an exposure-response relationship in several of the studies reviewed, whereby increasing levels of exposure were associated with increasingly large decrements in measures of neurodevelopment (Rauh et al., 2011; Rauh et al., 2006). For the analyses in which continuous distributions of exposure markers were used, the levels were often, but not always, log transformed prior to entry in statistical models. The estimated association between the biomarker level and the outcome in these cases is not linear; rather, the association between the log-transformed exposure and the outcome is linear. In such cases, results are interpretable as changes in mean level/risk of the outcome of interest for a given percentage increase in the exposure. Examination of departures from linearity of exposure-response relationships was reported in only a small subset of the reviewed articles. For example, Rauh et al. (2011) reported that, in the Columbia cohort, “the dose-effect relationships between CPF [chlorpyrifos] exposure and log-transformed Working Memory Index and Full-Scale IQ scores are linear across the range of exposures in the study population, with no evidence for a threshold” (Rauh et al., 2011). Departures from linearity were not statistically significant. Two possible explanations for this finding are that 1) the exposure response is linear on the scale assessed, or that 2) the studies did not have sufficient power to detect departures from linearity in the shape exposure-outcome relationship. The smoothed exposure-response curve superimposed upon the scatter plot of log-transformed working memory appears to be monotonically decreasing; however, a similar curve superimposed upon the scatter plot of log-transformed IQ score and chlorpyrifos levels appears flat if not slightly positive, before turning negative beyond an inflection point at about 5 pg/gram of chlorpyrifos. As such, it may be that both explanations for the null finding regarding departures from linearity are operating in this study. As another example, Bouchard et al used cubic splines to evaluate

the shape of dose-response curves, test the linearity assumption, and investigate potential thresholds (Bouchard et al., 2011). Again, no statistically significant departures from linearity were observed.

Due to the voluntary cancellation by registrants of indoor residential uses of chlorpyrifos-containing pesticide products, children born after the year 2000 were likely exposed to far lower levels of chlorpyrifos, on average, than children born prior to 2001. In their report, Whyatt et al. (2004) took advantage of this "natural experiment" by stratifying their analyses on birth date prior to, or after, January 1, 2001. Although reported household use of pesticides in general did not change over the same time period in this study, maternal chlorpyrifos in maternal air samples were significantly lower after the voluntary cancellation (4.9 ng/m^3), relative to those taken before 2001 (8 ng/m^3; $p<0001$). Associations between birth outcomes and organophosphate insecticide levels in maternal personal air samples were not statistically significant in both un-stratified analyses and among subgroups stratified by birth prior to or after January, 2001. However, the associations between birth weight and length and cord plasma chlorpyrifos were highly significant ($p \leq 0.007$) among newborns born before the 2001 cancellation of registrants. Among newborns born after January 2001, no association with fetal growth was observed ($p > 0.8$) (Whyatt et al., 2004). Supplemental analysis suggested this observation persisted among additional number of infants born after the period of the voluntary cancellation (n=193) (Whyatt & Rauh, 2011). This finding by Whyatt et al provides evidence that the association "can be altered (prevented or ameliorated) by an appropriate experimental regimen," as advocated by AB Hill in the assessment of causality in observational studies. Changes in exposure were reflected in decreasing chlorpyrifos levels, and appear to have been associated with changes in developmental outcomes. However, as noted by the 2008 FIFRA SAP Panel in their report, this study was not designed to test this hypothesis, specifically.

It must also be considered that given the many selection criteria applied within each of the three children's health cohorts, the generalizability of study results may have been affected. These studies reflect a somewhat narrowly defined sub-population who likely experienced a greater range of exposures and higher peak exposures than the general U.S. population; a comparison of maternal urinary concentration of TCPy in the three children's health cohorts with the general population (NHANES) illustrates higher exposures occurred among the cohort participants. The exception is with the urinary concentration data collected by Whyatt et al. (2009) which were lower than the U.S. general population; however, these data were collected 2001-2004, a period after the voluntary cancellation of indoor residential uses of chlorpyrifos (see Section 5.0). In addition, the external validity of the study findings may be limited if the organophosphate pesticide exposure-fetal development association is modified by factors that are more, or less, prevalent in the study populations relative to the population(s) to which inference is being made, or to populations with a substantially different exposure range. In the studies reviewed, the exploration of gene-by-environment, and phenotype-by-environment interactions is a strength of many of the analyses, although as noted above the studies are uniformly underpowered to assess such interactions. There was scant assessment of interaction between chlorpyrifos and other environmental co-exposures conducted in these investigations including blood lead levels.

4.4 Conclusions

The FIFRA SAP panel convened in September 2008 concluded that given the strengths of these studies, the effect of biases likely present, and rigorous statistical analyses performed, the observation of associations across these studies can be interpreted as a conclusion that chlorpyrifos likely played a role in the neurodevelopmental outcomes observed among children more highly exposed to chlorpyrifos in the *in utero* environment. Since that time, researchers with each of the three children's health cohorts have published several new etiologic and methodological studies and performed supplemental analyses as suggested by the 2008 Panel which extend the knowledge base, and reduce several major sources of uncertainty present in the 2008 database (*i.e.*, confounding by SES, exposure validation in the Columbia Mothers and Newborn study, further evaluation of *PON1* genotype and PON activity level). In the current document, EPA utilized the draft "Framework for Incorporating Epidemiology into Risk Assessment"[22], including use of modified Bradford Hill considerations, to assess the strengths and limitations of the studies in this database on the association of interest, and to clarify the epidemiologic evidence in support of a causal role for chlorpyrifos in infant and child adverse neurodevelopmental outcomes.

In EPA's analysis of the strengths and weaknesses of these studies, the chance that positive associations observed are false positives due to systematic errors in the studies cannot be excluded; however, it is more likely that error present in these studies would lead to the under-estimation of the true association. Therefore, while alternative explanations for positive association can be hypothesized (*e.g.*, additional unmeasured or poorly measured positive confounding variables), these explanations are judged to be less plausible than the alternative that associations have been missed or under-estimated due to non-differential measurement error and low sample size across exposure strata. In occupational settings, exposure measurement error has been shown to more greatly influence epidemiology study results than unknown or unmeasured confounding variables (Blair et al., 2011). Additionally, the elimination of temporal bias due to the prospective study design employed within each of the three children's health cohorts assures that prenatal exposures preceded neurodevelopmental outcomes measured at birth, and in early and later childhood through age 7 years. The strength of the associations measured in some studies was notably strong. However, associations in many instances were weak to moderate, possibly due to measurement error. Associations with neurodevelopmental outcomes were consistently identified with respect to the number of abnormal reflexes in the neonatal period, the presence of mental and behavioral issues as well as gross motor delays were pronounced especially in later toddler years of 24-36 months, and the observation of intelligence decrements were seen across the three cohorts using different measures of prenatal chlorpyrifos exposure, although not consistently statistically significant. EPA notes that other organophosphates may be involved as well as the CHAMACOS and Mt. Sinai Child Development study utilized DAP which measure exposure to several different organophosphates, and that use of chlorpyrifos in the CHAMACOS study region was low at the time of the study. However, links with DEP which result from either chlorpyrifos or diazinon exposure were observed as well. In general, given that the intended mode of action of chlorpyrifos toxicity is neurological dysfunction, *i.e.*, inhibition of acetyl cholinesterase, a biologically plausible role for chlorpyrifos in adverse neurodevelopmental outcomes in the developing brain can be posited. However, this issue is discussed in greater detail elsewhere in this paper.

In summary, while the strengths and limitations of the studies would be more likely to lead to an under-estimation of the true effect, the possibility of false positive associations cannot be entirely ruled out.

[22] http://www.epa.gov/scipoly/sap/meetings/2010/020210meeting.html#materials

The temporal association, strength of the statistical associations observed and presence of exposure response across some (but not all) of the investigations, as well as some notable consistencies in findings within this epidemiologic database tend to support a role for chlorpyrifos in this relation. It is true that the Columbia Mothers and Newborn study is the only cohort to have measured chlorpyrifos parent compound directly; replication in other cohorts that use this exposure metric would aid causal inference. The presence of factors which may have over-estimated effects, the lack of consistency in many neurodevelopmental domains, and the lack of a clear mechanism of action may argue against a true association. Overall, the current database supports the conclusion from 2008 Panel that "chlorpyrifos likely played a role in the birth and developmental outcomes noted in the three cohort studies." Additionally, the subsequent studies made available since the time of the 2008 evaluation helps to clarify the potential role for chlorpyrifos in adverse neurodevelopmental health effects.

5.0 Exposure Profile & Biomonitoring Research

In the previous Section, the Agency performed a targeted evaluation of recent epidemiologic results from the Columbia Mother's and Newborn Study, Mt. Sinai Child Growth and Development study, and CHAMACOS children's health cohorts. Each of these prospective cohorts utilized exposure assessment methodologies based on observed levels of chlorpyrifos or metabolites in environmental media and biological samples; however, only in the Columbia Mothers and Newborn study were etiologic analyses were all performed using biological markers of chlorpyrifos exposure.

In this section, EPA presents a comparison of biomonitoring studies involving the general population and specific sub-populations, including children, farm workers, and pregnant women. The purpose of the comparison is to establish a frame of reference to help characterize and evaluate the range of exposures to chlorpyrifos observed in each of the children's health cohorts. Specifically, TCPy biomonitoring results from the three children's health cohorts are compared with other biomonitoring results from other major observational exposure studies in the United States. Section 5.1 first provides an overview of the main chlorpyrifos biomarkers and important scientific considerations that should be evaluated when interpreting biomonitoring results. Section 5.2 then provides a comparison of the children's health cohorts with other biomonitoring studies involving pesticide applicators, farm families, children, and the general population, which expands the Agency's biomonitoring review presented during the September 2008 SAP on chlorpyrifos. Section 5.3 updates the Agency's previous review of the association between chlorpyrifos biomonitoring data and AChE levels. Finally, Section 5.4 discusses various approaches for interpreting biomonitoring data and provides a discussion of PBPK modeling efforts for chlorpyrifos.

5.1. Overview of Biomarkers and Scientific Considerations

A summary of biomarkers of chlorpyrifos exposure are summarized in Table 11 below. The biomarkers most widely monitored to evaluate exposure to chlorpyrifos are the urinary metabolites TCPy and the non-specific DAP metabolites DEP and DETP. In addition to these urinary biomarkers of exposure, chlorpyrifos can also be measured directly in blood and other biologic media.

Table 11 Common Chlorpyrifos Biomarkers of Exposure.

Biomarker/Analyte	Biologic Matrix	Population Group
3,5,6-trichloro-2-pyridinol (TCPy)	Urine	All
	Meconium	Pregnant Females, Neonates
Diethylphosphate (DEP)	Urine	All
Diethylthiophosphate (DETP)	Urine	All
Chlorpyrifos	Blood	All
	Cord Blood	Pregnant Females, Neonates

The National Academy of Sciences established a biomonitoring framework that recommends assessing biomonitoring data quality based on scientific and interpretive considerations that include: biomarker specificity; pharmacokinetics and intra-individual variability; and study design considerations (NAS, 2006). The remainder of this section provides an evaluation of these scientific and interpretative considerations with respect to the primary urinary and blood biomarkers that have been used to evaluate chlorpyrifos exposure in exposure studies and epidemiologic research. Several review articles

in the peer-reviewed literature have attempted to characterize the data quality of chlorpyrifos biomonitoring. Barr and Angerer (2006), in particular, provide a detailed review of chlorpyrifos biomarkers that discusses several of the considerations outlined by NAS. Similarly, Eaton et al. (2008) provides discussion of chlorpyrifos biomarkers in their extensive review of the toxicology of chlorpyrifos. Likewise, Egeghy et al. (2011) of the Agency's Office of Research and Development (ORD) reviewed pesticide urinary biomarkers from ORD observation exposure studies with an emphasis on pharmacokinetic factors.

Urinary Biomarkers of Exposure

As previously discussed in *Section 4*, TCPy, DEP, and DETP are commonly used as urinary biomarkers of chlorpyrifos to exposure in etiologic research. DEP and DETP are considered non-specific biomarkers because they are metabolites of multiple of OP pesticides, including chlorpyrifos-methyl, coumaphos, diazinon, disulfoton, ethion, parathion, phorate, sultotepp, and terbufos (CDC, 2009). As such, it is more difficult to attribute urinary levels of DEP and DETP to chlorpyrifos exposure without additional evaluation of exposure routes involving all potential OP parent compounds.

In contrast, the main parent compounds of TCPy are chlorpyrifos, chlorpyrifos-methyl, and trichlopyr, so urinary TCPy levels are attributable to exposure to fewer parent compounds that the DAP metabolites. For this reason, TCPy is generally considered a more specific biomarker of exposure than the DAP metabolites. However, studies have demonstrated that chlorpyrifos degrades into TCPy in environmental media and food. As such, urinary TCPy concentrations may reflect exposure to the parent compound chlorpyrifos and/or to its degradate, TCPy, in the environment. Morgan et al. (2005, 2011) and Wilson et al. (2003) have observed that TCPy concentrations were at least 10 times higher than chlorpyrifos concentrations in 48-hour duplicate diet samples (Eaton et al., 2008). Similarly, DEP, DETP, and the other non-specific DAPs have also been detected in the environmental media and foods. For example, Lu et al. (2005) observed that OP pesticides in fortified fruit juice samples degraded into DAPs. Based on these findings, and as noted by epidemiology study authors, use of urinary measurements of TCPy (or DEP and DETP) to assess chlorpyrifos exposure may overestimate chlorpyrifos exposure if it is assumed that exposure to the parent compound is the only source of the urinary biomarkers (Eaton et al., 2008). It is not clear, however, the extent to which environmental exposure to the metabolites contribute to urinary concentration of DAPs or TCPy. Thus it is important to consider the relative contribution of exposure to the parent chlorpyrifos compound and the environmental metabolite when interpreting epidemiology results.

Longitudinal variability is another important consideration when interpreting chlorpyrifos biomonitoring results. Studies evaluating chlorpyrifos exposure typically measure TCPy or DAP levels through collection of a single spot urine samples, 24-hour voids, or repeated samples over an extended period of time. Spot urine samples generally provide less reliable estimates of exposure if the goal of the study is to estimate the periodicity of exposure over an extended period of time (Barr and Angerer, 2006); however, if exposure is relatively constant, a spot urine sample may accurately reflect chronic exposure. Barr and Angerer (2006) suggest that previous studies indicate that spot-urine samples cannot accurately quantify pesticide exposure, but also indicate that more recent studies have shown that a single sample may be adequate for assigning broad exposure classifications in epidemiology studies. This conclusion was supported by methodological research performed by investigators with children's health cohort studies. Specifically, epidemiology investigators have illustrated some correlation among biomarkers reflecting short and long exposure durations, suggesting a one-time exposure measure may accurately reflect a longer period of exposure (Whyatt et al. 2009).

Blood Biomarkers of Exposure

Monitoring chlorpyrifos directly in blood can reduce the uncertainty associated with urinary biomarkers because blood measurement provides a more direct measure of internal dose that is specific to chlorpyrifos (Barr and Angerer, 2006). While monitoring of chlorpyrifos in blood can provide a better measure of exposure in some cases, blood measurement can place a greater burden on study participants and require more sensitive analytical methods. Additionally, chlorpyrifos has a short biological half-life (<27 hours), therefore, blood concentrations of chlorpyrifos may represent a much narrower exposure period relative to urinary concentrations (NAS, 2005). These challenges may explain why researchers have typically monitored TCPy and other urinary biomarkers to evaluate OP exposure. While collecting and monitoring blood samples for chlorpyrifos may be challenging, collection of umbilical cord blood can be more feasible because larger quantities of blood can be collected without invasive collection procedures (Barr and Angerer, 2006).

It appears that only a limited number of biomonitoring studies in the United States have measured chlorpyrifos in blood, perhaps for the reasons suggested by Barr and Angerer (2006). Two of these studies have previously been reviewed by the Agency during its 2008 SAP on chlorpyrifos (USEPA, 2008) and involved administration of a dose of chlorpyrifos to adult volunteers in order to examine the association between chlorpyrifos blood levels and AChE activity (Nolan et al., 1982; Kisicki et al., 1999).[23] Additionally, one study by Whyatt et al. (2003), also previously reviewed by the Agency, measured chlorpyrifos in both maternal and cord blood of the Columbia Mother's and Newborn study.

5.2 Chlorpyrifos Biomonitoring Study Comparison

This section presents a summary comparison of selected TCPy urinary biomonitoring studies in order to help characterize the range of potential exposures in the both the general U.S. population and study populations that have been targeted by researchers. The comparison focuses on urinary TCPy because it has been widely monitored in a range of different studies and has greater specificity to chlorpyrifos than the DAP analytes. Urinary TCPy concentrations are commonly reported on a volume and/or a creatinine-adjusted basis. Creatinine is a common correction method to adjust for variable dilutions in urine samples. However, there can be considerable variation in creatinine concentrations in urine amongst different age and ethnic populations, due partially to differences in lean muscle mass (Barr et al., 2005). For example, children generally have less muscle than active adults and African-Americans generally have more muscle mass than Caucasians. As a result, interpretation of creatinine-adjusted concentrations requires additional adjustment of typical creatinine urinary levels by age, sex, and race. Additionally, it has also been suggested that creatinine adjustment may not improve the interpretation of biomonitoring when urinary creatinine is highly variable (Kissel et al., 2005). For these above reasons, only TCPy concentrations on urine volume basis (µg/L) are presented in this section.

The studies selected are not the entire universe of all available biomonitoring studies. Rather, the majority of selected studies have previously been evaluated as part of biomonitoring review conducted as part of the Agency's 2008 SAP on the toxicity profile of chlorpyrifos (USEPA, 2008), reviewed by Eaton et al. (2008), or discussed in more detail in Section 4 of this report. In addition to these studies, a

[23] EPA has determined that it will not rely on Kisicki et al. (1999) because it was intentional human chlorpyrifos dosing study involving children less than 18 years old.

literature review was performed to identify relevant studies that were published following the Agency's previous 2008 SAP.

While the selected studies do not represent the entire universe of available studies, they are generally considered the major monitoring and epidemiologic studies that have monitored exposure to chlorpyrifos. CDC's National Health and Nutrition Examination Survey (NHANES) provides reference ranges of TCPy urinary levels in the general population during the 1999-2000 and 2001-2002 survey cycles (CDC, 2009).[24] Similarly, EPA's Office of Research and Development (ORD) has conducted or funded a number of observational studies to investigate children's exposure to pesticides and other contaminants, including: National Human Exposure Assessment Survey (NHEXAS); Minnesota Children's Pesticide Exposure Study (MSCPES); Children's Total Exposure to Persistent Pesticides and Other Persistent Organic Pollutants Study (CTEPP); Biological and Environmental Monitoring for Organophosphate and Pyrethroid Pesticide Exposures in Children Living in Jacksonville, Florida Study (JAX); and Children's Pesticide Post-Application Exposure Study (CPPAES) (Egeghy et al., 2011). The sample collection period of these studies ranged from 1995-1997 (NHEXAS) to 2001 (CTEPP), representing time periods when chlorpyrifos was registered for residential applications and the start of the phase-out.

Dow AgroSciences, the primary registrant of chlorpyrifos products, has also submitted a number of observational studies to the Agency that involved biomonitoring of TCPy following application of chlorpyrifos by adults in occupational and residential use scenarios. The majority of these studies have been incorporated into the Agency's assessment of occupational and residential human health risk assessments for the Chlorpyrifos Reregistration Eligibility Decision (EPA, 2006) and, more recently, the 2011 updated Occupational and Residential Exposure Assessment (EPA, 2011]. Dow's studies were intended to quantify environmental levels and exposures to chlorpyrifos following pesticide application in occupational and residential scenarios. For the purposes of comparison, a subset of Dow biomonitoring studies is presented in the section (MRID 44458201, MRID 43013501, MRID 43062701, MRID 43138102). The subset of studies was selected because it represents the high and low range of TCPy urinary levels observed in all the occupational and residential use scenarios evaluated by Dow.

In addition to the NHANES, ORD research, and registrant studies described above, a number of biomonitoring studies help further characterize the range of exposures in different population groups and the role of different exposure pathways. Several of the studies have previously reviewed during the Agency's 2008 SAP (EPA, 2008). Eaton et al. (2008) has also performed a review of exposure studies that have measured urinary TCPy levels. The studies reviewed include several that have measured to TCPy levels to investigate differences in pesticide exposure in farm and non-farm communities (Curwin et al., 2005, 2007; Alexander et al., 2006; Fenske et al., 2002). These studies help characterize the role of take-home exposure in agricultural communities. Similarly, biomonitoring studies have been conducted to investigate the role of dietary exposure (Lu et al., 2006, 2008). In these studies, children's conventional diets were replaced with organic foods to determine if an organic diet could lower children's dietary exposure to OP pesticides.

[24] CDC has not publically released TCPy urinary data from more recent NHANES survey cycles.

Comparison of Study Results

The monitoring studies described above can help characterize the exposure levels in the CHAMACOS, Columbia University, and Mt. Sinai cohorts described previously in Chapter 3. Specific studies from these cohorts include Eskenazi et al. (2007) and Castorina et al. (2010) from the CHAMACOS cohort, Whyatt et al. (2009) from the Columbia University cohort, and Berkowitz et al. (2003) from the Mount Sinai cohort. Based on the studies described above, comparisons of TCPy biomonitoring results for adult and child study populations are provided in Figure 4 and Figure 5, respectively. These figures provide information on the range of observed TCPy levels based on summary results reported by the investigators. As such, traditional boxplots could not be generated independently because raw study data from individual studies were not available. Whenever possible, boxplots are presented in which the box length indicates 25th and 75th percentiles, central line indicates median, and whiskers indicate 5th and 95th percentiles. However, several studies only reported central tendency values and the range of observed values. In these cases, study summary data are represented by a line that indicates the central tendency value (median unless noted) and whiskers indicating the range of observed values. In addition, the horizontal axis of each figure provides the study reference and description of study population. As part of the study description, the type of population is described, often based upon Table 17 of Eaton et al. (2008), along with the year of sample collection and sample size.

Child Study Populations

Figure 4 first provides a general reference range of urinary TCPy levels of 6 to 12 year old children in the general U.S. population based on NHANES 1999-2000 and 2001-2002, respectively (CDC, 2009). Based on comparison of these survey periods, 50th percentile TCPy levels were 2.8 and 3.1 µg/L, respectively. Likewise, 95th percentile urinary TCPy levels were fairly similar during the two survey periods and were 16.0 and 15.3 µg/L, respectively.

As has been previously described by Egeghy et al. (2011), median urinary TCPy levels were fairly similar across ORD monitoring studies of children and ranged from 5.1 µg/L in CTEPP to 12.0 µg/L in NHEXAS-AZ. These median levels were roughly 2-4 times higher than the median TCPy levels of 6-12 years old children in the 1999-2000 and 2001-2002 survey periods of NHANES. Comparison of studies that monitored children living in farm families or involved in farm activities (Alexander et al., 2005; Fenske et al., 2002), yielded results that were more similar to NHANES, although these studies were smaller in scale, making it more difficult to make direct comparisons.

The results of longitudinal studies examining the contribution of dietary pesticide exposure (Lu et al., 2006, 2008) were also comparable to the general range of TCPy levels observed in 6-11 year old children in NHANES. While both of these studies were comparable to NHANES, both studies also reported significant decreases in TCPy levels in their cohort following replacement of diet with organic foods. Lu et al. (2003) observed that median TCPy levels decreased from 7.2 µg/L during the initial conventional diet phase of the study to 1.7 µg/L during the subsequent organic diet phase. Following the re-introduction of conventional foods, the median TCPy level in children increased to 5.8 µg/L. Similarly, while not represented by the data presented in Figure 4, Lu et al. (2008) had study participants switch to organic diets during part of the summer and fall periods of their study and reported similar trends.

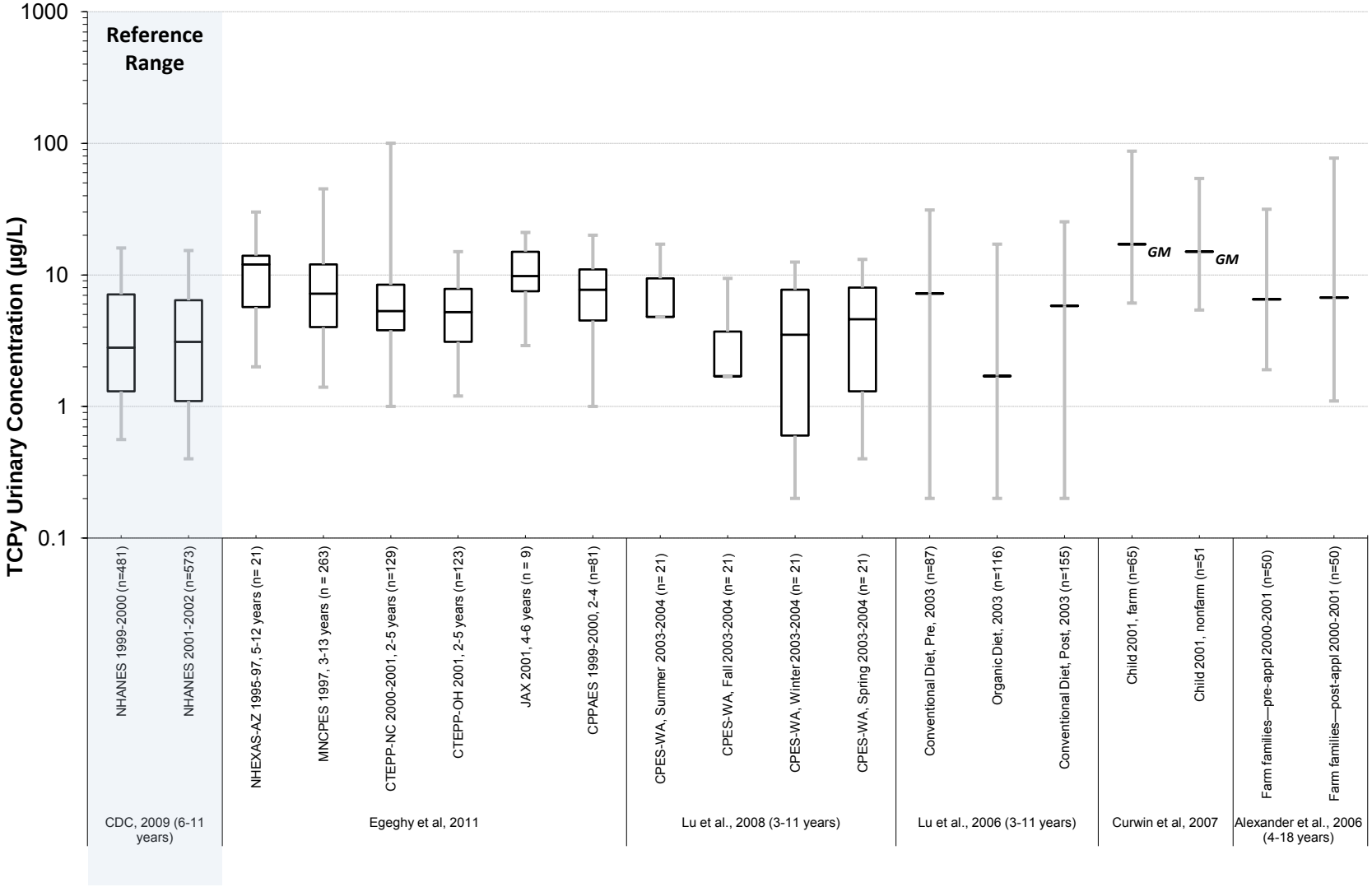
Adult Study Populations

As with the previous figure, Figure 5 first provides a general reference range of TCPy levels in the general adult U.S. population based on NHANES 1988-1992,1999-2000 and 2001-2002, respectively (Hill et al., 1995; CDC, 2009). Based on comparison of these three different survey periods, 50th percentile TCPy levels were 3, 1.5, and 1.9 µg/L, respectively. While this suggests that 50th percentile concentration may have declined relative to 1988-1992, the 95th percentile TCPy levels were 13, 9.9, and 10.9 µg/L, respectfully. As such, there appear to be smaller differences at the upper end of the distribution of TCPy levels in adults during the three survey periods.

Comparison of the range of TCPy levels observed in NHANES with other monitoring yielded mixed results. In studies that specifically monitored farmers or their families, some studies reported TCPy levels in adults that were within the range observed in NHANES (Curwin et al., 2005; Alexander et al., 2005), whereas Curwin et al. (2007) observed geometric mean TCPy levels in adults that were higher than the 95th percentiles in all three NHANES survey periods. On the other hand, TCPy levels in the registrant-submitted worker and residential applicator studies were generally higher than the range observed in NHANES. This was true for registrant studies that represented the both the lowest and highest TCPy levels in all eleven monitoring studies that have been submitted to the Agency.

TCPy levels of mothers that were part of the CHAMACOS, Columbia, and Mount Sinai cohorts also varied in comparison with the general range of TCPy levels observed in NHANES. Eskenazi et al. (2007) from the CHAMACOS cohort observed median levels that were higher than all NHANES adults aged 20-59 years old. Similarly, Castorina et al. (2010) performed a subanalysis of NHANES, which suggests that median TCPy levels observed in prenatal spot urine samples were slightly higher than the median levels TCPy levels of pregnant women in NHANES 2001-2002. Berkowitz et al. (2003) of Mount Sinai also observed median TCPy levels that were higher than NHANES. In particular, Berkowitz et al. observed the median TCPy level of their cohort was 7.6 µg/L, which was higher than the median TCPy levels observed in NHANES during the three survey periods. On the other hand, the TCPy levels reported by Whyatt et al. (2009) of the Columbia cohort appear to be lower than the TCPy levels observed in adults in the general U.S. population. Specifically, Whyatt et al. (2009) reported median TCPy levels that ranged from <LOD (0.26 µg/L) to 0.61 µg/L during pre/postnatal sample collection; however EPA notes that in contrast to the other cohorts mentioned these data were collected, in part, during a time period of rapid decline in use of chlorpyrifos (subsequent to the voluntary cancellation of indoor residential uses).

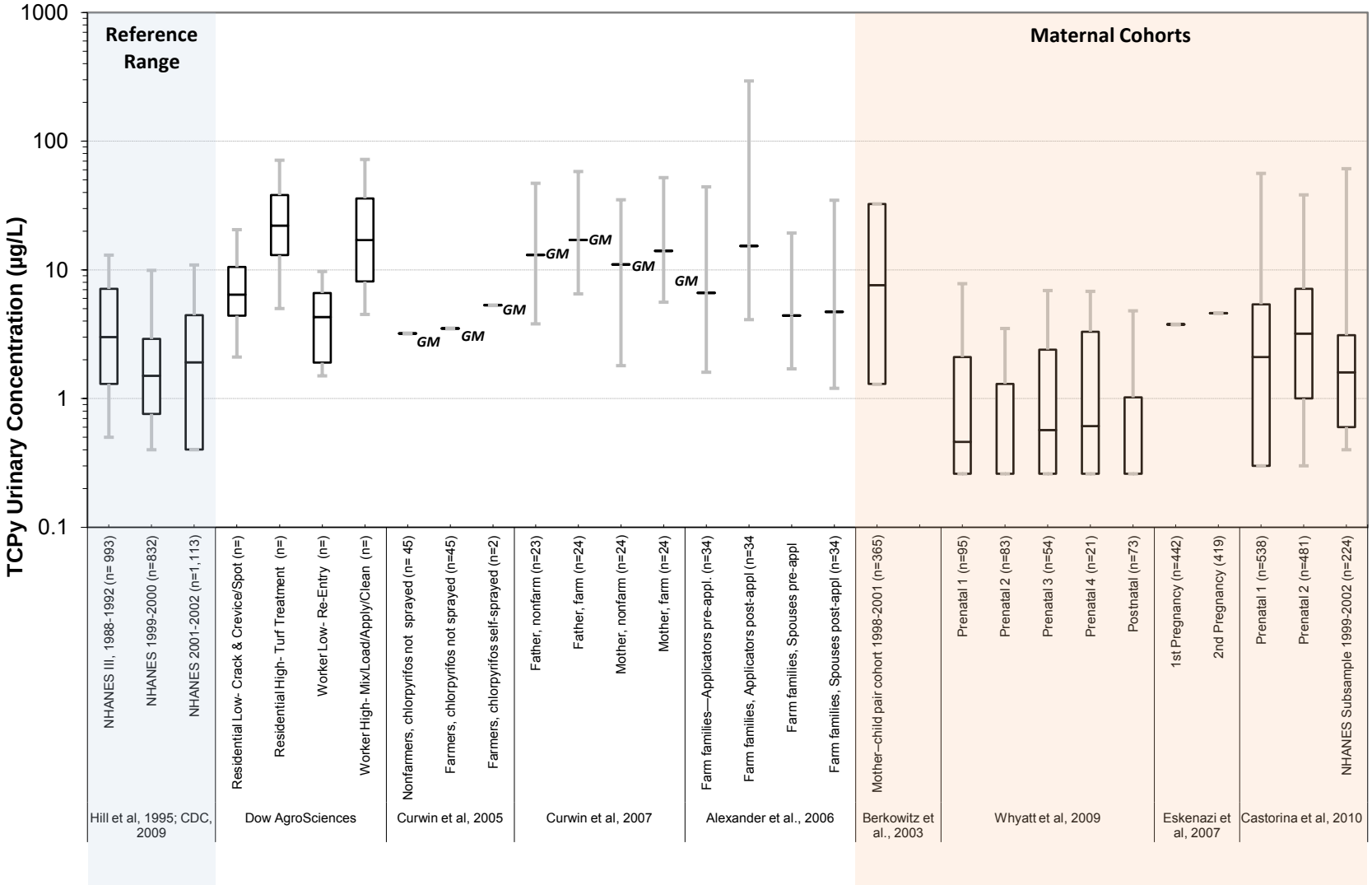
Figure 4 Summary of Comparison of TCPy Biomonitoring Studies Involving Child Study Populations.[a]



[a] Boxplots are presented in which the box length indicates 25th and 75th percentiles, central line indicates median, and whiskers indicate 5th and 95th percentiles. However, several studies only reported central tendency values and the range of observed values. In these cases, study summary data are represented by a line that indicates the central tendency value (median unless noted) and whiskers indicating the range of observed values.

GM – Geometric Mean

Figure 5 Summary of Comparison of TCPy Biomonitoring Studies Involving Adult Study Populations.[a]



[a] Boxplots are presented in which the box length indicates 25th and 75th percentiles, central line indicates median, and whiskers indicate 5th and 95th percentiles. However, several studies only reported central tendency values and the range of observed values. In these cases, study summary data are represented by a line that indicates the central tendency value (median unless noted) and whiskers indicating the range of observed values.

GM – Geometric Mean

Table 12 Summary Results of TCPy Biomonitoring Studies (Modified from Eaton et al., 2008)

Reference	Cohort	Type of Study Population	Collection Period	Collection Method	n	% Det.	Urinary TCPy Concentration (µg/L)									
							LOD	AM	GM	Min	5th	25th	50th	75th	95th	Max
Hill et al., 1995	NHANES III	20-59 year olds	1988-1992	Spot	993	82	1	4.5	-	-	<LOD	1.3	3	5.9	13	77
CDC, 2009	NHANES	6-11 year olds	1999-2000	Spot	481	-	0.4	-	2.88	-	0.56	1.3	2.8	7.09	16	-
	NHANES	6-11 year olds	2001-2002	Spot	573	-	0.4	-	2.67	-	<LOD	1.1	3.09	6.36	15.3	-
	NHANES	20-59 year olds	1999-2000	Spot	832	-	0.4	-	1.53	-	<LOD	0.76	1.5	2.9	8.9	-
	NHANES	20-59 year olds	2001-2002	Spot	1,113	-	0.4	-	1.91	-	<LOD	<LOD	1.91	4.44	10.9	-
Egeghy et al. 2011	ORD - NHEXES, AZ	5-12 year olds	1995-1997	Spot – Morning Void	21	100	-	12	9.3	2	-	5.7	12	14	26	30
	ORD - MNCPES	3-13 year olds	1997	Spot – Morning Void	263	92	-	9.2	6.6	<1.4	-	4	7.2	12	23	45
	ORD - CTEPP,NC	2-5 year olds	2000-2001	Spot – Three Voids	129	98	-	7.5	5.5	<1.0	-	3.8	5.3	8.4	16	100
	ORD - CTEPP, OH	2-5 year olds	2001	Spot – Three Voids	123	100	-	5.9	4.9	1.2	-	3.1	5.2	7.8	12	15
	ORD - JAX	4-6 year olds	2001	Spot – Morning Void	9	100	-	11	9.1	2.9	-	7.5	9.8	15	21	21
	ORD - CPPAES	2-4 year olds	1999-2000	Spot – Morning Void	81	93	-	8	6.4	<1.0	-	4.5	7.7	11	18	20
Lu et al., 2008	CPES-WA	3-11 year olds	2003, Summer	Spot - Longitudinal	23	86	0.2	6.4	-	-	0	0	2	4.8	9.4	-
	CPES-WA	3-11 year olds	2003, Fall	Spot - Longitudinal	21	88	0.2	2.6	-	-	0	0	0.6	1.7	3.7	-
	CPES-WA	3-11 year olds	2004, Winter	Spot - Longitudinal	20	96	0.2	5.1	-	-	0.2	0.6	1.7	3.5	7.7	-
	CPES-WA	3-11 year olds	2004, Spring	Spot - Longitudinal	19	97	0.2	5.6	-	-	0.4	1.3	2.5	4.6	8	-
Lu et al., 2006	WA Children	3-11 year olds	2003	Spot - Longitudinal	87	78	0.2	7.2	-	<LOD	-	-	6	-	-	31.1
	WA Children	3-11 year olds	2003	Spot - Longitudinal	116	50	0.2	1.7	-	<LOD	-	-	0.9	-	-	17.1
	WA Children	3-11 year olds	2003	Spot - Longitudinal	155	78	0.2	5.8	-	<LOD	-	-	4.3	-	-	25.3
Alexander et al., 2006	Farm families—pre appl	4-18 year olds	2000-2001	Spot	50	-	-	-	7.6	1.9	-	-	6.5	-	-	31.5
	Farm families—post-appl	4-18 year olds	2000-2001	Spot	50	-	-	-	7.6	1.1	-	-	6.7	-	-	77.1
	Farm families—pre appl	Applicators	2000-2001	Spot	34	-	-	-	7.8	1.6	-	-	6.6	-	-	44.1
	Farm families—post-appl	Applicators	2000-2001	Spot	34	-	-	-	19	4.1	-	-	15.3	-	-	293
	Farm families—pre appl	Spouses	2000-2001	Spot	34	-	-	-	4.7	1.6	-	-	4.4	-	-	24.1
	Farm families—post-appl	Spouses	2000-2001	Spot	34	-	-	-	5	1.2	-	-	4.7	-	-	34.7
Fenske et al., 2002	Applicator children	≤ 6 years olds	1995	Spot	49	20	8	-	-	<LOD	-	-	-	-	-	100
	Farm worker children	≤ 6 years olds	1995	Spot	12	33	8	-	-	<LOD	-	-	-	-	-	53
	Agricultural children	≤ 6 years olds	1995	Spot	61	23	8	-	-	<LOD	-	-	-	-	-	100
	Reference children	≤ 6 years olds	1995	Spot	14	29	8	-	-	<LOD	-	-	-	-	-	27
Curwin et al., 2005	Nonfarmers, not sprayed		2001, Spring-Summer	Spot - Longitudinal	45	89	0.5	-	3.3	-	-	-	-	-	-	-
	Farmers, not sprayed		2001, Spring-Summer	Spot - Longitudinal	45	89	0.5	-	3.5	-	-	-	-	-	-	-
	Farmers, self-sprayed		2001, Spring-Summer	Spot - Longitudinal	2	100	0.5	-	54.2	-	-	-	-	-	-	-

ATTACHMENT H

Reference	Cohort	Type of Study Population	Collection Period	Collection Method	n	% Det.	Urinary TCPy Concentration (µg/L)									
							LOD	AM	GM	Min	5th	25th	50th	75th	95th	Max
Curwin et al., 2007	Father, nonfarm		2001, Spring-Summer	Spot	23	94	3.32	-	13	3.8	-	-	-	-	-	47
	Father, farm		2001, Spring-Summer	Spot	24	100	3.32	-	17	6.5	-	-	-	-	-	58
	Mother, nonfarm		2001, Spring-Summer	Spot	24	95	3.32	-	11	1.8	-	-	-	-	-	35
	Mother, farm		2001, Spring-Summer	Spot	24	100	3.32	-	14	5.6	-	-	-	-	-	52
	Child, nonfarm		2001, Spring-Summer	Spot	51	100	3.32	-	15	5.4	-	-	-	-	-	54
	Child, farm		2001, Spring-Summer	Spot	65	100	3.32	-	17	6.1	-	-	-	-	-	87
Berkowitz et al., 2003	Mt. Sinai		1998-2001	Spot	365	-	-	-	-	-	-	1.8	7.5	25.7	-	-
Whyatt et al., 2009	Columbia	Prenatal 1	2001-2004	Spot	95	52	0.26	-	-	-	-	<LOD	0.46	2.1	7.8	-
	Columbia	Prenatal 2	2001-2004	Spot	83	48	0.26	-	-	-	-	<LOD	<LOD	1.3	3.5	-
	Columbia	Prenatal 3	2001-2004	Spot	54	56	0.26	-	-	-	-	<LOD	0.57	2.4	6.9	-
	Columbia	Prenatal 4	2001-2004	Spot	21	62	0.26	-	-	-	-	<LOD	0.61	3.3	6.8	-
	Columbia	Postnatal	2001-2004	Spot	73	40	0.26	-	-	-	-	<LOD	<LOD	1.02	4.8	-
Castorina et al., 2010	CHAMACOS	Prenatal 1	1999-2000	Spot	538	71.2	0.3	-	-	-	<LOD	<LOD	2.1	5.4	16.9	56.1
	CHAMACOS	Prenatal 2	1999-2000	Spot	481	81.9	0.3	-	-	-	<LOD	1	3.2	7.1	17.9	38.1
	NHANES, 1999-2002	Preg. Subsample	1999-2002	Spot	224	79	0.4	-	-	-	<LOD	0.6	1.6	3.1	7.4	61
Eskenazi et al., 2007	CHAMACOS	1st Preg.	2000-2003	Spot	442	71	-	-	-	-	-	-	3.76	-	-	-
	CHAMACOS	2nd Preg.	2000-2003	Spot	419	82	-	-	-	-	-	-	4.6	-	-	-

5.3 Chlorpyrifos Biomonitoring and AChE Inhibition

Monitoring of ChE has been performed in both research and state-based occupational surveillance to measure potential changes in blood ChE levels following exposure to OP and carbamate pesticides (Barr and Angerer, 2006; Lessenger, 2005; WA State, 2011). The most common biomarkers used to monitor ChE activity include erythrocyte AChE and plasma BuChE. Inhibition of erythrocyte AChE is generally considered the most relevant measure of potential toxicity of chlorpyrifos and other compounds that act as ChE inhibitors (Eaton et al., 2008). While AChE is believed to have greater toxicological relevance, plasma BuChE has been shown to be inhibited at lower levels of exposure than erythrocyte AChE, making plasma BuChE a more sensitive biomarker for the purposes of monitoring potential physiological change.

Although analytical methods are available to measure AChE and BuChE levels, evaluation of inhibition requires careful study design and analysis. Most notably, AChE and BuChE activity can have relatively high inter- and intra-individual variability. Due to this variability, reference ranges of activity have not been established to evaluate inhibition in the general population or occupational groups. Rather, researchers and state-based occupational surveillance programs evaluating inhibition have typically utilized longitudinal study designs that compare AChE/BuChE activity during a "baseline" period where only background exposure is expected to occur with an "exposure" period where exposure may occur in excess of background levels (e.g., season in which pesticide applicators apply chlorpyrifos). AChE/BuChE activity can also be influenced by a number of other factors, including pregnancy status, disease status, exposure to other ChE inhibitors, and illegal drug use. As such, it may be important to control for these types of factors when evaluating inhibition. Another important consideration is that changes in AChE activity may be less sensitive to lower levels of environmental exposure that may be experienced by the general population (Barr and Angerer, 2006). For this reason, examination of AChE inhibition has generally been more common in occupational studies involving chemical manufacturing or agricultural study populations. While it may be difficult to monitor AChE inhibition in non-occupational study populations, some investigators have measured AChE and BuChE as indicators of pesticide exposure in epidemiologic investigations. Eskenazi et al. (2004), for example, measured AChE and BuChE in maternal and umbilical cord blood and evaluated the association between AChE and BuChE and length of gestation and fetal growth.

Previous Agency Evaluation and Findings

The Agency previously evaluated the relationship between both chlorpyrifos blood and urinary TCPy levels and AChE activity during its 2008 SAP (EPA, 2008).[25] In its evaluation, the Agency considered human toxicity studies that measured chlorpyrifos blood levels and AChE/BuChE inhibition in adults that were administered a single dose of chlorpyrifos (Nolan et al., 1982). For additional context, the Agency also compared Nolan et al. (1982) with other biomonitoring studies in order to determine how the chlorpyrifos blood levels of study participants compared to blood levels reported in epidemiologic and biomonitoring studies in the literature. Based on its evaluation, the Agency noted AChE inhibition in some study participants, but indicated that there was large variability in response to the administered dose of chlorpyrifos, making it difficult to draw definitive conclusions about the relationship between

[25] This section does not consider Kisicki et al. (1999), an intentional exposure study that monitored both TCPy and AChE. EPA has determined that it will not rely on Kisicki et al. (1999) because it was intentional human chlorpyrifos dosing study involving children less than 18 years old.

urinary TCPy levels and AChE inhibition. In addition, the Agency also noted that the chlorpyrifos blood levels were orders of magnitude lower than cord and maternal blood levels observed by Whyatt et al. (2009) in the Columbia Cohort which included births during the pre- and post-voluntary cancellation period.

Recent Studies on the Association between Urinary TCPy and AChE Inhibition

Following the 2008 SAP, two observations studies have been published that further evaluate the relationship between TCPy on a creatinine-adjusted basis and AChE inhibition (Galabrant et al., 2009; Farahat et al., 2011). Both of these studies involved occupational populations exposed to chlorpyrifos at levels that were much higher than the general U.S. population, based on comparison with the NHANES 1999-2000 and 2001-2002 survey cycles (See Table 13 below).[26] While these studies involved study populations that were exposed to much higher levels than the general U.S. population, both studies utilized effective study designs for evaluating the association between urinary TCPy and AChE/BuChE inhibition.

Table 13 : Comparison of Creatinine-Adjusted Urinary TCPy levels (µg /g creatinine) reported by Galabrant et al. (2009) and Farahat et al. (2011) with the General U.S. Population.

Population Group	Time Period	n	TCPy (µg /g creatinine)		
			Average	Median (95% CI)	95th Percentile (95% CI)
NHANES (CDC, 2009)					
General Population	1999-2000	1,994	*NR*	1.5 (1.2-1.7)	8.4(6.3-11.6)
	2001-2002	2,508	*NR*	1.9 (1.6-2.2)	9.2 (6.9-12.3)
Galabrant et al, 2009					
Exposure Group	Fall 1999	53	*NR*	37.5	432.6
	Spring 2000	53	*NR*	40.7	436.3
	Turnaround	50	*NR*	59.2	7,403.8
	Fall 2000	52	*NR*	51.5	987.2
Reference group	Fall 1999	60	*NR*	3.2	13.9
	Spring 2000	57	*NR*	4.8	11.2
	Turnaround	57	*NR*	6.6	13.0
	Fall 2000	58	*NR*	6.3	15.5
Farahat et al, 2011					
Applicators	1- to 2- week Exposure Period	14	6,437	*NR*	*NR*
Technicians		12	184	*NR*	*NR*
Engineers		12	157	*NR*	*NR*

NR – Not reported

In Galabrant et al. (2009), the association between AChE/BuChE was evaluated in a group of exposed workers at a chlorpyrifos manufacturing facilities and a reference group of chemical manufacturing workers. The study utilized a longitudinal design over a year-long period (1999-2000) that involved collection of blood and urine samples during two physical examinations and two additional first morning void samples that were collected during the spring and fall of the study period, respectfully. Based on

[26] Investigators only reported results on a creatinine-adjusted basis, so the results could not be incorporated into the TCPy comparison provided in the previous section.

their statistical analysis, the investigators reported that no relationship was between urinary TCPy levels and AChE activity was observed over the entire range of TCPy levels. While no association was reported in relation to AChE, the investigators reported that there was a statistically significant association between TCPy levels and BuChE levels above a cutpoint of 110 µg TCPy/g creatinine.

The second recent study that has examined the association between TCPy and AChE/BuChE involved cotton field workers in Egypt (Farahat et al., 2011)[27]. In the study, daily urine samples and weekly blood samples were collected before, during, and after 9 – 17 consecutive days of chlorpyrifos application to cotton fields. All study participants were observed to have detectable levels of urinary TCPy at the start of the study, which ranged from 4.1 to 4,080 µg TCPy/g creatinine. Following the start of application, TCPy levels increased and were reported to be significantly higher in applicators (6,437 µg/g creatinine) than in technicians (184 µg/g) and engineers (157 µg/g). Baseline BuChE activity was reported to range widely during the baseline period of the study. Following the start of the chlorpyrifos exposure period, the investigators reported that BuChE activity was suppressed, but a specific quantification of suppression was not reported. The AChE activity of study participants was reported to be less variable than BuChE during the baseline period of the study. Following exposure, the investigators reported that average AChE activity was inhibited by 43, 73, and 70% during the July-2, July-10, and July-24 collection time points, respectively.

Based on their statistical analysis, Farahat et al. (2011) reported that they identified an inflection point representing a significant decrease in AChE activity at TCPy levels greater than 3,148 µg TCPy/g creatinine. In discussing their results, the investigators noted that few studies have attempted to identify a dose-effect relationship between urinary TCPy and blood ChE activity. They also discussed their results in comparison with Galabrant et al. (2009) and indicated that both studies observed similar inflection point of inhibition of BuChE activity. In particular, Galabrant et al. (2009) reported an inflection point of BuChE inhibition at 110 µg TCPy/g creatinine and Farahat et al. (2011) reported a similar reflection point at 114 TCPy/g creatinine.

Galabrant et al. (2009) and Farahat et al. (2011) provide evidence that suggests that there is an association between TCPy and BuChE at higher levels of exposure. Both studies utilized effective study designs for evaluation of the association between urinary TCPy and AChE/BuChE inhibition. In particular, each study utilized a longitudinal design. Farahat et al. had both a “baseline” period where occupational exposure was not reasonable expected to occur and an “exposure” period with repeated measurement of urinary TCPy levels and blood AChE/BuChE activity. Galabrant et al. had a similar longitudinal design, but did not establish “baseline” AChE/BuChE levels for the purpose of evaluating inhibition. While Galabrant et al. were unable to establish baseline AChE/BuChE activity, their inclusion of a reference population enabled them to evaluate the relationship between absolute AChE activity and TCPy. Limitations were also noted by the investigators. Galabrant et al., for example, reported that there study population only included workers and did not include potentially sensitive populations, such as children or elderly. Their study population did include some female study participants (22.6% and 26.7% of the exposure and reference groups, respectively).

[27] While Farahat et al. (2011) provides information on the relationship between TCPy levels and AChE inhibition, the levels of exposure observed in the Egyptian farmworker population may not be representative of U.S. farm practices. As such, it may not be appropriate to use Farahat et al. (2011) to make conclusions about farmworker exposure in the U.S.

5.4 Approaches to Interpreting Biomonitoring Data

There is a need in environmental exposure assessment to quantitatively interpret the results of biomonitoring studies. Quantitative analysis involves the reconstruction of human exposure estimates and scenarios most likely associated with measurements of analytes in human biological specimens such as blood and urine. The previous section provided an overview of the available TCPy data and discussed some of the challenges in its interpretation. The TCPy data are presented in this document as published. The Agency will be soliciting comments on approaches to best interpret such biomonitoring data given the discrete and intermittent nature of human environmental exposures and chemical-specific pharmacokinetic behavior. As part of the consideration for the SAP, the following section summarizes various approaches that can be used in the interpretation of the TCPy data. The approaches are generally discussed in order of increasing sophistication (*i.e.*, qualitative to quantitative) including the most sophisticated physiologically based pharmacokinetic (PBPK) modeling approach. The strengths and limitations of each approach are briefly discussed.

5.4.1 Consideration of Approaches for Interpretation of Biomonitoring Data

- *Qualitative-Noncomparative:*
  This approach is based on simply laying out biomonitoring data as in the previous section. Interpretation is in the context of measures of exposure (for example, pre- and post- mitigation periods) without any quantitative link to exposure levels to chlorpyrifos, exposure route, lifestage, or health outcome.

- *Qualitative-Comparative:*
  Similar to Qualitative-Noncomparative except that the analysis will also include gross comparisons to the available database of human deliberate dosing studies with chlorpyrifos. The primary comparison will be to a controlled human study performed by Nolan and co-workers in which human volunteers were administered a single oral dose of 0.5 mg/kg bw chlorpyrifos and the resulting blood and urinary TCPy levels were measured as well as plasma cholinesterase inhibition as a potential marker of early effects (Nolan et al., 1984). Uncertainties arise when comparing biomonitoring data lacking critical information on timing of exposure to those obtained from a controlled study involving a single oral dose of chlorpyrifos in a single gender (males) (Nolan et al., 1984). In general, biomonitoring data collected in observational studies reflect repeated environmental exposures, multiple exposure sources and routes, unknown timing of exposures, and unless specified, from both genders and different life-stages. Historically, the Nolan data have been used in the evaluation of occupational chlorpyrifos exposures. A number of biomonitoring studies were submitted by Dow AgroSciences in which pre- and post-exposure urine measures were conducted and measured for TCPy. Measures of total chlorpyrifos (µg) absorbed were calculated using the "kinetic method." This method estimated the amount of chlorpyrifos absorbed by fitting the amount of TCPy excreted in the individual urine specimens to a one compartment pharmacokinetic model that described the time course of TCPy in urine of volunteers following application of the chlorpyrifos to their forearm (Nolan et al., 1984). Based on the Nolan et al., 1984 study, it was assumed that 72% of the absorbed chlorpyrifos was excreted into the urine as TCPy. This methodology is more fully described in the example biomonitoring study report (Honeycutt and Day 1994).

- *Semi-Quantitative Screening:*
  This approach would be based on the derivation of "biomonitoring equivalents (BE)" as a screening tool for comparison to chemical levels in biomonitoring data. The BE approach does not generate human environmental exposure estimates per se, but rather uses pharmacokinetic information and modeling for forward dosimetry analysis to estimate biomarker levels of exposure likely to be associated with regulatory human external exposure guidelines (e.g., RfD). The resulting estimates can then be compared to those in biomonitoring data. The main caveat with this approach is that uncertainty arises if the biomarker lacks specificity as biomarker of exposure and/ordoes not closely align with a toxicologically relevant internal dose metric. Both of these represent the case with the use of TCPy as a biomarker of exposure. TCPy can be formed as a result of direct oxidative dearylation of chlorpyrifos (i.e., without the formation of chlorpyrifos-oxon as intermediate) and also happens to be an environmental degradate to which human direct exposures take place. Therefore, caution should be exercised in the interpretation of biomonitoring data of TCPy based on the BE approach.

- *Semi-Quantitative Reverse Dosimetry:*
  This approach, like that proposed by Mage and co-workers (Mage et al., 2004), makes use of information on body surface area, daily creatinine excretion rate to normalize urinary analyte levels, and most importantly for bounding dose estimates, the ratio of moles of excreted analyte expected per mole of pesticide intake. The resulting human dose estimate represents an estimate from all relevant routes of exposure without accounting for route-specific bioavailability. The Mage et al. (2004) approach has been applied to the interpretation of at least two large datasets: urinary measurements in the NHANES III study (Mage et al., 2004) and in a comparative analysis of pesticide dose estimates for children of Iowa farmers and non-farmers (Curwin et al., 2007). The Agency has utilized the Mage et al., 2004 approach, in combination with other approaches that do not use creatinine normalization, in the analysis of biomonitoring data associated with triclosan (RED 2008). A major limitation of the Mage et al. (2004) approach is that the human dose estimate does not account for route-specific bioavailability and the contribution of each route of exposure would need to be carefully elucidated for any type of interpretation. Moreover, as with any approach, the contribution of direct dietary exposures to TCPy as an environmental degradate would need to be carefully considered in any analysis associated with environmental exposures to chlorpyrifos.

- *Quantitative PBPK Approach:*
  By far, the preferred approach for reconstructing exposure estimates is a well-calibrated and evaluated PBPK model. These models provide an anatomically/physiologically bounded quantitative structure within which the pharmacokinetics of chemicals can be described. Because PBPK models can predict internal dosimetry, they can be used in reconstructing human exposures (including scenarios) that may be associated with biomonitoring data. The major disadvantage of PBPK models is that their development including parameterization, calibration, and evaluation requires a significant amount of data.

  There is a well-developed PBPK model available for the AChE inhibiting mode of action of chlorpyrifos that provides the critical internal dose metrics associated with AChE

inhibition. The model was developed based on adult non-pregnant rats and was then scaled and calibrated for the human counterpart (*i.e.*, adult, non-pregnant human) (Timchalk et al. 2002). The model in its current state can only simulate oral exposures to chlorpyrifos and does not account for any lifestage-related changes applicable to fetuses, infants, or children.

Table 14 Comparison of Approaches for Interpretation of Chlorpyrifos (CPF) Biomonitoring Data

Approach	Strengths	Weaknesses/Uncertainties
Qualitative- Noncomparative	- Simple - No need for pharmacokinetic information or modeling	-Gross comparison with no link to exposure (levels or route), lifestage, or health outcome
Qualitative-Comparative	- Simple - No need for pharmacokinetic information or modeling	-Comparison is to controlled human oral studies irrespective of dosing regimen, exposure route, lifestage, or gender
Semi-Quantitative Screening	- Simple - Makes use of some pharmacokinetic information/modeling - Dose estimates can be compared to a exposure regulatory guideline (*e.g.*,RfD) - Can incorporate some lifestage-specific information in clearance estimates	-Screening tool - Urinary levels of TCPy may not be the most relevant dose metric
Semi-Quantitative Reverse Dosimetry	-Simple - Can incorporate some lifestage-dependent changes in body surface area and creatinine excretion rate. - Provide quantitatively bound estimates based on expected moles of urinary analyte per mole of parent chemical	- Provides absorbed dose estimates without route-specific information - Assumes that analytes are derived from a single parent chemical
PBPK-PD model	- Provides critical internal dose metrics associated with AChE inhibition (levels of CPF-oxon and AChE inhibition) - Provides internal dose metrics for potential non-cholinergic effects (levels chlorpyrifos and oxon metabolite)	- Does not address all susceptible lifestages - Does not incorporate all relevant routes of exposure

Chlorpyrifos is a unique in that its database is extensive; there is a wealth of information on metabolism, mechanism of cholinesterase inhibition, markers of exposure and effects, and a robust PBPK-PD model developed for adult non-pregnant adults. Furthermore, its biomonitoring database is also extensive and continues to grow. The Agency is considering approaches to best evaluate the current biomonitoring information and is soliciting input from the SAP. There is no ideal reverse dosimetry analysis approach. Each approach needs to be carefully considered for its strengths and limitations. Table 14 provides a brief synposis of several approaches discussed above. The following section 5.4.2 provides a more extensive background of the PBPK-PD model available for chlorpyrifos.

5.4.2 Physiologically Based Pharmacokinetic-Pharmacodynamic Model for Chlorpyrifos: State of the Science

A physiologically based pharmacokinetic model (PBPK) model for chlorpyrifos that was originally developed by Timchalk and coworkers in 2002 (Timchalk et al., 2002) has been refined over the years as more data has become available (Busby-Hjerpe et al., 2010; Cole et al., 2005; Garabrant et al., 2009; Lee et al., 2009; Lowe et al., 2009; Lu et al., 2010; Marty et al., 2007; Timchalk and Poet, 2008; Timchalk et al., 2005; Timchalk et al., 2006). The model is based on the mechanistic mode of action information that chlorpyrifos exerts its cholinergic toxicological effects through metabolic activation to chlorpyrifos-oxon and detoxification to TCPy and DAP metabolites DEP and DMP. Chlorpyrifos-oxon inhibits AChE through covalent modification of an active site serine active site, resulting in build-up of acetylcholine and hyperstimulation at synapses in brain, spinal cord, and peripheral nervous system (Timchalk et al., 2002). The chlorpyrifos PBPK model has evolved to include the key event in the cholinergic toxicity mode of action: AChE inhibition. Thus, the PBPK model includes pharmacodynamics (*i.e.*, a PBPK-PD model) that can be used to predict the critical dose metrics associated with cholinergic toxicity following chlorpyrifos exposure: CPF-oxon levels and the resulting AChE inhibition in target tissues. As CPF-oxon is very reactive and can modify AChE at non-target sites such as red blood cells (RBCs) as well as other proteins with esterase activity, the model also includes inhibition of AChE activity in RBCs and plasma cholinesterase as potential biomarkers of early effects. Also predicted by the model are TCPy levels (in plasma and excreted urine) as a biomarker of exposure given that this metabolite is associated with detoxification of chlorpyrifos.

Although variations of the original Timchalk et al. (2002) PBPK-PD model have been published, the fundamental model structure has remained largely the same. The Agency's SAP recommended in 2008 that the Agency consider further refinement of the Timchalk et al. (2002) model for use in the human health risk assessment of chlorpyrifos (FIFRA SAP, 2008). In recent collaborative research effort between Battelle and Dow Agrosciences (DAS et al., 2011), the chlorpyrifos PBPK-PD model was linked to the probabilistic dietary exposure model CARES and expanded it to include different age groups: infants (6 months), children (3 year-olds), and adults (30 year olds). The final composite model represents a "source-to-outcome" model that, if robust enough, could be used to assess population risk to cholinesterase inhibition starting with data on crop residues levels of chlorpyrifos. The Agency's Science SAP reviewed the final composite model at its February 2011 meeting (FIFRA SAP, 2011). Although the Panel commended the effort, confidence was not expressed in using the model developed for the typical adult non-pregnant human to infer on the pharmacokinetics of chlorpyrifos (and its metabolites) in earlier lifestages. One concern reason cited by the Panel was that the model did not include gestational and lactational exposures which require explicit attention. Further, the Panel expressed concern about the *in vitro*-based parameterization of metabolism in the model based on a limited number of age-appropriate samples (FIFRA SAP, 2011).

5.4.2.1 Lifestages considered in the current chlorpyrifos PBPK-PD model

The current PBPK-PD model for chlorpyrifos is based on the typical non-pregnant adult rat and human and does not inform about lifestage-related susceptibility. This is a key deficiency in the model for use in human health risk assessment because there is a large body of evidence indicating that the developing nervous system may be a particular susceptible target of toxicity associated with chlorpyrifos. The evidence comes from rodent studies from multiple laboratories indicating neurobehavioral changes resulting from peri-natal exposures to chlorpyrifos. Epidemiological studies have also supported associations between prenatal exposures to OPs including chlorpyrifos, and cognitive delays in children.

At the February 2011 SAP meeting, the Panel specifically noted the lack of maternal and fetal pharmacokinetics and pharmacodynamics in the current PBPK-PD model as a deficit to inform about pregnancy and early lifestages (FIFRA SAP 2011) and criticized the use of a simple multiplicative factor to account for pregnancy-related changes in order to make comparisons to biomonitoring data (DAS et al. 2011).

Attempts have been made to extrapolate the current chlorpyrifos PBPK-PD model to include earlier lifestages. In another effort by Timchalk and coworkers (Timchalk et al., 2007), the existing adult rat PBPK-PD model was extrapolated to preweanling rats by incorporating age-dependent changes in tissue volumes, blood flows, and CYP450 and PON-1 metabolic activities. The modeling results supported the notion that the increased susceptibility of young rats may due to pharmacokinetic differences; chlorpyrifos-oxon levels in blood and brain increased disproportionally with chlorpyrifos dose as compared to adult non-pregnant rats. With the exception of not incorporating *in utero* and lactational exposures, the age-dependent rat model provided a good starting point for developing a human equivalent PBPK model that would address exposures to infants and children (Timchalk et al., 2007).

A more recent effort to extrapolate the model to earlier lifestages was published in the peer-reviewed literature by Lu and co-workers (Lu et al., 2010). The modeling effort focused on developing a PBPK model to predict urinary levels of TCPy in children. The model was implemented using the Agency's Exposure Related Dose Estimating Model (ERDEM) to accommodate for aggregate exposures to chlorpyrifos including bolus ingestion, inhalation, and ingestion based on non-dietary routes (hand-to-mouth)(Blancato et al., 2006). The dermal route was not included in the analysis because chlorpyrifos was not detected on any of the children's hands. Although a significant amount of data based chlorpyrifos measurements in indoor air, food, and house dust was used in the effort, the resulting model performed poorly by significantly under-predicting levels of TCPy in children (Lu et al., 2010). The Agency is also aware of a PBPK modeling effort led by Dr. Dale Hattis of Clark University in collaboration with the Columbia University epidemiology team. This PBPK model may, in the future, be useful in clarifying the exposure concentrations that correspond to the chlorpyrifos levels in umbilical cord blood associated with statistically significant neurodevelopmental effects. However, the development of the Hattis effort is further behind than other research groups (Hattis, 2011)

In summary, there is a robust PBPK-PD model available for chlorpyrifos for describing its AChE-inhibition mode of action; however, the model is restricted to the "typical" non-pregnant adult human and does not inform about the most susceptible lifestages which includes pregnancy, infants, and children. Although several modeling efforts have been undertaken to extrapolate the current model to different lifestages, the efforts have yet to produce a robust PBPK model that can be used for human health risk assessment applications (as opposed to for AChE-inhibition mode of action). Modeling future efforts aimed PBPK modeling for chlorpyrifos needs to include *in utero* exposures along with dermal and/or inhalation routes of exposure. The absence of gestational stages in the model was regarded by the SAP as a major deficit that prevents the chlorpyrifos PBPK-PD model from being an optimal prototype for risk assessments (FIFRA SAP, 2011).

5.4.2.2 Routes of Exposure simulated by the PBPK-PD model

The current PBPK-PD model for chlorpyrifos is limited to oral exposures to chlorpyrifos with particular emphasis on dietary exposures (excluding nursing infants), even though dermal and inhalation are also important routes of exposure, particularly in occupational settings. The pharmacokinetics of chlorpyrifos (and its metabolites) and resulting cholinesterase inhibition profile via other relevant routes of exposure are expected to be distinct as compared to the oral route profile predicted by the model. For a rapidly and extensively metabolized chemical such as chlorpyrifos whose cholinergic toxicity is dictated by bioactivation, route-specific metabolism can have a dramatic influence on the site, extent, and temporality of toxicity. For instance, in a rat study where dosing was performed via the oral route, cholinesterase inhibition occurred in plasma far earlier and at a lower dose than in AChE inhibition in brain tissue (Mendrala et al. 1998). In contrast, when chlorpyrifos was administered to adult non-pregnant rats via an aerosol through the inhalation route, the result was cholinesterase inhibition occurring far earlier in lung tissue followed by plasma, while no AChE inhibition was reported in brain tissue at any of the exposure levels tested (Hotchkiss et al. 2010). A dermal toxicity study in rats resulted in a similar profile as via the oral route (plasma cholinesterase inhibition occurred to a greater extent than RBC AChE inhibition). However, it is important to note that, via the dermal route, blood flow to the liver, the major site of metabolism of chlorpyrifos, will be limited by the input of the hepatic artery which can lead to temporal changes in cholinesterase inhibition as compared to other routes of exposure.

Therefore, the uncertainties associated with the pharmacokinetics of chlorpyrifos via dermal and inhalation routes significantly limit the utility of the current chlorpyrifos PBPK model for risk assessment applications. The SAP also supported this conclusion at its February 2011 meeting by stating: “the important pharmacokinetic differences between oral dosing and other routes of exposure, first pass metabolism, can have a dramatic influence on the extent and timing of the functional changes under consideration following OP exposure.” (FIFRA SAP 2011)

5.5 Conclusions

A large number of biomonitoring studies have evaluated exposure to chlorpyrifos by measuring urinary TCPy levels. While the biomonitoring database is extensive and continues to grow, the relationship between urinary TCPy levels and external chlorpyrifos exposure remains uncertain, making it difficult to directly evaluate biomonitoring data in a risk assessment context. One important interpretative limitation is that TCPy lacks the desired specificity as biomarker because urinary TCPy reflects concurrent human exposures to chlorpyrifos and TCPy as an environmental degradant. Further, reverse dosimetry approaches including a PBPK model are available, but each has limitations that need to be carefully considered for any type of interpretation. The current PBPK model for chlorpyrifos is limited to simulating oral exposures to chlorpyrifos and thus cannot be used to evaluate urinary TCPy which is likely also reflective other routes of exposure such as inhalation and dermal as well as direct exposures to TCPy. Further, the model has limited utility for evaluating maternal/umbilical cord blood measurements of chlorpyrifos since it does not incorporate the critical physiological and chemical-specific changes associated with pregnancy and in utero exposures. The ideal PBPK/PD model would accommodate oral, dermal, and inhalation routes and be informative of multiple lifestages (e.g., gestation, birth, childhood, and adulthood). This ideal model may not be available for the revised risk assessment. In the absence of such a model, the Agency is soliciting comments from the Panel on approaches for interpreting the biomonitoring data currently available for chlorpyrifos.

6.0 Summary & Preliminary Conclusions

Chlorpyrifos is a widely studied pesticide using an expansive suite of toxicity tests conducted in accordance with harmonized, scientifically peer-reviewed study protocols and published in the scientific literature. Thus, chlorpyrifos has a robust database across many scientific areas (*e.g.* metabolism, mode of action, biomonitoring, and human health toxicity studies) and lines of scientific evidence (*i.e. in vitro*, laboratory animal, *in vivo,* epidemiology), reflective of different levels of biological organization, and across various lifestages (fetuses, postnatal, pregnant and non-pregnant adult).

It is well established that AChE inhibition is the primary mode of action/adverse outcome pathway for OPs, like chlorpyrifos. Because AChE inhibition is the initiating event for this mode of action/adverse outcome pathway, using AChE inhibition as a regulatory endpoint is protective of downstream cholinergic effects. Moreover, historically, given the sensitivity of AChE inhibition data for OPs, these data have been considered to be protective of other potential toxicities and/or modes of action for OPs. As discussed in detail throughout this draft issue paper, newer lines of research on chlorpyrifos, including epidemiological studies in mothers and children, have posed the issue of whether AChE inhibition is the most sensitive endpoint, and thus have raised some uncertainty in the chlorpyrifos risk assessment. In order to determine the degree to which these recent studies are appropriate for incorporation into risk assessment (qualitatively and/or quantitatively) the Agency is taking a stepwise, objective and transparent approach to evaluate and interpret all the lines of scientific information related to the potential for adverse neurodevelopmental effects in infants and children as a result of prenatal exposure to chlorpyrifos, as well as to characterize thoroughly the strengths and uncertainties associated with these studies.

The evaluation of these recent studies is stepwise process which began with the September 2008 FIFRA SAP involving a preliminary review of the literature for chlorpyrifos, with a particular focus on women and children. Subsequent Agency activities have involved developing approaches for performing risk assessment of semi-volatile pesticides, and developing the draft "Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment" for integration of epidemiology with other types of experimental data (USEPA, 2010a, b; FIFRA SAP 2010a,b). In summer 2011, the Agency released its preliminary human health risk assessment that focused on the AChE inhibiting potential of chlorpyrifos. This focus is consistent with the recommendation from the 2008 SAP that AChE data provide the most appropriate endpoint and dose-response data for deriving point of departure for purposes of risk assessment. Moreover, because of the Agency's long experience with assessing the potential risk to chlorpyrifos and other OPs, and because both the dose response approach based on AChE inhibition and also the exposure methodologies used (as noted in previous sections of this issue paper) in the 2011 assessment have been vetted by numerous SAPs, there is high confidence in those analyses.

The 2011 risk assessment noted that a full weight of the evidence analysis that explicitly considers uncertainty and implications of modes of toxic action related to experimental and epidemiologic lines of evidence using factors such as biological plausibility, strength, consistency, dose-response and temporal concordance, will be conducted in the future. A full weight of the evidence including characterization of uncertainty has not yet been conducted; however, the April 2012 FIFRA SAP is an important step toward this effort. Unlike the dose-response and exposure analyses conducted for the 2011 chlorpyrifos risk assessment, the types of information reviewed here (*e.g.*, epidemiology studies, biomonitoring data, modes of action/adverse outcome pathways for neurodevelopmental outcomes, etc) are less often

available and approaches for their use in risk assessment (qualitatively or quantitatively) are less well established. The input from the SAP will be helpful as the Agency moves to the next stages of the weight of evidence analysis and uncertainty characterization including presenting various risk assessment alternatives or options and describing the range of potential risks.

This 2012 draft issue paper builds on the 2008 SAP review and is informed by the public comments received in 2011 in response to the preliminary chlorpyrifos human health risk assessment. In previous sections of this issue paper, the state of the science with respect to lifestage susceptibility, modes of action/adverse outcome pathways for AChE inhibition and for neurodevelopmental outcomes, epidemiology studies in mothers and children, biomonitoring data, and PBPK modeling have been fully reviewed. Each of these lines of evidence is important for the pending weight of the evidence analysis and uncertainty characterization.

- *AChE inhibition:*

In preparation for the 2008 SAP, the Agency performed a comprehensive review of the literature for AChE inhibition. This review has been supplemented with the newest data; together, the preliminary 2008 literature review and the newest studies inform the selection of point of departures for risk assessment (Section 3.1). The approach of using AChE inhibition data for derivation of point of departures is consistent with the advice of the SAP from 2008 in that AChE data remain the most sensitive and robust dose-response data in experimental toxicology studies. Consistent with the concept that younger juvenile animals are more sensitive due to immature metabolic systems, PND11 rat pups provide the most sensitive cholinesterase inhibition data for deriving the acute oral POD ($BMDL_{10}$ 0.36 mg/kg). As the metabolic systems mature at or near adult levels, this age-dependant sensitivity disappears. In repeated dosing studies, pups exhibit similar sensitivity to adult rats and it is pregnant dams which provide the most sensitive data for deriving a repeated exposure oral point of departure ($BMDL_{10}$ 0.03 mg/kg/d).

- *Neurodevelopmental Outcomes in Laboratory Animals:*

In the 2008 preliminary review, the Agency evaluated the neurobehavioral studies available at that time. Since then, the database of studies with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent effects into adulthood has grown (Section 3.2.2). This literature consistently shows effects related to *in utero* and/or early post-natal exposure; however, there is considerable variability in the outcomes. This literature includes a range of exposure periods, dosing scenarios, testing strategies, and specific methodologies and equipment used. Given the wide array of testing that has been conducted, some variability is not unexpected and in fact, the consistency of finding neurological effects is more striking.

Many studies report effects at a dose of 1 mg/kg/d, a dose that produces some amount of brain ChE inhibition when given directly to the pups postnatally, but may or may not alter fetal brain ChE activity when given to the dams gestationally. One study (Braquenier et al., 2010) using lower doses, administered to the dam on GD15-LD14, reported a NOEL of 0.2 mg/kg/d. The AChE inhibition studies described in section 3.1 provide robust dose-response data with which to conduct BMD modeling for derivation of points of departure, but the animal data on outcomes such as behavior do not provide dose-response data amenable to such modeling. Many studies use only one, maybe two doses, and in several cases the lower but not higher dose

level produced significant effects. Comparing a NOEL of 0.2 mg/kg/d to a repeated dosing $BMDL_{10}$ of 0.03 mg/kg/d suggests that AChE inhibition is a sensitive and protective endpoint; however, ideally studies that thoroughly evaluate neurodevelopmental effects at lower, non-inhibitory doses could be helpful. Additional studies that thoroughly characterize (time-course, dose-response) the potential for fetal brain AChE inhibition during gestational exposures and/or or pup brain ChE inhibition during postnatal exposures, could be combined with behavioral studies with which to quantitatively evaluate potential AChE inhibition compared with long-term effects.

In addition, numerous studies on the possible mechanistic aspects of neurodevelopmental effects have been published (Section 3.2.1). The results have led some research groups to propose that changes in brain connectivity and/or neurochemistry may underlie the long-term *in vivo* neurobehavioral changes observed into adulthood. While multiple biologically plausible hypotheses are being pursued by researchers, no one pathway has sufficient data to be considered more credible than the others. There is some evidence of possible effects which are similarly sensitive or more sensitive than AChE inhibition. Because of insufficient data establishing the causal linkages among different levels of biological organization to adversity, a mode of action or adverse outcome pathway leading to effects on the developing brain cannot be established. For example, while there is *in vitro* evidence relating binding of chlorpyrifos/chlorpyrifos oxon to AChE and the subsequent decrease in neurite outgrowth at the cellular level, the relationship between neurite outgrowth and neurodevelopmental consequences has not been established. As described in the 2007 NRC report on Toxicity Testing in the 21st Century, to develop an adverse outcome pathway, not only does one have to establish plausible relationships among the key events, but quantitative relationships also need to be established, *i.e.*, how much of a change in one key event is needed to result in an adverse effect at the next level of biological organization? Thus, certain exposure to a chemical may impact normal physiological responses in a way that may not necessarily be adverse and thus, the AOP concept requires an understanding of adaptive/homeostatic capacity of biological systems and their limits, relative to concentration and duration of exposure.

To follow the example above, the concentration response relationship relating the amount of chlorpyrifos/chlorpyrifos oxon binding to AChE, the inhibition of neurite outgrowth, and the level of neurodevelopmental deficit has not been established. Some molecular initiating events and some cellular effects occur below exposure levels that inhibit cholinesterase. Most of the data, however, are limited to reports from one laboratory and are not anchored to an adverse neurodevelopmental effect at the tissue or organism level.

While it is difficult to establish an adverse outcome pathway in the developing nervous system due to on-going dynamic developmental processes and multiple critical windows of development, the following experimental design aspects are critical to establishing a relationship between chlorpyrifos exposure and adverse effect.

- Concentration/dose-response relationship for each level of biological organization of the adverse outcome pathway. Ideally, this should include concentrations/doses that do not inhibit cholinesterase.
- *In vitro* studies in neural cell cultures need to be translated to effects in more complex biological systems, *i.e.*, developing, intact nervous system.
- It would be desirable to look at the molecular initiating event and the downstream events in the same biological system.

- Demonstration of a relationship between an alteration(s) of a key neurodevelopmental event at one level to that in the intact nervous system. For example, how much of a change in neuronal proliferation causes neurodevelopmental deficits?

The Agency will be soliciting comment from the SAP on the Agency's preliminary conclusions (see below) and on the strengths and uncertainties associated with the *in vivo* and *in vitro* experimental toxicology studies with a particular focus on the extent to which these studies suggest potential for long-term effects of chlorpyrifos from early-life exposure and/or the extent to which they suggest neurodevelopmental effects below doses which elicit 10% AChE inhibition.

- *Epidemiology studies in mothers and children:*

In Section 4.0 of this issue paper, EPA summarized the available epidemiologic data concerning the relation between gestational exposure to chlorpyrifos and adverse neurodevelopmental outcomes in infants and children from three major children's health cohort studies in the U.S. The currently available database expands upon those studies available at the time of the 2008 FIFRA SAP meeting on chlorpyrifos adding both new epidemiologic evaluations and new methodological studies concerning key measures in these studies. In addition, supplementary analyses which address some areas of uncertainty identified by the 2008 FIFRA SAP are also available for consideration. Overall, the newly available data support and strengthen the conclusion of the 2008 FIFRA SAP that prenatal exposure to chlorpyrifos likely plays a role in adverse neurodevelopmental outcomes measured in children. Specifically, high-quality etiologic studies from each of the children's health cohorts consistently illustrate evidence for a putative role of prenatal chlorpyrifos exposure on intelligence measured at school age (7-years). In these concurrent publications (Spring 2011), authors performed numerous model checking exercises and sensitivity analyses to reduce or eliminate alternative explanations for the findings, strengthening the overall interpretation of these data. Additionally, primarily within the Columbia Mother's and Newborn study, investigators performed exposure validation research suggesting the use of a measure of prenatal chlorpyrifos concentration in cord blood collected at delivery may be a reasonably accurate measure to categorize (rank) exposure among cohort participants. Additionally, the contribution of the assessment of infant neurodevelopment as measured by Bayley Scores in relation to prenatal organophosphate exposure by researchers with the Mt. Sinai Child Development study as well as the evaluation of effect modification by *PON1* genotype or PON1 phenotype by researchers with two of the three children's health cohorts adds to the totality of the database. Overall, epidemiology studies available since the 2008 FIFRA SAP review strengthen the conclusion that prenatal chlorpyrifos exposure likely played a role in these adverse neurodevelopmental outcomes observed in children through school-age.

While both strengths and notable limitations are present in these studies, EPA believes that the limitation present in the design, conduct, and analyses of these observational studies would likely under-estimate, rather than over-estimate measured associations (*i.e.*, measures of relative risk). Specifically, given the prospective study design many sources of error are eliminated or minimized, including temporal bias and most instances of differential measurement error. Errors in the measurement of both neurodevelopmental outcomes and exposure most likely occurred in these studies. While the magnitude of this error is difficult to

quantify, this source of error would unlikely be related to the either the outcomes or exposures measured in these studies, respectively, and therefore, would tend to under-estimate measured risks (non-differential measurement error). Alternatively, factors which may have biased the reported risk estimates away from the null (over-estimated risks) may also be present including unmeasured or poorly measured factors related to socio-economic status or other environmental exposures both of which may be positively correlated with the outcome (adverse neurodevelopmental effects) and the exposure (chlorpyrifos). Considering both supplemental analyses and methodological research published since the 2008 FIFRA SAP review, available evidence does not suggest a substantial bias of this nature; however, the possibility cannot be completely excluded.

The epidemiology studies reviewed are high-quality, well-conducted studies with numerous strengths, as well as some limitations. These limitations include use of non-specific biomarkers of chlorpyrifos exposure in some of these studies, lack of measurement of postnatal chemical exposure, and the proportion of missing data among several key variables in these investigations. There is evidence of consistent associations, of a moderately strong magnitude with some evidence of an exposure-response across many of these investigations; however, instances of inconsistency across studies of the same association are also observed, which may reflect exposure measurement variability or true inconsistency in observed effects. Currently, while biologically plausible explanations can be posited, a clear mode of action for these effects has not yet been elucidated, as discussed in detail in the paper. The lack of an established mode of action for neurodevelopmental effects is problematic for both causal inference as well as determining appropriate points of departure among other uses in quantitative risk assessment. The Agency will be soliciting comment from the SAP on the strengths and uncertainties associated with the epidemiology and its incorporation into risk and uncertainty characterization.

- *Biomonitoring data:*

Chlorpyrifos is one of a small set of pesticides for which there is a large database of biomonitoring studies available for a variety of lifestages and subpopulations (Section 5.0). Biomonitoring data provide real world information on exposed individuals; however, their interpretation is challenging given that the timing, magnitude, and source of exposures are most often unknown or only partially known. Since the 2008 SAP, there has been significant growth in the scientific literature on approaches for interpreting biomonitoring data. Such approaches range in their level of sophistication and the availability of the pertinent data to inform them (Section 5.4). In the epidemiology studies, measured exposure likely underestimates actual exposure experienced by participants, in most instances, therefore direct use of exposure estimates from epidemiologic analyses for use in establishing points of departure or modeling exposure-response associations in quantitative risk assessment is less than optimal. Therefore, exposure estimates from observational studies would likely require supplementary information or use of PBPK/PD modeling-type approach to approximate actual exposure. Given the findings of the children's health cohorts and the Columbia cohort in particular, the Agency has an interest in considering the degree to which other groups of individuals in the U.S. may be exposed to levels of chlorpyrifos comparable with those from Columbia University cohort, particularly prior to the cancellation of the indoor residential uses. The Agency will be soliciting comment from the SAP on the strengths and uncertainties associated with such approaches for interpreting biomonitoring data for use in risk characterization and uncertainty analysis. One

approach would be to use a PBPK-PD model which accounts for all relevant routes of exposure and incorporates a gestational component combined with a dose reconstruction analysis. At this time, the existing PBPK-PD models are not equipped for this sophisticated analysis. It is proposed that this is an area where additional research could significantly improve the Agency's ability to determine whether actual exposures experienced by participants in the epidemiology studies were above or below AChE inhibition concentrations.

As discussed in previous sections of this draft issue paper, two of the key scientific questions being considered in preparation for the weight of the evidence are: 1) the degree to which scientific data suggest that chlorpyrifos causes long-term neurodevelopmental effects from fetal or early life exposure and 2) the degree to which adverse effects can be attributed to doses lower than those which elicit 10% inhibition of AChE, *i.e.*, the dose levels previously used for regulatory decision making. The evaluation of these scientific questions are multi-faceted and require integration of multiple lines of data and an understanding of the nature and degree of the uncertainties around the data and impact on hypotheses or alternative interpretations. At this time, this evaluation is incomplete, and thus in the absence of a full weight of the evidence, the Agency has not yet made firm conclusions. The Agency has, however, made preliminary interpretations and conclusions on the information summarized above which will be reviewed by the SAP.

- Experimental toxicology studies in rodents suggest that long-term effects from chlorpyrifos exposure may occur. Due to the dose selections in most of these *in vivo* studies evaluating effects such as behavior and cognition, it is not known whether such adverse effects would be shown at doses lower than those used for derivation of point of departures. Despite this uncertainty, however, comparing a NOEL of 0.2 mg/kg/d to a repeated dosing $BMDL_{10}$ of 0.03 mg/kg/d suggests that ChE inhibition is a sensitive and protective endpoint.

- With respect to adverse effects in humans, the Agency has not yet developed a full weight of the evidence analysis to consider the extent to which causality can be postulated based upon all of the available evidence including the observational studies in the human population. Consistent with the 2008 SAP, however, the Agency has preliminarily concluded that the epidemiologic data support and strengthen a finding that chlorpyrifos likely played a role in the neurodevelopmental effects observed in children in association with exposure during gestation. Although actual level of such exposure during the critical window(s) of susceptibility is not known, the cord blood and other (meconium) measures from the Columbia University study provide qualitative, strong evidence of exposure to the fetus during gestation. While there are significant uncertainties remain about the actual exposure levels experienced by participants in the three children's health cohorts, particularly during the time period prior to the voluntary cancellation of indoor residential uses of chlorpyrifos containing pesticide products, and the degree to which those levels were above, at, or below those which would elicit 10% AChE inhibition, exposures measured in the range reported in the epidemiology studies (pg/g plasma) are likely low enough that is unlikely to result in AChE inhibition.

- At this time, a mode of action/adverse outcome pathway has not been established for neurodevelopmental outcomes. This growing body of literature does demonstrate, however, that chlorpyrifos and/or its oxon are biologically active on a number of processes that affect the developing brain. Although these studies are not appropriate for deriving point of departures and quantitative relationships between these perturbations and adversity is not known, it is notable that some of these effects are similarly sensitive or more sensitive than AChE inhibition.

Moreover, the lack of established mode of action/adverse outcome pathway does not undermine or reduce the confidence in the qualitative conclusions of the epidemiology studies. The context of quantitative risk assessment, the lack of an established mode of action/adverse outcome pathway complicates the way in which the epidemiological studies can be used in relation to important factors such as dose-response, critical duration of exposure, and window(s) of susceptibility.

- With respect to the key questions being considered by the Agency with this 2012 draft issue paper (*i.e*, whether chlorpyrifos causes long-term effects from fetal or early life exposure and if adverse effects can be attributed to doses lower than those which elicit 10% inhibition of AChE), when taken together the evidence from 1) the experimental toxicology studies evaluating outcomes such as behavior and cognitive function; 2) mechanistic data on possible adverse outcome pathways/modes of action; and 3) epidemiologic and biomonitoring studies:

    - Qualitatively, the Agency preliminarily concludes that these lines of evidence together support a conclusion that exposure to chlorpyrifos results in adverse neurodevelopmental outcomes in humans, at least under some (still unclear) conditions.

    - Quantitatively, the dose –response relationship of AChE inhibition across different life stages is established, but other adverse outcome pathways/modes of action are not established. The question posed regards the nature and degree of uncertainty around points of departure based on 10% AChE inhibition to protect against neurodevelopmental outcomes.

        - The database of *in vivo* animal toxicology neurodevelopmental studies on adverse outcomes includes only a small number of studies at or near the Agency's point of departure. Despite this uncertainty, the Agency's chronic oral point of departure is approximately 10-fold lower than doses used in repeated dosing studies.

          With respect to the mechanistic data, there are some effects which are similarly sensitive or more sensitive than AChE inhibition. The fact that there are, however, sparse data to support the *in vitro* to *in vivo* extrapolation, or the extrapolation from biological perturbation to adverse consequence significantly limits their quantitative use in risk assessment.

        - As noted above, the lack of an established adverse outcome pathway/mode of action makes quantitative use of the epidemiology study in risk assessment challenging, particularly with respect to dose-response, critical duration of exposure, and window(s) of susceptibility. Despite this uncertainty, the cord blood and other measures (meconium) provide qualitative, strong evidence of exposure to the fetus during gestation. Moreover, exposure levels in the range measured in the epidemiology studies (pg/g) are likely low enough that is unlikely to result in AChE inhibition; however, the actual level of such exposure during the critical window(s) of susceptibility is not known.

- There are a variety of dose reconstruction and reverse dosimetry approaches of to evaluate the extent that exposures experienced by the mothers in the Columbia cohort are potentially high enough to elicit AChE inhibition. These vary in their sophistication. One robust approach to inform the scientific question of the degree to which neurodevelopmental outcomes may/may not occur below AChE inhibiting doses used for regulatory decision making would involve reverse dosimetry and dose reconstruction using a PBPK/PD model that predicts a variety of dose metrics including AChE inhibition and blood/urinary biomarker levels and can simulate different exposure scenarios. Such an analysis would also require a more in-depth evaluation of the specific exposure conditions likely experienced during the time period of these epidemiology studies. Preferably such a PBPK/PD model could accommodate oral, dermal, and inhalation routes in addition to multiple lifestages (*e.g.*, gestation, birth, childhood, adulthood). This ideal model may not be available for the revised risk assessment. In the absence of such a model, the Agency is soliciting comments from the Panel on approaches for interpreting biomonitoring data, synthesizing data from several lines of evidence to support standard assumptions regarding exposure assessment and reconstruction, in addition to integrating exposure and hazard information more comprehensively than has been performed. In the absence of a sophisticated, life-stage appropriate PBPK/PD model, there may be other more practical options to pursue that would be fruitful for inclusion in the quantitative chlorpyrifos risk assessment.

The Agency will solicit comment on these preliminary conclusions, their strengths and uncertainties along with the data and analysis that underlie them at the April, 2012 SAP. In addition, the Agency will solicit comment on the further analyses that may be conducted to further inform the potential for long-term effects of early life exposure to chlorpyrifos and to evaluate the extent to which these potential effects may (or may not) occur at or below 10% AChE inhibition levels previously used for regulatory decision making.

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ATTACHMENT I

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF CHEMICAL SAFETY AND POLLUTION PREVENTION

July 11, 2012

MEMORANDUM

SUBJECT: Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting held April 10-12, 2012 on "Chlorpyrifos Health Effects"

TO: Steven Bradbury, Ph.D.
Director
Office of Pesticide Programs

FROM: Fred Jenkins, Jr., Ph.D. 7/11/12
Designated Federal Official
FIFRA Scientific Advisory Panel
Office of Science Coordination and Policy

THRU: Laura Bailey, MS 7/11/12
Executive Secretary
FIFRA Scientific Advisory Panel
Office of Science Coordination and Policy

Frank Sanders 7/11/12
Director
Office of Science Coordination and Policy

Attached, please find the meeting minutes of the FIFRA Scientific Advisory Panel open meeting held in Arlington, VA on April 10-12, 2012. This report addresses a set of scientific issues associated with "Chlorpyrifos Health Effects."

Enclosure

cc:
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SAP Minutes No. 2012-04

A Set of Scientific Issues Being Considered by the Environmental Protection Agency Regarding Chlorpyrifos Health Effects

April 10 – 12, 2012
FIFRA Scientific Advisory Panel Meeting
Held at the
Environmental Protection Agency Conference Center
Arlington, VA

NOTICE

These meeting minutes have been written as part of the activities of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), Scientific Advisory Panel (SAP). The meeting minutes represent the views and recommendations of the FIFRA SAP, not the United States Environmental Protection Agency (Agency). The content of the meeting minutes does not represent information approved or disseminated by the Agency. The meeting minutes have not been reviewed for approval by the Agency and, hence, the contents of these meeting minutes do not necessarily represent the views and policies of the Agency, nor of other agencies in the Executive Branch of the Federal Government, nor does mention of trade names or commercial products constitute a recommendation for use.

The FIFRA SAP is a Federal advisory committee operating in accordance with the Federal Advisory Committee Act and established under the provisions of FIFRA as amended by the Food Quality Protection Act (FQPA) of 1996. The FIFRA SAP provides advice, information, and recommendations to the Agency Administrator on pesticides and pesticide-related issues regarding the impact of regulatory actions on health and the environment. The Panel serves as the primary scientific peer review mechanism of the Environmental Protection Agency, Office of Pesticide Programs (OPP), and is structured to provide balanced expert assessment of pesticide and pesticide-related matters facing the Agency. FQPA Science Review Board members serve the FIFRA SAP on an ad hoc basis to assist in reviews conducted by the FIFRA SAP. Further information about FIFRA SAP reports and activities can be obtained from its website at http://www.epa.gov/scipoly/sap/ or the OPP Docket at (703) 305-5805. Interested persons are invited to contact Fred Jenkins, Jr., Ph.D., SAP Designated Federal Official, via e-mail at jenkins.fred@epa.gov.

In preparing these meeting minutes, the Panel carefully considered all information provided and presented by EPA, as well as information presented by public commenters.

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Chlorpyrifos Health Effects

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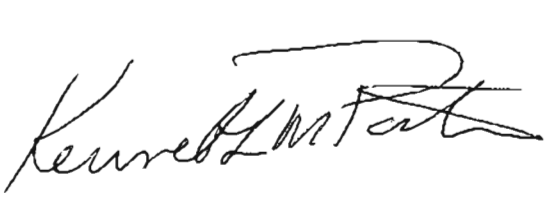
Arlington, VA



Kenneth M. Portier, Ph.D.
FIFRA SAP Chair
FIFRA Scientific Advisory Panel
Date: JUL 10 2012

Fred Jenkins, Jr., Ph.D.
Designated Federal Official
FIFRA Scientific Advisory Panel
Date: JUL 10 2012

Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP)
Chlorpyrifos Health Effects
April 10 – 12, 2012

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INTRODUCTION

Chlorpyrifos (0,0-diethyl--3,5,6-trichloro -2-pyridyl phosphorothioate) is a broad-spectrum, chlorinated organophosphate (OP) insecticide. Like other OPs, chlorpyrifos binds to and phosphorylates the enzyme acetylcholinesterase (AChE) in both the central (brain) and peripheral nervous systems. This can lead to accumulation of acetylcholine and, ultimately, at sufficiently high doses, to clinical signs of toxicity. In 2011, the Agency released a preliminary human health risk assessment for chlorpyrifos. The focus of this assessment was on the cholinesterase (ChE) inhibiting potential of chlorpyrifos. Consistent with this focus, EPA evaluated the extensive database of ChE data for multiple lifestages and selected points of departure (PoDs) based on consideration of all quality and reliable data. There is, however, a growing body of literature with laboratory animals (rats and mice) indicating that gestational and/or early postnatal exposure to chlorpyrifos may cause persistent effects into adulthood. The results of both *in vivo* and *in vitro* studies on chlorpyrifos have led some research groups to propose that changes in brain connectivity and/or neurochemistry may underlie these changes into adulthood. In addition, there are epidemiology studies evaluating pre- and post-natal chlorpyrifos or other OP exposure in mother-infant pairs that have reported associations with birth outcomes, childhood neurobehavioral and neurodevelopment outcomes in the offspring when evaluated in neonates, infants, and young children.

In 2008, the FIFRA Scientific Advisory Panel (SAP) reviewed a draft science issue paper on the human health effects of chlorpyrifos which provided a preliminary review of the scientific literature on experimental toxicology and epidemiology studies available at that time. In 2010, the Agency developed a draft "Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment" which provides the conceptual foundation for evaluating multiple lines of scientific evidence in the context of the understanding of the adverse outcome pathway (or mode of action). This draft framework uses modified Bradford Hill Criteria to evaluate the sufficiency of evidence to establish key events within a mode of action(s) and explicitly considers such concepts as strength, consistency, dose response, temporal concordance and biological plausibility.

Since the 2008 SAP on chlorpyrifos, the Agency has performed further analyses on the existing and new epidemiology results in mothers and children, available biomonitoring data, and experimental toxicology studies evaluating proposed adverse outcome pathways in the context of human health risk assessment. Specifically, the Agency is evaluating available literature on the potential for chlorpyrifos to cause long term adverse effects from early life exposure, *in vivo* and *in vitro* studies evaluating mechanistic aspects of chlorpyrifos, and the potential for adverse effects below doses established from ChE inhibition that are used for regulatory purposes. At this time, the Agency is working towards a weight of evidence evaluation integrating the epidemiology studies with the experimental toxicology studies for the neurodevelopmental outcomes. This analysis is complex and multifaceted as it involves different lines of scientific evidence (*i.e.*, *in vivo* & *in vitro* experimental toxicology studies, explicit consideration of adverse outcome pathways, exposure, epidemiology, and biomonitoring data). As such, the Agency believes that peer review on the status of the current analysis is important.

Opening remarks at the meeting were provided by:
Steven Bradbury, Ph.D., Director, Office of Pesticide Programs (OPP), EPA; Karen Whitby, Ph.D., Acting Director, Health Effects Division, OPP, EPA; Anna Lowit, Ph.D., OPP, EPA; William R. Mundy, Ph.D., Office of Research and Development (ORD), EPA; Ginger Moser, Ph.D., DABT, Fellow ATS, ORD, EPA; Carol H. Christensen, Ph.D., MPH, OPP, EPA; Lieutenant Aaron Niman, US Public Health Service, OPP EPA

Public Comments

Public comments were provided by:
Dow AgroSciences
Julie E. Goodman Ph.D. and Lorenz Rhomberg, Ph.D. of Gradient on behalf of Dow Agrosciences
Abby Li, Ph.D. of Exponent on behalf of Dow Agrosciences
Jennifer Sass, Ph.D. of Natural Resources Defense Council (NRDC)
Dale Hattis, Ph.D. of Clark University on behalf of himself

Summary of Panel Discussion and Recommendations

Charge 1: Mode of action/adverse outcome pathway: Acetylcholinesterase (AChE) inhibition

Question 1.0

It is well established that AChE inhibition is the primary mode of action/adverse outcome pathway for OPs, like chlorpyrifos. Because AChE inhibition is the initiating event for this mode of action/adverse outcome pathway, using AChE inhibition as a regulatory endpoint is protective of downstream cholinergic effects. Moreover, historically, given the sensitivity of AChE inhibition data for OPs, these data have been considered to be protective of other potential toxicities and/or modes of action for OPs. In 2008, the Agency performed a comprehensive review of the available AChE data from multiple lifestages. This review has been supplemented with the newest studies. Consistent with the recommendations from the 2008 SAP, the Agency believes that AChE data remain the most robust dose-response data for deriving points of departure in *in vivo* experimental toxicology studies with laboratory animals. *Please comment on the Agency's preliminary conclusion that AChE data remain the most robust source of data for deriving points of departure for chlorpyrifos. Please include a discussion of the strengths and uncertainties of this preliminary conclusion.*

The Panel concurs with the Agency's position that AChE data continue to be the strongest resource of data for deriving points of departure for chlorpyrifos. The Panel's conclusion is based on the premise that all studies reporting neurobehavioral changes following *in vivo* prenatal or postnatal exposures to chlorpyrifos have been accompanied by AChE inhibition when measured at an appropriate time following administration of chlorpyrifos.

The Panel additionally notes that studies evaluating neurodevelopmental effects entailed experimental designs that do not permit an efficient means of determining a point of departure for chlorpyrifos. Thus, just as the in the 2008 SAP, this Panel advises that the Agency continue to use AChE data at the most sensitive lifestages for dose-response analysis and deriving points of departure. Also in keeping with the 2008 SAP, this Panel expresses concern about the use of Dimethyl Sulfoxide (DMSO) as a vehicle because of its intrinsic toxicity, its potential influence on absorption and interaction with chlorpyrifos, and the impact of this interaction on the developing organism.

Charge 2.0: Mode(s) of action/adverse outcome pathway(s): Plausible pathways leading to potential neurodevelopmental outcomes

Question 2.1

As discussed in Section 3.2.1, although there are numerous mechanistic studies in the scientific literature, the research on different hypotheses does not provide sufficient data to establish causal linkages among different levels of biological organization to show how effects lead to adversity. As such, a mode of action or adverse outcome pathway leading to effects on the developing brain cannot be established at this time. Moreover, although multiple biologically plausible hypotheses are being pursued by researchers, based on the current state of the science, no one pathway has sufficient data to be considered more credible than the others. *Please comment on the Agency's preliminary conclusion that although there are multiple biologically plausible hypotheses being evaluated by research scientists, the mechanistic experimental toxicology data do not yet support a coherent set of key events in a mode of action/adverse outcome pathway.*

The Panel agrees with the Agency's conclusion that based on the current state of the science, no one pathway has sufficient data to be considered more credible than the others with respect to a causal link between chlorpyrifos exposure and neurodevelopmental outcome.

In regard to the Agency's case study demonstrating domoic acid's adverse outcome pathway, the Panel contends that the linear connections of the pathway demonstrated in this case study appear likely to be rare and unique. They also note that it is more likely that other such neurotoxicological pathways are non-linear. Expectations of a linear pathway specifically in the case of chlorpyrifos may be artificially elevated and potentially unrealistic for risk assessment.

Question 2.2

Although a mode of action/adverse outcome pathway has not been established, qualitatively, the growing body of mechanistic studies does demonstrate that chlorpyrifos and/or its oxon are biologically active on a number of processes that affect the developing brain. Some mechanistic studies provide evidence of possible effects which are similarly sensitive or more sensitive than AChE inhibition (*e.g.*, neurite outgrowth, binding to muscarinic receptors, axonal transport; serotonergic nervous system development). Some of these comparisons must be considered with caution since the amount of change in the *in vitro* systems required to elicit an adverse effect *in vivo* is unknown. Moreover, extrapolation from *in vitro* perturbations to *in vivo* effects has not been established, which introduces additional uncertainties. *Given the doses/concentrations evaluated in the in vitro and in vivo mechanism studies, please comment on the degree to which these studies suggest that endpoints relevant to evaluating potential neurodevelopmental outcomes may or may not be more sensitive than AChE inhibition. Please include in your comments a discussion of the strengths and uncertainties. Please also include in your comments a discussion of the scientific understanding of dose-response relationships for biological perturbations and the magnitude, frequency and/or duration of such perturbations that can lead to adverse effects at higher levels of biological organization*

to support characterization of the likelihood of adverse outcomes in a human health risk assessment (as articulated in NRC 2007).

The Panel concurs with the Agency that caution should be applied in interpreting the *in-vivo* significance of the changes observed across the various *in vitro* studies. Several uncertainties and limitations are associated with the translation of *in vitro* study results to *in vivo* effects. The inherent complexity of the nervous system presents significant challenges to accomplishing this translation. An additional example of uncertainty is that cells that are isolated in culture within an *in vitro* experiment may be affected differently than they would if they were within their *in vivo* environment.

The Panel recommends continued literature review and analysis of published data with the goal of developing additional hypotheses linking *in vitro* findings to *in vivo* relevance. As an example, the analytical studies of the Lockridge group indicating that chlorpyrifos oxon can covalently modify key cytoskeletal proteins such as tubulin and motor proteins like kinesin, provide information that can contribute to the interpretation of findings of alterations in neurite outgrowth and axonal transport, respectively.

The Panel also recommends that the Agency consider other areas that might be added to the review such as the effect of chlorpyrifos on neurotrophins (growth factors). Several researchers have found early evidence of the potential for these effects (Pope *et al.,* 1995; Slotkin *et al.*, 2007; Betancourt and Carr, 2004).

The Panel cautions the Agency concerning their examination of the dose-response relationships. They particularly note that when evaluating these relationships, pharmocodynamic (PD) analyses should not be uncoupled from pharmacokinetic (PK) models given that PK differences can affect active site concentrations and hence, PD effects. Thus, PK models can significantly affect the magnitude and duration of an effect.

Lastly, the Panel raises concerns about the equivalency of developmental stages between ages of rodents to human. These are not well defined with regard to cell type compositions, brain region, cellular architecture, and physiological or biochemical processes. This lack of equivalence further limits the translation to the *in vivo* situation and the ability to provide a quantitative dose-response relationship that can be compared to that for AChE inhibition.

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Charge 3.0 Neurodevelopmental data from laboratory animals

Question 3.1

As discussed in Section 3.2.2, the experimental toxicology data in laboratory rodents show neurobehavioral effects following developmental exposure with changes in a number of neurological domains. In 2008, the SAP agreed to this preliminary conclusion, and the nine additional studies available since 2008 add further support. *Please comment on the degree to which these studies show changes in a number of neurological domains and support the qualitative conclusion that chlorpyrifos exposure during gestation and/or early post-natal period may result in long-term adverse effects on the developing nervous system. What evidence does and does not support this conclusion? Please also include in your comments a discussion of the strengths and uncertainties. Please also include in your comments a discussion of the scientific understanding of dose-response relationships for biological perturbations and the magnitude, frequency and/or duration of such perturbations that are can lead to adverse effects at higher levels of biological organization to support characterization of the likelihood of adverse outcomes in a human health risk assessment (as articulated in NRC 2007).*

The Panel agrees with the 2008 SAP conclusions that developmental neurobehavioral studies demonstrate adverse effects from chlorpyrifos exposure. However, the number of available neurobehavioral studies is limited leading to caution concerning this finding. Also many of these studies are statistically under-powered and prone to Type I errors and should be discounted in formulating the weight of evidence for or against neurobehavioral effects from developmental exposure to chlorpyrifos. The Panel also expressed caution with the significance of some of the experimental neurotoxicological outcomes that have not been validated. These included the tests of anxiety, depression, and social interactions. The Panel recommends these experimental outcomes be regarded as exploratory, and hypothesis-generating, as opposed to being evidence of toxicity. The lack of specificity in the direction of the neurobehavioral dose response findings is a problematic issue.

Despite the issues raised by the Panel about these studies, the overall evidence across these studies is persuasive in indicating that there are enduring effects on the Central Nervous System (CNS) from chlorpyrifos exposure at or above 1.0 mg/kg. The Panel recommends that future neurodevelopmental studies be focused on testing chlorpyrifos levels below 1.0 mg/kg/day and that these studies be geared towards identifying the correct testing paradigm and neural substrates for detecting possible effects. The Panel advises that cross-laboratory or collaborative studies may provide systematic comparison of the effects of chlorpyrifos on neurodevelopmental domains using unified exposure periods, dosing, age of testing, and methods, combined with urinary analysis of chlorpyrifos' metabolites, and accurate assessments of AChE inhibition.

Question 3.2

The dose-response data in the *in vivo* experimental neurodevelopmental toxicity studies are not amenable to empirical dose-response modeling as many studies use only one or two doses, and in some cases the lower dose, but not higher dose level, produced significant effects. Many studies report effects at a dose of 1 mg/kg/d-- a dose that produces some amount of brain ChE inhibition when given directly to the pups post-natally, but may or may not alter fetal brain ChE activity when given to the dams gestationally. One study (Braquenier *et al.*, 2010) using lower doses, administered to the dam on GD15-LD14, reported a NOEL of 0.2 mg/kg/d. Comparing the NOEL of 0.2 mg/kg/d to a repeated dosing AChE inhibition $BMDL_{10}$ of 0.03 mg/kg/d suggests that AChE inhibition is a sensitive and protective endpoint.

a. *Please comment on the scientific quality and robustness of the animal neurodevelopmental toxicity studies.*

The Panel notes that the quality of these studies vary. The "high quality" studies use multiple doses, adequate sample sizes, controls for litter effects, sound behavioral methods, and appropriate statistical methods to analyze the data. Since all these studies demonstrate long-term neurobehavioral effects, the data generated by them (especially the findings that occurred at doses $\geq$ 1 mg/kg of chlorpyrifos) can be considered robust. The Panel has some concerns even with the high quality studies. For example, the rat strains used in some of the studies are considered by the Panel to be less than preferable. The Panel advises that studies that are considered to support regulatory decisions should be those that use a mainstream rat strain such as Sprague-Dawley from Charles River or Harlan because much more is known about their behavioral characteristics and they do not perform at the extremes of the distribution.

Despite the concerns expressed about the studies, the Panel concurs with the conclusions of the 2008 SAP findings and the EPA White Paper background document, and concludes that the collective weight of evidence from these studies demonstrate that it is probable that there are significant long-term adverse effects from chlorpyrifos exposure.

b. *Please comment on the degree to which studies that measured AChE inhibition and those that measured neurodevelopmental outcomes can be integrated to evaluate whether points of departure based on 10% AChE inhibition provide more sensitive endpoints than endpoints measured in the experimental neurodevelopmental studies (as reviewed in Section 3.2.2). Please include in your comments a consideration of the strengths and uncertainties associated with this assessment.*

The Panel concludes that since AChE inhibition recovers quickly the data are insufficiently refined to allow for a linkage between the mode of action and the neurodevelopmental effects (acute vs. chronic, respectively). They note that since the mode of action of these effects is not established and cannot be presumed to be related to AChE inhibition, these studies do not exclude the possibility that other mechanisms may

be involved, especially concerning long-term effects that may be unmasked at later life-stages. Additionally, since the neurodevelopmental effects may be independent of AChE inhibition, the Agency should consider whether AChE inhibition represents the critical marker for derivation of points of departure for chronic studies.

Charge 4.0 Epidemiology Regarding Children's Health

Question 4.1
Section 4.0 and Appendices 5 and 6 provide the Agency's review of the available epidemiology studies from the Columbia Mothers and Newborn study, the Mt. Sinai Child Development study, and the Center for Health Assessment of Mothers and Children of Salinas Valley (CHAMACOS) study. Consistent with the 2008 SAP recommendations, the Agency has considered information offered from each of the three cohort investigations; however EPA acknowledges the primacy of the Columbia cohort data for the purposes of informing risk assessment because researchers measured chlorpyrifos parent compound directly in this study. *Please comment on the sufficiency, clarity, and quality of the Agency's epidemiology review as contained in Section 4.0 and Appendices 5 and 6 of the draft issue paper with respect to identifying the major strengths and limitations of each study.*

The Panel considers the Agency's epidemiology review to be very clearly written, accurate, and to generally provide a very thorough review of the epidemiology literature. In addition, the Panel commends the Agency for putting their epidemiology review in the context of the modified Bradford Hill criteria, as recommended by the 2010 SAP. The Panel believes that the epidemiology review appropriately concludes that the studies show some consistent associations relating exposure measures to abnormal reflexes in the newborn, pervasive development disorder at 24 or 36 months, mental development at 7-9 years, and attention and behavior problems at 3 and 5 years of age, in addition to less consistent results for reduced mental and psychomotor development at 12 and 24 months. Inconsistent results are found for associations between exposure and measures of fetal growth.

The Panel views the Agency's epidemiology review as an excellent description of the strengths and limitations of the studies conducted to examine the relation of chlorpyrifos to children's growth and neurodevelopment. It is noted that studies of this nature are logistically difficult to implement because of the potentially large burden imposed on the study participants in terms of time and effort, often with little or no specific benefit to the participants.

Although in agreement with the Agency that chlorpyrifos could have played a role in the neurodevelopmental outcomes observed in the Columbia cohort, some panel members expressed concern about associating the observed deficits in neurodevelopmental outcomes in children with a single chemical. This is because the studies entail a multi-chemical exposure spanning a multi-year period that encompasses an important period of sequential developmental processes necessary for brain maturation. Thus, panel members caution that it is very difficult to attribute the independent physiological effects to a single chemical in this type of multi-chemical exposure scenario. An additional

concern raised by the Panel is the modest sample sizes of the studies. They deem inadequate sample size as one of the most important limitations of these studies.

Question 4.2

Similar to the initial conclusions from 2008, the Agency has preliminarily concluded that, qualitatively, chlorpyrifos likely played a role in the neurodevelopmental outcomes reported in the epidemiologic studies, and that information available since 2008, including both new etiologic investigations as well as epidemiologic methods papers, strengthens this conclusion. *Please comment on the Agency's preliminary, qualitative conclusion that chlorpyrifos likely played a role in the neurodevelopmental outcomes observed in the epidemiologic studies. Please include in your comments a discussion of the strengths and uncertainties associated with this preliminary conclusion.*

Overall, the Panel concurs with the 2008 SAP and the Agency in concluding that chlorpyrifos likely plays a role in impacting the neurodevelopmental outcomes examined in the three cohort studies. Although exposures to other AChE-inhibiting compounds cannot be excluded as contributing to neurodevelopmental (adverse) outcomes, the potential combination and/or additive effects of these compounds does not rule out the role of chlorpyrifos. As a result, it cannot be concluded that chlorpyrifos is the only contributor to the observed outcomes.

The strengths of the three studies support the Panel's conclusion. There are nine strengths identified by the Panel which are discussed in the detailed response section of this report. Some of the strengths noted include, but are not limited to: 1) the longitudinal designs which permit clear indications of the temporal relation of chlorpyrifos exposure to adverse neurodevelopmental outcomes, 2) the inclusion of biomarkers of exposure as well as self reported exposure, and 3) the relative consistency of findings in different populations while using similar standardized exposure and outcome measures.

Question 4.3

As discussed in Question 2.0, a mode of action/adverse outcome pathway has not yet been fully elucidated for the potential neurodevelopmental outcomes as a result of prenatal chlorpyrifos exposure. Although this does not undermine the qualitative interpretation of these studies, and the preliminarily conclusion stated above (Question 4.2), the identification of the dose-response for neurodevelopmental effects based on mode of action is not possible. Further, given the urine and cord blood sampling frequency in the study there is a large degree of uncertainty in estimating absolute exposure-response relationships, as opposed to establishing relative exposure groups for evaluating associations. With respect to dose-response, critical durations of exposure, and windows of susceptibility are unknown. In 2008, the SAP cautioned against using the Columbia cohort data for deriving a point of departure due, in part, to only measuring biomarkers (3rd trimester maternal, cord blood, meconium) at one point in time, and because they cannot exclude possibility that the effects seen were due to chlorpyrifos in combination with other pesticides. In 2008, the SAP advised against using data from the epidemiology studies (including the Columbia Mothers and Newborn study which measured chlorpyrifos directly) for deriving a point of departure due to limitations of the

exposure assessment in these epidemiology studies for the purpose of risk assessment, *e.g.*, lack of repeated exposure estimates to ascertain more specifically the variability and periodicity of exposure over time (*i.e.*, predominant use of one-time exposure estimate).

Question 4.3

a. Due to the limitations of exposure assessment performed in the epidemiologic investigations for the purposes of quantitative risk assessment, the Agency has concluded that the epidemiologic data are not sufficient for deriving points of departure for quantitative risk assessment. The Agency proposes that AChE inhibition data from laboratory animals remain the most appropriate data to use for dose-response modeling and the derivation of points of departure. *Please comment on the scientific evidence that does and does not support this conclusion, as well as the strengths and limitations of the evidence.*

The Panel recognizes the limitations of estimating chlorpyrifos exposures based on the exposure measures collected in the three longitudinal children's cohort studies (*i.e*., the Columbia study, the Mt. Sinai study, and the CHAMACOS study). Consequently, the Panel largely concurs with EPA that the data generated from these studies alone are not adequate enough to obtain a point of departure (POD) for the purposes of quantitative risk assessment.

However, despite the limitations of the exposure assessment of these three cohort studies, the Panel recognizes the significance of these data, and advises the Agency to explore additional ways of using these studies, especially the data from the Columbia study, to inform the dose response assessment of chlorpyrifos. This recommendation is underscored by the Panel's concerns regarding the proposed use of dose-response data on AChE inhibition in laboratory animals to derive points of departure for the chlorpyrifos risk assessment. The Panel notes that multiple lines of evidence suggest chlorpyrifos can affect neurodevelopment at levels lower than those associated with AChE inhibition. These multiple lines of evidence include: 1) the collective findings of the three cohort studies, 2) *in vivo* animal neurodevelopmental studies summarized in the Draft Issue Paper that report differential expression of oxidative stress genes and altered serotonergic tone in rat brain associated with early life chlorpyrifos exposures at doses below which acetylcholinesterase inhibition was detected, and 3) several *in vitro* mechanistic studies reported in the Draft Issue Paper demonstrating interference with neurite and axon outgrowth, reduced axonal transport, and increased oxidative stress in a variety of cell types exposed to chlorpyrifos concentrations that do not or are not expected to inhibit AChE. As noted in the response to Charge Question 2.2, additional evidence comes from studies not included in the Draft Issue Paper, reporting effects of chlorpyrifos on nerve growth factors and mitochondrial morphology at concentrations below which acetylcholinesterase inhibition is expected.

The Panel recommends that the Agency consider developing a functional physiologically based pharmacokinetic (PBPK) model for chlorpyrifos for pregnancy and the prenatal lifestage. The PBPK model could be utilized for additional dose-response analyses to further characterization of the dose estimates in the epidemiology studies. This model

could also become important in the future if the Agency decides to transition from using AChE inhibition to another outcome.

The Panel suggests additional research that may answer the key question of whether chlorpyrifos induces neurodevelopmental effects in humans at doses that do not cause AChE inhibition. More specifically the Panel suggests conducting studies that test whether red blood cell or brain AChE inhibition occurs as a result of chlorpyrifos concentrations in cord blood being associated with neurodevelopmental effects.

Additional Panel concerns about the use of AChE inhibition dose-response data to protect against neurodevelopmental effects is based on the potential for AChE inhibition and adverse neurodevelopmental effects to be two separate events. AChE inhibition is the result of an acute exposure scenario and neurodevelopmental effects likely being caused by chronic low level exposure to chlorpyrifos *in utero*.

Lastly, the Panel cautions the Agency dose-response data for AChE inhibition by chlorpyrifos in the pregnant rat may not be predictive of AChE inhibition in the human fetus given known interspecies differences in CYP450 isoforms, substrate affinities, fetal expression levels, and degree of polymorphism.

Question 4.3

b. The Agency does, however, believe that the epidemiologic data are useful to informing other key aspects of the chlorpyrifos risk assessment including hazard characterization, exposure characterization, and quantitative uncertainty characterization and analysis. *Please suggest approaches/analyses for potentially using the epidemiology data in different parts of the chlorpyrifos risk assessment including those noted above.* (Note: Some of these may also be covered in Question 5.4 below.)

The Panel agrees that the epidemiologic data are useful to inform key aspects of the chlorpyrifos risk assessment including exposure characterization, hazard characterization, and quantitative uncertainty characterization and analysis.

In regard to the exposure characterization, the Panel notes that environmental monitoring and biomonitoring data in these epidemiology studies can contribute to the overall database on estimation of exposure, including (particularly) population variability. These data can also enable the Agency to characterize exposure levels over time among diverse populations including production workers, agricultural workers, individuals exposed via residential use, general population, *etc*.

With respect to toxicological hazard characterization, the Panel suggests that these data can serve as the key source of support for the identification of prenatal exposures to chlorpyrifos as a cause of neurodevelopmental effects in humans. These data have many strengths. First there are consistencies in the findings of neurodevelopmental effects across the three cohort studies. Second, the levels of chlorpyrifos exposure experienced in these cohorts are comparable and well-characterized, being based on biomonitoring

(blood and urine measurements of chlorpyrifos, metabolites, *etc*.) and environmental monitoring measures (*e.g*., personal air monitoring in the Columbia study) and having similar levels observed in data collected from other studies of the general U.S. population (*e.g*., National Health and Nutrition Examination Survey (NHANES), for similar time periods (*i.e*., pre- and post-cancellation of residential uses).

In reference to the epidemiology data being used to support the quantitative uncertainty characterization and analysis, the Panel agrees with the 2008 SAP suggestion that at a minimum the Agency should use available data from these studies to at least "bound" reference doses developed on the basis of animal data. Given the potential significance of the epidemiological findings, the Panel advises the Agency to consider the potential impact of factors of study design and interpretation to bound the dose-response relationship from the human studies. For example, it would be useful to consider systematically (and at least semi-quantitatively) the potential impact of exposure measurement error, outcome ascertainment, confounding variables and statistical analysis methodology on the reported dose-response analysis.

To increase the confidence in the selected point of departure, the Panel recommends a simple experimental protocol to determine whether chlorpyrifos levels measured in the cord blood in the Columbia study inhibit either red blood cells or brain AChE. The results of such an exercise could potentially contribute to the essential question of whether or not a causal association between chlorpyrifos exposure and neurodevelopmental effects in the absence of AChE inhibition is plausible for humans.

Charge 5.0 Exposure Profile & Biomonitoring Research

Question 5.1

a. Section 5 of the draft issue paper presents an overview of the principal chlorpyrifos biomarkers and a comparison of biomonitoring studies that measured urinary TCPy levels in a range of study populations involving both the general population and potentially vulnerable populations, including children, workers, and farm families. *Please comment on the degree to which the Agency identified the primary chlorpyrifos biomarkers of exposure, appropriately discussed the strengths and limitations of such biomarkers, and how the strengths and limitations affect the interpretation of the chlorpyrifos biomonitoring data.*

The Panel notes that the Agency was thorough in its coverage of the literature on biomarkers of chlorpyrifos exposure. The Panel recommends that chlorpyrifos in blood be the first choice for a biomarker, particularly because of its specificity, the availability of standard methods for measuring it, the relevance of its concentration levels, and the number of laboratories that are capable of conducting the measurements. However, the Panel acknowledges that this is a most challenging assay, and has been used in only a small percentage of published research. The second biomarker of choice is 3,5,6-trichloro-2-pyridinol (TCPy) followed by diethylthiophosphate/diethylphosphate (DETP/DEP), both measured in urine. These have roughly the same equivalence and

neither is equivalent to measuring chlorpyrifos directly in blood because of the frequent presence of these environmental degradates of the active ingredient. Total DAPs (as DMP and DEP) are not selective enough to be a useful biomarker for chlorpyrifos although DAPs may be more appropriate in a global risk calculation model when all AChE inhibiting chemicals are considered together when evaluating risk.

The Panel suggests that more importance should be afforded to the direct intake of TCPy which is mainly present in foods. A growing body of research developing since the 1990s, has established the significance of this factor.

The Panel also acknowledges the capability of measuring AChE and BuChE as biomarkers of exposure. However, inhibitions of these enzymes are even less specific than DAPs, although they are more indicative of potential health risk. Unfortunately, the ability to measure small changes in these enzymes differs widely among laboratories and among study designs.

The Panel also recommends including in future considerations the phase II conjugation products of chlorpyrifos (namely, glucuronidase and sulfonates). Quantifying the conjugative metabolism will ensure that levels of biomarkers are correctly interpreted with respect to biomonitoring data and for performing reverse dosimetry.

b. Section 5 of the draft issue paper compares biomonitoring findings from the three children's health cohorts with other major observational exposure studies in the United States. Based on comparison with NHANES 2001-2002, median TCPy levels in the CHAMACOS and Mount Sinai cohorts were slightly higher than in the general population. It should be noted that the exposures experienced by the CHAMACOS and Mount Sinai cohorts overlapped the start of the residential chlorpyrifos phase-out. By contrast, median TCPy levels in the Columbia cohort, for which sampling occurred when chlorpyrifos use should have rapidly declined due to the voluntary cancelation, were slightly lower than the levels measured by NHANES in the general population. *Please comment on the adequacy of the Agency's comparison for the purposes of evaluating chlorpyrifos exposure levels in the three children's health cohorts. Are there any additional biomonitoring studies that should be included in the Agency's comparison?*

The Panel concurs with EPA that the human studies discussed in this section are currently the best available, primarily because they are carefully designed and well executed. The Panel recommends the following additional biomonitoring studies listed in the ordered that they should be considered: 1) NHANES 1999-2004, 2) Barr *et al*. 2010, 3) Bradman *et al*. 2005 (because the families studied are likely to continue to see significant exposure which should be validated by the next round of NHANES data), 4) the Children's Pesticide Exposure Study (CPES) by Lu et. al. 2008 and Children's Post-Pesticide Application Exposure Study (CPPAES) studies, and 5) studies that are either currently in process or completed and will be published soon. This last group includes: 1) The Children's Pesticide Exposure Study that focuses on dietary intake of children and related pesticide exposures being conducted at the Harvard School of Public Health. 2) The

Children's Exposure to Environmental Pesticides study which is evaluating the utility of biomarkers of pesticide exposures, *e.g.*, DAPs and pesticide-specific markers of OP and pyrethroid exposures, and environmental levels measured in soil, house dust, and food being conducted by Emory University, and 3) the SAWASDEE cohort study, that is examining pesticide biomarker concentrations in pregnant mothers and similar markers in their newborn children, run by Emory University and Chiang Mai University in Northern Thailand. The Panel advises that in comparing the results among these different studies, it is important to verify that analytical results from the studies are directly analogous, especially in the methods used to control for the effect of small day-to-day variations in a laboratory's AChE results when trying to quantify small changes in an exposed population.

Question 5.2

In Section 5.0 of the draft issue paper, the Agency summarized the 2008 preliminary findings on the association between urinary TCPy levels and AChE/BuChE inhibition and discussed two recent studies involving manufacturing workers in the US and Egypt. *Please comment on the scientific quality of these studies and their findings. Please include a discussion of their strengths and limitations. Please comment on the strengths and limitations of the evidence from this research to show an association between TCPy and AChE/BuChE inhibition at exposure levels experienced by occupational populations.*

The Panel notes that both studies were in general well designed and implemented. They both have adequate power to demonstrate an association between TCPy and AChE or/and BuChE inhition at exposure levels experienced by occupational populations. In addition the cholinesterase data from the studies verify the ability of the PBPK model to predict that once chlorpyrifos is absorbed, it interacts first with BuChE and only starts to inhibit RBC AChE and AChE in the central nervous system after BuChE is more than 50% inhibited.

The Panel points out several primary weaknesses in both studies as it relates to using them within the weight of evidence paradigm. These include high levels of TCPy in pre-exposure samples indicative of prior exposure to chyorpyrifos or environmental degradates either from food or accumulated residues in the workplace. Neither study reported analyses that adjusted for levels in control groups.

The Panel recommends that the Agency separate the scenarios for occupational exposures, as reported in these two studies, from exposures to residential sources. The extrapolation of these data to the population as a whole is subject to criticism. The subjects in these two studies are adults and issues such as the "healthy worker effect" and the notion that low-level exposure and high-level exposures are likely to be detoxified by differing mechanisms make extrapolation difficult. Additionally, the Panel suggested that studies of agricultural workers and their families could provide a better avenue of investigation that compares "occupational-levels" exposure with other members of their families likely to see slightly "elevated" but lower levels of exposure, and to study the potential impact on the offspring in such cohorts either *in utero* or otherwise. The Panel

recommends that in the future, the Agency consider the quality of ChE measurements before pursuing further uses of these data in exposure and risk assessments.

Question 5.3

Several approaches ranging from qualitative to the most sophisticated PBPK/PD modeling approach were introduced as potential options for analyzing the chlorpyrifos biomonitoring data. *Please comment on the strengths and limitations of these approaches. In addition, please suggest, if appropriate, alternative approaches or analyses not identified by the Agency.*

There are a rising number of data-informed options for interpreting biomonitoring data. Choosing the adequate option relies on the extent of the data available, on the toxicokinetics of the relevant population subset, on the mode of action, and on the integration of these data. Integrating these data through a verified PBPK model has the potential to be the most informative approach while also being the most data intensive.

The Panel advises EPA that at the very least the chlorpyrifos biomonitoring data should be utilized as a means of "ground truthing" total external exposures under a variety of use conditions. Considering the availability of chlorpyrifos biomonitoring data on the general population, and as a basis to support its maximal consideration of public health, the Panel also recommends that the Agency seriously consider developing a "biomonitoring equivalent" at the same time the reference dose for chlorpyrifos is derived. The biomonitoring equivalent is defined as a calculated level of a biomarker associated with exposures consistent with health protective guidance values for the general population. The Panel also recommends that the Agency utilize a verified PBPK model which will provide a robust opportunity to integrate the considerable available data on external and internal exposures (*i.e.*, biomonitoring) to chlorpyrifos at different life stages under different conditions of exposure. With respect to a specific PB/PK model for the Agency to consider, the Panel recommends a sophisticated model such as the SimCYP pediatric model (SimCYP Company, Sheffield, UK) for children that is currently available.

Question 5.4

Characterization of chlorpyrifos exposure experienced by women in the Columbia cohort, particularly during the pre-cancellation period, remains an important uncertainty in using these data in quantitative risk assessment. Exposure levels in the range measured in the cord blood data from the epidemiology studies (pg/g plasma) are probably low enough that is unlikely that the cohort mothers were experiencing AChE inhibition at the time of delivery; however, the biomonitoring data were taken after birth and not necessarily associated in time with an application of chlorpyrifos. As such, the actual level of such exposure particularly during any critical window(s) of susceptibility is not known, and a better understanding of the range of possible exposures and the degree to which they may or may not have elicited inhibition of AChE, remains a key scientific question. *In light of Panel discussions of Questions 4.3 and 5.3, please suggest approaches and/or analyses which would inform the understanding of the degree to which exposure levels experienced by the Columbia cohort participants may or may not have been below doses*

which result in 10% inhibition of AChE in the most sensitive lifestage. Please discuss the strengths and uncertainties associated with such analyses. Please include in your discussions approaches involving chlorpyrifos and its metabolites and also chlorpyrifos plus other AChE-inhibiting pesticides (propoxur, diazinon) which the cohort participants were exposed too.

The Panel notes that it is important to realize that the short half-life of chlorpyrifos and its metabolites in the body calls into question any "spot data" that might be used. Large cross-sectional studies may capture some exposure but they do not put these exposures into context. Longitudinal investigations with frequent samplings are more likely to provide data that are more useful. Thus, the Panel recommends that a longitudinal study with measurement throughout the pregnancy (rather than a few samples in the last trimester) would fill many of the data gaps that currently exist for this group. Such a study is needed given the potential for neurodevelopmental effects on the fetus as well as the metabolic differences in pregnant women versus the workers from the 1984 study.

As discussed in the response to the previous question, the Panel again recommends that a more sophisticated PBPK model may provide better data particularly if the model is pertinent to the population being studied, *i.e.* pregnant women and small children.

Studies discussed in Question 5.1 provide data on the concentration of chlorpyrifos in various media (*i.e*. house dust, air and water) while market basket data exists on the concentration of chlorpyrifos on food. These data provide the main tools for developing an effective exposure assessment and a subsequent reconstruction of potential dose. Dose reconstruction can be used to evaluate the efficacy of the PBPK model since its prediction of excretion rates can potentially be validated with an accurate estimate of dose. This assessment of the PBPK model through reconstructed dose may bridge some of the data gaps in assessing risk by validating the PBPK model.

The Panel discusses the issue of mixtures of chlorpyrifos + Diazinon /chlorpyrifos + Propoxur or chlorpyrifos/Propoxur/Diazinon. The Panel recommends that the Agency address the following questions: "Do mixture components affect each other's half lives, distributions and clearance through metabolic competition?" "Are the net AChE effects of mixtures additive or multiplicative?, and "Do they share mechanistic pathways?"

Lastly, the Panel expresses concern over the Agency's focus on a 10% AChE activity reduction. They point out that to their knowledge there is no proposed mechanism whereby a 10% AChE activity reduction in pregnant women would be responsible for a cognitive defect or developmental delay in their offspring.

Charge 6: Characterizing the range of potential risks.

The 2009 NRC report, *Science and Decisions*, focused on improving the *technical analysis* through the development and use of scientific knowledge and information to promote more accurate characterizations of risk, and thus improving the *utility* of risk assessment for risk-management decisions. The NRC report also pointed out that regulatory risk assessment does not routinely approach public health and environmental problems by arraying a wide range of options for dealing with them. *In the case of chlorpyrifos, in light of the discussions of Questions 1-5, please provide guidance for assessing and presenting the range of plausible responses at given doses, and the effect of the overall uncertainty and variability around that range.*

With regard to characterizing the probable response at given doses, the Panel recommends that the Agency use the dose-response data to establish multiple points of departure. For instance, it would be informative for risk management purposes to fully characterize the nature of the risk above the reference dose. The Panel also recommends that the Agency maximizes its use of available data on dose response from the epidemiology studies as a basis to at least "bound" reference doses developed on the basis of points of departure from animal data. As advised by the Panel, options for dose-response analysis for acute effects should be considered independent from those based on long term exposures, *i.e.*, measures representing acute adverse neurological outcomes (ChE inhibition) commonly associated with occupational exposure versus those potentially related to lower level long term exposure in the general population, such as neurobehavioral disorders.

The Panel suggests that the Agency focus on the data of chlorpyrifos in the cord blood as a means for creating the point of departure for chronic exposures to chlorpyrifos based on the PBPK/PD model.

Another consideration that the Panel deems important is the degree to which epidemiological data on neurotoxicity is consistently used within EPA to establish points of departure for chemicals with the potential for neurotoxic outcomes (*e.g.*, mercury and lead). The Panel asks: "How is the evidence from epidemiological studies weighted in the assessment for these compounds, and how does this compare with what is proposed for chlorpyrifos?"

DETAILED PANEL DELIBERATIONS AND RESPONSE TO CHARGE

It is well established that acetylcholinesterase (AChE) inhibition is the primary mode of action/adverse outcome pathway for organophosphorus chemicals (OPs) such as chlorpyrifos. In June 2011, consistent with the recommendations from the Scientific Advisory Panel (SAP) in 2008, the Agency performed a risk assessment utilizing AChE inhibition data in laboratory animals for deriving points of departure and for dose-response analysis as the Agency believes these data remain the most robust and most sensitive information available for regulatory risk assessment. However, newer lines of research on chlorpyrifos such as epidemiological studies in mothers and children, have posed the issue of whether AChE inhibition is the most sensitive health outcome, leading to questions about the chlorpyrifos risk assessment.

In order to determine the degree to which these recent studies are appropriate for incorporation into risk assessment (qualitatively and/or quantitatively), the Agency is taking a stepwise, objective and transparent approach to evaluate and interpret all the lines of scientific information related to the potential for adverse neurodevelopmental effects in infants and children as a result of their prenatal exposure to chlorpyrifos, as well as to characterize thoroughly the strengths and uncertainties associated with these studies. The issue paper entitled "Scientific Issues Concerning Health Effects of Chlorpyrifos" extends the Agency's September 2008 review of the available experimental toxicology and observational epidemiology data. This 2012 review incorporates experimental data available since the time of the last review relating to AChE inhibition and both cholinergic and non-cholinergic adverse outcomes, including neurodevelopmental studies on behavior and cognition effects. Similarly, the Agency also performed a more in-depth analysis of the epidemiologic studies from three major children's health cohort studies in the U.S., plausible hypotheses on modes of action/adverse outcome pathways (MOA/AOP) leading to neurodevelopmental outcomes, along with biomonitoring and physiologically-based pharmacokinetic/pharmacodynamic (PBPK/PD) modeling than was conducted in 2008. Overall, the Agency has updated and extended its evaluation of multiple lines of evidence informing the chlorpyrifos risk assessment.

As discussed the 2012 issue paper, two of the key scientific questions are: 1) the degree to which scientific data suggest that chlorpyrifos causes long-term neurodevelopmental effects from fetal or early life exposure and 2) the degree to which adverse effects can be attributed to doses lower than those which elicit 10% inhibition of AChE, *i.e.*, the dose levels previously used for regulatory decision making. The evaluation of these scientific questions requires integration numerous types of data, and consideration of the nature and degree of the uncertainties surrounding the data, including the extent to which alternative interpretations may be supported. This step is vital to robust risk characterization and uncertainty analysis. The 2011 preliminary risk assessment noted that a full weight of the evidence analysis that explicitly considers uncertainty and implications of experimental and epidemiologic lines of evidence using factors such as biological plausibility, strength, consistency, and dose-response and temporal concordance, will be conducted in the

future. A full weight of the evidence and full uncertainty characterization has not yet been conducted; the 2012 SAP is an important step toward this effort.

Question 1.0 Mode of action/adverse outcome pathway: Acetylcholinesterase (AChE) inhibition

Question 1.0

It is well established that AChE inhibition is the primary mode of action/adverse outcome pathway for OPs, like chlorpyrifos. Because AChE inhibition is the initiating event for this mode of action/adverse outcome pathway, using AChE inhibition as a regulatory endpoint is protective of downstream cholinergic effects. Moreover, historically, given the sensitivity of AChE inhibition data for OPs, these data have been considered to be protective of other potential toxicities and/or modes of action for OPs. In 2008, the Agency performed a comprehensive review of the available AChE data from multiple lifestages. This review has been supplemented with the newest studies. Consistent with the recommendations from the 2008 SAP, the Agency believes that AChE data remain the most robust dose-response data for deriving points of departure in *in vivo* experimental toxicology studies with laboratory animals. *Please comment on the Agency's preliminary conclusion that AChE data remain the most robust source of data for deriving points of departure for chlorpyrifos. Please include a discussion of the strengths and uncertainties of this preliminary conclusion.*

Response

The Panel agreed with the Agency's conclusion that the AChE data remain the most robust source of data for deriving points of departure for chlorpyrifos. This is based on the observation that all studies reporting neurobehavioral changes following *in vivo* prenatal or postnatal exposures to chlorpyrifos have been accompanied by AChE inhibition when measured at an appropriate time following administration of chlorpyrifos. Moreover many studies reporting persistent neurobehavioral changes used a potentially confounding vehicle (*e.g.*, DMSO). Most importantly, the experimental design for essentially all experimental studies evaluating neurodevelopmental effects of chlorpyrifos do not allow for the effective determination of a point of departure. There are candidates that may replace AChE as a more sensitive indicator but, at this juncture, these have not been fully validated and their alteration has not been determined to result in a well-defined, measurable neurotoxic outcome.

As in 2008, the Panel recommended that the Agency continue to use AChE data at the most sensitive lifestages for dose-response analysis and deriving points of departure. When looking at obstetric outcomes and pediatric exposures, life-stage levels of red blood cells (RBC) AChE activity in humans, which has been reported as significantly lower in fetal cord blood than in adults (de Peyster *et al.* ,1994), needs to be taken into account to eliminate potential uncertainties.

The Panel concurred with the 2008 Panel in expressed caution on the use of DMSO as a vehicle because of its intrinsic toxicity and potential influence on absorption. Again, uncertainty was expressed about potential interactions between DMSO and low doses of

chlorpyrifos and the effect of this interaction on the developing organism. In addition to the three papers cited by the 2008 SAP (FIFRA Scientific Advisory Panel, 2008b), more recent evidence is available to support the potential toxicity of DMSO. Hanslick *et al.* (2009) reported that following acute intraperitoneal injection of DMSO into 7 day-old mice, there was a significant increase in the number of apoptotic neurons at dosages as low as 0.3 ml/kg. An increased number of apoptotic neurons was also observed at 1 ml/kg which is the most frequent volume of DMSO administered in the cited studies using DMSO as a vehicle. Recent reports from the zebrafish literature suggest that DMSO has the capacity to directly induce neurobehavioral effects. Exposure to 0.05% DMSO induces anxiolytic behavior in adult zebrafish (Sackerman *et al.*, 2010) and exposure to 0.01% DMSO alters locomotor activity in larval zebrafish exposed embryonically (Chen *et al.*, 2011). Also, based on earlier studies observing that DMSO induces a stress protein response in zebrafish embryos (Hallare *et al.*, 2004; 2006), Turner *et al.*, (2012) reported that levels of DMSO as low as 25 µl/L (0.0025%) were sufficient to induce gene expression changes in embryonic zebrafish. While altered gene expression does not indicate a toxic response, it suggests disruption of homeostasis by low levels of this solvent. While the experimental studies reviewed in the White Paper all had controls with DMSO only, there is no way to rule out the potential for an interaction between DMSO and the OP. For example, Fossum *et al.* (2008) reported that 2% DMSO had no effect when microinjected into the periaquaductal gray region of rat brain, but it enhanced the potency of morphine when co-administered. In this case, if morphine was dissolved in 2% DMSO and the controls received DMSO only, the interpretation of the findings are confounded. It should be noted that the concentration of 100% DMSO is approximately 14 M. Because of the potential biological/cellular changes noted above, the lack of evaluation of potential interactions between DMSO and chlorpyrifos, and the well-known effects of DMSO on membrane permeability (Gurtovenko and Anwar, 2007), caution should be exercised in the use of data for quantitative risk assessment from *in vivo* (or *in vitro*) studies using DMSO as a solvent.

Question 2.0 Mode(s) of action/adverse outcome pathway(s): Plausible pathways leading to potential neurodevelopmental outcomes

Question 2.1

As discussed in Section 3.2.1, although there are numerous mechanistic studies in the scientific literature, the research on different hypotheses does not provide sufficient data to establish causal linkages among different levels of biological organization to show how effects lead to adversity. As such, a mode of action or adverse outcome pathway leading to effects on the developing brain cannot be established at this time. Moreover, although multiple biologically plausible hypotheses are being pursued by researchers, based on the current state of the science, no one pathway has sufficient data to be considered more credible than the others. *Please comment on the Agency's preliminary conclusion that although there are multiple biologically plausible hypotheses being evaluated by research scientists, the mechanistic experimental toxicology data do not yet support a coherent set of key events in a mode of action/adverse outcome pathway.*

Response

The Panel acknowledged the efforts EPA has taken to review all of the relevant data addressing the various cellular and mechanistic based studies on chlorpyrifos and relevant associated neurobiologically-based studies. Research scientists are examining multiple biologically plausible hypotheses regarding cellular mechanisms of chlorpyrifos neurotoxicity. Over the past approximately 15 years a number of studies have evaluated changes in neurite outgrowth, axonal transport, dendritic growth, and other cellular processes following chlorpyrifos or chlorpyrifos oxon exposure that could potentially disrupt the development of the nervous system. In no case, however, is there a defined, coherent set of events from alteration of any of these cellular functions to disrupted development of the nervous system sufficient to explain a variety of neurobehavioral changes. There is also limited evidence that these current research efforts are directed in such a manner to link the *in vitro* findings to a structural or functional change in the animal. The Panel agreed with the Agency that, based on the current state of the science, no single pathway has sufficient data to be considered more credible than the others with respect to a causal link between chlorpyrifos exposure and toxicological outcome.

As defined, the progression of events from molecular initiation to adverse outcome requires a logical sequence of changes in the mode of action/adverse outcome pathway. The Panel raised the issue that the example of domoic acid as a linear connection is likely to be a unique case that can provide components at each level of the pathway but may also generate a non-linear pattern. A linear pathway from mode of action to adverse outcome appears rare. Thus, the Panel agreed that while laudable, expectations of the existence of such a pathway may be artificially elevated and potentially unrealistic for risk assessment.

Question 2.2

Although a mode of action/adverse outcome pathway has not been established, qualitatively, the growing body of mechanistic studies does demonstrate that chlorpyrifos and/or its oxon are biologically active on a number of processes that affect the developing brain. Some mechanistic studies provide evidence of possible effects which are similarly sensitive or more sensitive than AChE inhibition (*e.g.*, neurite outgrowth, binding to muscarinic receptors, axonal transport; serotonergic nervous system development). Some of these comparisons must be considered with caution since the amount of change in the *in vitro* systems required to elicit an adverse effect *in vivo* is unknown. Moreover, extrapolation from *in vitro* perturbations to *in vivo* effects has not been established, which introduces additional uncertainties. *Given the doses/concentrations evaluated in the in vitro and in vivo mechanism studies, please comment on the degree to which these studies suggest that endpoints relevant to evaluating potential neurodevelopmental outcomes may or may not be more sensitive than AChE inhibition. Please include in your comments a discussion of the strengths and uncertainties. Please also include in your comments a discussion of the scientific understanding of dose-response relationships for biological perturbations and the magnitude, frequency and/or duration of such perturbations that can lead to adverse effects at higher levels of biological organization to support characterization of the likelihood of adverse outcomes in a human health risk assessment (as articulated in NRC 2007).*

Response
The Panel agreed with EPA that caution must be exercised in interpreting the *in-vivo* relevance of the changes observed across the various *in vitro* studies. There are a number of cellular processes such as, neurotransmitter receptor activation, and others that are currently under study; however, the studies lack the necessary data on the relevance of these changes observed *in vitro* and effects occurring *in vivo*. Much of this work is speculative because the underlying process being studied is assumed a process critical to brain development. *In vitro* work is best suited for testing hypotheses that can be further explored *in vivo*, but given their reductionist approach, they lack the experimental power and demonstrated predictive validity of an *in vivo* effect at this stage to be of scientific value for risk assessment considerations. The *in vitro* models that have been utilized to address the effects of chlorpyrifos and oxon such as neurite outgrowth, M2 acetylcholine receptor binding, mitochondrial morphology and axonal transport, as well as oxidative stress, may potentially provide information on non-cholinesterase related mechanisms.

The Panel discussed the limitations of the cellular *in vitro* systems to translate to *in vivo* systems as well as to identify a human health risk. They noted that the inherent complexity of the nervous system cannot be replicated in a cellular *in vitro* system. The majority of these studies were conducted to explore the potential for chemical exposure to induce a change in cell physiology and to further examine underlying mechanisms. Thus, while the reductionist approach provides information on possible mechanisms of action for chlorpyrifos, it does not translate to *in vivo* effects. The extrapolation of data from *in vitro* to *in vivo* is filled with uncertainty factors. Adding to this uncertainty is the possibility that although chlorpyrifos has a well-established molecular target (AChE) for cholinergic toxicity, it may not be the only toxicologically relevant target. In addition, due to the inherent complexity of the nervous system, which contains multiple regions and systems that are connected and interact with one another, effects on the molecular target could induce downstream effects that can result in a neurotoxic response that may not be directly attributable to the molecular target. This complexity and these connections cannot be replicated *in vitro*.

Issues of concern raised by the Panel were not specific to chlorpyrifos, but rather these concerns were directed toward the use of *in vitro* systems in general. Such concerns entailed the isolated nature of the cells in culture and the inability to address critical regulatory components of the *in vivo* environmental niche. For instance, the Panel noted that the *in vitro* model system can influence the effects observed in isolated cells, but the responses may be changed or absent when these same cells are co-exposed with other cells normally within their *in vivo* environment. The Panel also raised questions about how the isolated nature of the various model systems could deal with altered homeostasis or dose response differences that may occur as a function of differential recruitment of processes *in vivo*. In addition, the actual amount of compound or AChE inhibition at the target site is one critical factor toward determining if a toxicological event observed *in vitro* can occur *in vivo*. The Panel mentioned that that until such *in vivo* translation can be established, it is difficult to determine what level, magnitude, or duration of change is required within each model system to be indicative of a change that may occur *in vivo*.

The Panel also discussed the likelihood that using such models may not provide a linear dose-response relationship for chlorpyrifos or for AChE inhibition. Thus, providing a direct assessment of the relative sensitivity to AChE inhibition is difficult. Changes in neurite outgrowth, dendritic spine development, axonal transport and other cellular responses that have been seen with chlorpyrifos or chlorpyrifos oxon exposure *in vitro* have been reported to occur at levels of exposure below those necessary to inhibit AChE. These comparisons must be made with caution. For example, adding chlorpyrifos oxon directly to hippocampal neurons *in vitro* in tissue culture medium is unrealistic with regards to how the oxon would reach a neuron *in vivo*. Many detoxifying/binding proteins are not included in *in vitro* conditions, potentially modifying the interaction of the chemical with the cell. Similarly, if acetylcholinesterase in a disrupted tissue (*e.g.*, a homogenate) such as liver and is inhibited *in vitro* by chlorpyrifos oxon, its sensitivity is much higher than if that same enzyme is immunoprecipitated and then inhibited by chlorpyrifos oxon *in vitro* under similar conditions. Such effects of tissue components, in the relative potency of chlorpyrifos or chlorpyrifos oxon, make extrapolation of *in vitro* effects to *in vivo* settings difficult. In general, changes in neurodevelopmental endpoints require relatively higher exposures in *in vivo* models. Reviews of mechanistic/cellular studies with neurodevelopmental outcomes following chlorpyrifos exposure suggest that such responses may occur at dose levels at, near, or above those necessary to induce AChE inhibition. The development of a physiologically-based pharmacokinetic (PBPK) model that can estimate the *in vivo* dosage required to reach toxicant concentration at the target site that is similar to the *in vitro* concentration at which the effects were observed would be especially valuable. Such a model would assist in determining the plausibility that the effect observed *in vitro* also occurs during an *in vivo* exposure.

The Panel was in agreement that further mining of the published literature may provide significant information on how one might utilize the available data obtained for chlorpyrifos to generate plausible hypotheses for future evaluation. As an example, the analytical studies of the Lockridge group indicating that chlorpyrifos (chlorpyrifos) oxon can covalently modify key cytoskeletal proteins such as tubulin and motor proteins like kinesin provide information that can contribute to the interpretation of findings of alterations in neurite outgrowth and axonal transport, respectively. Such an integrated effort may allow for the design of specific targeted studies to test the hypothesis *in vivo* as an effort to obtain predictive validity (Jiang et al., 2010; Grigoryan et al., 2009).

The Panel considered items that might be added to the review. One topic is the effect of chlorpyrifos on neurotrophins (growth factors). Pope *et al.* (1995) provided the first evidence that OPs might alter the activity of growth factor-like (neurotrophic) molecules. Data suggesting that growth factors could be altered within the brain tissue, *in vivo,* has recently been provided in neonatal rats across low dose levels of chlorpyrifos. Alterations were observed in mRNA levels for specific members of the fibroblast growth factor (FGF) superfamily of neurotrophic factors (Slotkin *et al*., 2007). Further, early postnatal exposure to chlorpyrifos has also been associated with decreases in nerve growth factor (NGF) in the rat forebrain (Betancourt and Carr, 2004). These effects are not limited to the immature rodent in that adult exposure to chlorpyrifos can result in protracted alterations in NGF-related signaling proteins (*e.g.*, the high affinity nerve

growth factor receptor TrkA and its activated form, phospho-TrkA in the prefrontal cortex) (Terry *et al.*, 2007). Also, as a note, in the Middlemore-Risher *et al*. (2011) study, alterations in mitochondrial morphology and decreases in axonal transport were observed in primary cortical neurons exposed to chlorpyrifos and chlorpyrifos oxon at concentrations including ones that did not inhibit acetylcholinesterase.

The Panel raised the issue that oxon and protein adducts likely serve as a potentially important pathway for cellular/protein damage. The oxon has an incredibly rapid half life and despite a relatively high affinity for ChE, one would expect that they would occasionally bind non-ChE cellular components 3,5,6-trichloro-2-pyridinol (TCPy) and diethylthiophosphate (DETP) are conjugated by -O-sulfotransferases or gluruconosyltransferases; and as presented in public comments from Dow Chemicals, both sulfonylates and glucuronides are "equally prevalent" but relative affinities (Km) appear to be unknown. In translating from animal to human, it was considered of importance by the Panel that Uridine 5'-diphospho-glucuronosyltransferase enzymes (UGTs) develops late in children. In this case the sulfotransferase (SULTS) are usually present during gestation, despite the fact that the AChE adducting oxon would not be conjugated. One must consider that if glucuronidation is the rate-limiting pathway in children, then other metabolites may accumulate to toxic levels due to ontogenetic inadequacy of UGTs. This could result in a potential for error in biomarker analysis and generate errors in dosimetry estimations.

The Panel discussed the types of dose-response relationships that may be observed and allowed for non-linear patterns or no clear dose-response association to be observed. The Panel cautioned that when examining the dose-response relationship, one should not uncouple PD analyses from PK models too far given that PK differences can affect active site concentrations and hence, PD effects. To this end PK can significantly affect the magnitude and duration of an effect. It was noted that the p450s and PON1 were well integrated into the PB/PK/PD models and this was considered a major strength. There were, however a number of weaknesses discussed including the fact that there is a substantial lack of knowledge about the high capacity of phase II conjugation for chlorpyrifos in humans. For example, there are species differences in Phase II where UGTs and SULTS may be non-orthologous between humans and rodents; hence any extrapolation of animal data to humans must take this into consideration. The Panel noted that this was highlighted by the public presentation from Dr. Hattis who noted that the human data he subsequently used and modeled demonstrated lower clearance than seen in rat data. An issue was raised by the Panel that pregnancy can be considered as a specific state and that information is needed relative to how PK differs in a pregnancy scenario relative to exposure and toxicity. In addition to maternal influences such as metabolism by the liver, the placenta was also raised as a unique component requiring attention in such PB/PK/PD models for gestational exposure.

The Panel raised a concern that equivalency developmental stages between ages of rodents to human are not well defined with regards to cell type compositions, brain region, cellular architecture, and physiological or biochemical process. This is not a problem that the Agency needs to address but rather the Panel emphasized that specific

developmental periods in which perturbation may occur appear to be ill defined and may not translate between rodent and human species. The numerous *in vitro* mechanistic studies suggest that chlorpyrifos can alter numerous biological processes in normal brain development. However, these data do not permit translation to the *in vivo* situation nor do they provide a quantitative dose-response relationship that can be compared to AChE inhibition.

Question 3.0 Neurodevelopmental data from laboratory animals

Question 3.1

As discussed in Section 3.2.2, the experimental toxicology data in laboratory rodents show neurobehavioral effects following developmental exposure with changes in a number of neurological domains. In 2008, the SAP agreed to this preliminary conclusion, and the nine additional studies available since 2008 add further support. *Please comment on the degree to which these studies show changes in a number of neurological domains and support the qualitative conclusion that chlorpyrifos exposure during gestation and/or early post-natal period may result in long-term adverse effects on the developing nervous system. What evidence does and does not support this conclusion? Please also include in your comments a discussion of the strengths and uncertainties. Please also include in your comments a discussion of the scientific understanding of dose-response relationships for biological perturbations and the magnitude, frequency and/or duration of such perturbations that are can lead to adverse effects at higher levels of biological organization to support characterization of the likelihood of adverse outcomes in a human health risk assessment (as articulated in NRC 2007).*

Response

In order to address the first part of this charge question, the Panel critically reviewed these toxicology data in laboratory rodents. This review included a total of 21 developmental neurobehavioral effects studies which also entailed the nine studies published since the 2008 SAP review (These studies are identified in Appendix 3 of the Agency's Draft Issue Paper: Scientific Issues Concerning Health Effects of Chlorpyrifos.). Based upon their review, the Panel agreed with the 2008 SAP conclusions that developmental neurobehavioral experiments show adverse effects of chlorpyrifos exposure. However, the Panel cautioned that the existing neurobehavioral studies are limited and a number are under-powered and prone to Type I error (meaning the null hypotheses may have been falsely rejected) and therefore should be discounted in determining the weight of evidence for or against neurobehavioral effects from developmental exposure to chlorpyrifos. An additional concern was raised by the Panel in the inclusion of tests that have not been validated as to neurotoxicological significance. Such assessments included anxiety tests, depression tests, or social interactions. The Panel concluded that, in the current state, such outcomes should be regarded as exploratory, and hypothesis-generating, rather than evidence of toxicity. The Panel considered that the lack of observable effects below dosages equal to or exceeding 1.0 mg/kg/day in some studies could be due to the possibility that the effects at lower dosages might not be the same as those observed at higher dosages. In addition, low dose exposure may lack a sufficient level of response to elicit a physiological compensation

response that would occur with higher dose levels. In this scenario, the toxic effects of the higher and lower dosages may manifest themselves in different ways. If the response is through the same physiological target, the expected dose-response curve may be altered (such as a U-shaped or inverted U-shaped curve). In contrast, the response of the lower dosages may be quite different from that of the higher dosages and require a different behavioral paradigm.

Many of the 21 studies reviewed included 2 or 3 dose levels of chlorpyrifos (ranging from 0.2 in one case up to as high as 10 mg/kg in another but more commonly from 1-7 mg/kg), but dose-effect outcomes were, more often than not, not observed. Some of the most consistent effects are from the Slotkin-Levin experiments (see below) where radial arm maze deficits and several other effects have been replicated but the studies are seldom designed with three doses levels and even when two dose levels were included the findings were not dose-dependent in most cases. The Panel agreed with the Agency that the lack of specificity of direction of the neurobehavioral findings is problematic. A statistically significant change in isolated markers of certain behaviors may not be supported by other studies under similar dosing paradigms thereby raising concern regarding the biological significance of the observed change. The Panel questioned the Agency's interpretation that a change in either direction or a specific behavior is necessarily indicative of an adverse effect. Rather, such discrepancies may suggest methodological error, problems in study execution, or a predilection toward searching data for positive rather than negative findings. Dose-response and attention to methodological issues should play a role in evaluating the weight of evidence within and across the different studies. The Panel agreed that the overall evidence across these studies is persuasive, indicating that there are enduring effects from chlorpyrifos exposure at 1.0 mg/kg or above on the CNS. Future neurodevelopmental studies need to focus on levels below 1.0 mg/kg/day and to expand the studies to identify the correct testing paradigm to detect these effects and possibly identify a neural substrate. The Panel also considered the possibility that additional negative data exists at lower dose levels but is not available in the published literature. The Panel suggested that cross-laboratory or collaborative studies could provide systematic comparison of the effects of chlorpyrifos on neurodevelopmental domains using unified exposure periods, dosing, age of testing, and methods, combined with urinary analysis of chlorpyrifos' metabolites, and accurate assessments of AChE inhibition.

In evaluating the inconsistencies among these studies, the Panel suggested that the Agency should consider factors such an the distinct ontogeny of the various brain regions, cellular components, and neurotransmitter systems in the fetal/gestational exposures and that the structural and functional maturation of each system is unique and thus may be at different stages during the age of exposure. In addition, the redundancy and compensatory capability of each system should be considered as the level of insult may be required to reach a substantial level before it manifests as a neurobehavioral change.

The Panel's detailed review of each these developmental neurobehavioral effects studies is provide in Appendix A of this report. However, the following provides a collective summary of the Panel's observations and conclusions regarding the studies.

Among the 21 reviewed articles (which include more than 21 experiments), many effects are reported at chlorpyrifos doses ranging from at 1.0 to 7.0 mg/kg (and in one study 10 mg/kg). However, these are dose levels known to significantly inhibit cholinesterase in RBC, therefore, based on these 21 studies, cholinesterase inhibition is an adequate threshold as no credible evidence of neurobehavioral effects below 1.0 mg/kg were found.

Three studies tested doses <1.0 mg/kg chlorpyrifos. Two studies used 0.3 mg/kg and one used 0.2 mg/kg (Jett *et al*., 2001;Braquenier *et al*., 2010;Maurissen *et al*., 2000). Of these, two found no effects at these lower doses (Maurissen *et al.*, 2000;Braquenier *et al.*, 2010). Only one study found effects at 0.3 mg/kg (Jett *et al*., 2001), however, this study contains serious methodological flaws which are of sufficient magnitude to cast serious doubt on the credibility of the findings. While the data from Jett *et al*. (2001) raise the possibility of neurobehavioral effects at 0.3 mg/kg/d, these data require replication in a study that is properly designed, adequately powered, and appropriately analyzed. Until such time as new data at such lower doses become available, it is concluded that no dose <1.0 mg/kg in any neurodevelopmental behavioral studies shows evidence of adverse effects (or of any effects, even including those outcome measures of indeterminate/unknown toxicological significance).

In addition, effects of chlorpyrifos at 1.0 mg/kg are difficult to interpret because of methodological limitations, inconsistencies, and variation in study design, sometimes lack of control for litter effects, oversampling issues, behavioral methods used, and lack of dose-response findings.

At doses exceeding 1.0 mg/kg, the data show somewhat more consistency, but even here, dose-response experiments are the exception. A 5.0 mg/kg of chlorpyrifos, reduced body weight is sometimes seen, and at doses above 5.0 mg/kg increased mortality may occur along with other evidence of toxicity. Given this, it is a significant gap in the literature that more dose-response studies are not available in the range downward toward 0.2 mg/kg and extending up to and including doses previously tested of 1.0-2.0 mg/kg in order to determine what, if any, dose-effect curve occurs in this range for neurobehavioral effects.

It appears that prenatal and prenatal-neonatal exposures are more sensitive than neonatal exposure alone on neurobehavioral outcomes. This implies that prenatal exposure may be the exposure period contributing to this observation, but unfortunately, most of the pre- and neonatal studies are not entirely informative because the neonatal exposure was to the dam rather than directly to the progeny. This makes it unclear what the exposure to the offspring actually was or whether it was at similar levels to those reaching the embryo and fetus. More studies, especially dose-response studies, in the lower dose ranges with exposure from implantation to the end of major neurogenesis (approximately P20) are

needed, again with doses below 1.0 mg/kg and with concomitant measurement of maternal, fetal, and neonatal cholinesterase activity.

Many of the existing studies expose for only a narrow interval during gestation or the neonatal period. Prenatal exposures should be from E6-20 to 21 for rats, and E6-18 or 19 in mice in order to span most of early brain development (equivalent to human first and part of second trimester). And for neonatal treatment, exposures should be from shortly after birth to approximately P20 (equivalent to the latter half of second and all of third trimester equivalent brain development comparable to that for humans). If the critical period or most sensitive period is within this range, then such comprehensive exposure should cover the entire span of CNS development that represents the species being modeled, *i.e.*, human beings.

In the prenatal studies, the use of timed-pregnant females shipped from breeders is problematic for behavioral studies because maternal stress, even if regarded as equivalent across dams assigned to the treated and control groups, introduces a variable that has the potential to interact with the independent variable. If maternal stress were to interact with chlorpyrifos, it would confound the outcome and make a result difficult to interpret (which is exactly what is found in many of the reviewed studies). Since no one has tested for this, it is currently impossible to rule it out.

Many studies use diurnal and some nocturnal testing. If additional dose-response studies are undertaken, this factor should be held constant so that results can be better compared.

Question 3.2

The dose-response data in the *in vivo* experimental neurodevelopmental toxicity studies are not amenable to empirical dose-response modeling as many studies use only one or two doses, and in some cases the lower dose, but not higher dose level, produced significant effects. Many studies report effects at a dose of 1 mg/kg/d-- a dose that produces some amount of brain ChE inhibition when given directly to the pups post-natally, but may or may not alter fetal brain ChE activity when given to the dams gestationally. One study (Braquenier *et al.*, 2010) using lower doses, administered to the dam on GD15-LD14, reported a NOEL of 0.2 mg/kg/d. Comparing the NOEL of 0.2 mg/kg/d to a repeated dosing AChE inhibition $BMDL_{10}$ of 0.03 mg/kg/d suggests that AChE inhibition is a sensitive and protective endpoint.

a. *Please comment on the scientific quality and robustness of the animal neurodevelopmental toxicity studies.*

Response

The quality of the studies in this category varies, but there are some of high quality. Overall, the studies by Slotkin (Dam *et al*., 2000; Icenogle *et al*., 2004) and Levin (Levin *et al*. 2001 and 2002), those by Carr *et al.* (Carr *et al.* 2001 and Johnson *et al.*, 2009), the study by Maurissen (Maurissen et all, 2000), and several of those from the Ricerri and Venerosi group (Ricerri *et al.*, 2003 and 2006 and Venerosi *et al.*, 2006, 2008, and 2010), are among the better ones because they generally used multiple doses, had adequate sample sizes, controlled for litter effects, used sound behavioral methods, and used

appropriate statistical methods to analyze the data. Because each of these studies found long-term neurobehavioral effects, these data may be regarded as robust within the limits of what was tested. These more persuasive findings only occurred at doses of 1.0 mg/kg of chlorpyrifos and above. This data set is, therefore, moot concerning effects at <1.0 mg/kg of chlorpyrifos. Among these studies, however, several concerns remain. In the Slotkin and Levin experiments, the use of commercially supplied timed-pregnant rats is a concern as is the use of Zivic-Miller (ZM) Sprague-Dawley rats. The ZM rat is known to be different from other Sprague-Dawley rats on some behavioral tests where it often performs as an outlier. For studies with regulatory implications, it is preferable to use a mainstream rat strain such as Sprague-Dawley from Charles River or Harlan where much more is known about their behavioral characteristics and they do not perform at the extremes of the distribution. Also, the concern about the RAM method as noted in Panel review in Appendix A should be taken into account with regard to robustness. Notwithstanding these caveats, the weight of evidence from the neurobehavioral studies is that there are too many long-term effects for them all to be attributable to Type I errors, hence, it is more likely than not that there are significant long-term adverse effects and in this the Panel concurs with the conclusions of the 2008 SAP findings and the EPA White Paper background document.

c. *Please comment on the degree to which studies that measured AChE inhibition and those that measured neurodevelopmental outcomes can be integrated to evaluate whether points of departure based on 10% AChE inhibition provide more sensitive endpoints than endpoints measured in the experimental neurodevelopmental studies (as reviewed in Section 3.2.2). Please include in your comments a consideration of the strengths and uncertainties associated with this assessment.*

Response

Data in the available studies, including the nine additional studies reported since 2008, provide qualitative (emphasis on qualitative) support for the effect of chlorpyrifos exposure during gestation and/or early post-natal period and long-term adverse effect on the developing nervous system. Several of these studies examined AChE activity in the brain after oral and/or subcutaneous chlorpyrifos exposures during postnatal periods, and inhibition of AChE within one day of exposure was observed in these studies at doses as low as 1 mg/kg/day. Since AChE inhibition recovers quickly the data are insufficiently refined to allow for a linkage between the mode of action and the neurodevelopmental effects (acute vs. chronic, respectively).

Since the mode of action of these effects is not established and cannot be presumed *a priori* to be related to AChE inhibition, these studies do not exclude the possibility that other mechanisms may be involved, especially long-term effects where functional characteristics may be unmasked at later life-stages due to neuroplasticity. A few studies have reported AChE inhibition when a dose of 1 mg/kg/d was administered directly to the pup postnatally (Dam, *et al*., 2000; Johnson, *et al*., 2009; Ricceri, *et al*., 2003). However, none of the neurobehavioral studies described in the Panel's review tested for fetal AChE inhibition when 1 mg/kg/d was given during gestation. A companion study to Maurissen

et al. (2000) reported no cholinesterase inhibition in samples taken from fetuses 4 h after dosing the dam when 1 mg/kg/d had been administered daily since E6 (Mattsson, *et al.*, 2000). Qiao *et al*. (2002) also reported no brain AChE inhibition in fetuses 24 h after the last dose of 1 mg/kg/d to the dam on E17-20. No other time points or days were assessed in either study. These results suggest, but do not confirm, that the fetus would not experience AChE inhibition at 1 mg/kg/d to the dam, further suggesting that the behavioral effects reported in those studies were not due to AChE inhibition.

The studies published since 2008 demonstrate alterations in a number of neurodevelopmental and biochemical outcomes. The amount of AChE inhibition required to elicit the various endpoints was however inconsistent and varied, because of differences in study designs, analysis of different endpoints, and how long animals were followed-up. Many of the studies measured AChE inhibition 24 hours or longer after dosing, which can underestimate the amount of AChE inhibition. Furthermore, since the neurodevelopmental effects may be independent of AChE inhibition, it needs to be considered whether AChE inhibition represents a critical marker for derivation of points of departure when considering chronic studies.

Finally, the Panel notes that there has been little consideration of the relationship to genetic variability on experimental outcomes, the exception being paraoxonase 1 (PON1). Recovery of AChE activity is linked to changes in AChE gene expression. It has been previously reported that molecular and behavioral effects may be attributable to alternative splicing of the AChE gene. Within the brain there are 2 variants: AChE-S (synaptic) and AChE-R (read-through splice variant) mRNA. Under normal conditions variant AChE-S dominates; however, under stress conditions, such as OP exposure (chlorpyrifos has yet to be studied), the transcription of AChE-R increases. Following stressful events, AChE-R increases to a level that is no longer adaptive and result in varied physiological changes. Of the neurobehavioral effects reported in the reviewed experiments that assessed AChE inhibition, no studies were identified that showed effects on behavior at low levels of AChE inhibition, including at 1.0 mg/kg of chlorpyrifos. Doses below 1.0 mg/kg/day chlorpyrifos did not show convincing evidence of neurobehavioral effect; hence, no extrapolation to lower doses in terms of AChE inhibition is possible from the data reviewed herein.

Question 4.0 Epidemiology Regarding Children's Health

Question 4.1

Section 4.0 and Appendices 5 and 6 provide the Agency's review of the available epidemiology studies from the Columbia Mothers and Newborn study, the Mt. Sinai Child Development study, and the Center for Health Assessment of Mothers and Children of Salinas Valley (CHAMACOS) study. Consistent with the 2008 SAP recommendations, the Agency has considered information offered from each of the three cohort investigations; however EPA acknowledges the primacy of the Columbia cohort data for the purposes of informing risk assessment because researchers measured chlorpyrifos parent compound directly in this study. *Please comment on the sufficiency, clarity, and quality of the Agency's epidemiology review as contained in Section 4.0 and*

Appendices 5 and 6 of the draft issue paper with respect to identifying the major strengths and limitations of each study.

Response

The Panel believes that the epidemiology section of the draft issue paper is very well-written, clear, accurate and fairly complete. The Panel commends the Agency staff on the thorough review of the epidemiology literature, for putting their epidemiology review in the context of the modified Bradford Hill criteria, as recommended by the 2010 SAP, and for reviewing the potential for selection and information biases in each of the studies. In particular, the Panel commends the Agency staff for the tremendous amount of work and thoughtfulness that went into Appendices 5 and 6. The Panel believes that the epidemiology review appropriately concludes that the studies show some consistent associations relating exposure measures to abnormal reflexes in the newborn (using the Brazelton Neonatal Behavioral Assessment Scale), pervasive development disorder at 24 or 36 months, mental development at 7-9 years, and attention and behavior problems at 3 and 5 years of age , in addition to less consistent results for reduced mental and psychomotor development (measured by Bayley scores) at 12 and 24 months. Inconsistent results were found for associations between exposure and measures of fetal growth.

The Agency's epidemiology review provided an excellent description of the strengths and limitations of the studies conducted to examine the relation of chlorpyrifos to children's growth and neurodevelopment. Epidemiologic studies such as these require that large numbers of mothers and infants/children be followed longitudinally for an extended period with extensive data collection at regular intervals to ascertain exposure measures, potential confounders, and health outcomes, all of which can change over time. These studies are logistically difficult to implement and require great commitment by the researchers and a potentially large burden in terms of time and effort on the part of the study participants over a lengthy period of time, often with little or no specific benefit or return to themselves.

The Agency's review adequately summarizes the challenges and scientific contributions of each of three studies: one conducted in an inner city sample of African American or Dominican initially pregnant women and their infants/children by Columbia University investigators; one conducted by Mt. Sinai investigators in a predominately Hispanic and African American sample in New York City, and one conducted in the Salinas Valley in California led by University of California Berkeley investigators. As noted in the review, all three studies had the significant strength of being longitudinal, prospective designs, the most effective design for establishing the temporal sequence in relating exposure to health outcomes, specifically in these studies relating exposure measures obtained prenatally and/or at delivery to outcomes measured at six months up to nine years of age.

Regarding exposure assessment, only the study by the Columbia investigators measured chlorpyrifos parent compound in cord blood, and conducted an exposure validation study and looked at correlations of cord blood measures with mothers blood and meconium, and used the cord blood as the measure of exposure in relation to scores on standard

neurodevelopmental test batteries for children at age 7 years (Rauh *et al.*, 2011). The Panel agrees with the Agency that because this study included the most specific exposure measure, particular attention should be focused on its results. The Panel disagrees with some of the public comments claiming that because the observed associations with adverse neurodevelopmental outcomes occurred at levels below those required for AChE inhibition, the results should be discounted. Instead, the Panel believes that these findings are derived from a well-designed and conducted study and thus suggest that the mechanism may be other than that of AChE inhibition. Further, the very large effect observed in this study for the relationship of exposure to attention deficits, reduced birth weight prior to the voluntary withdrawal of chlorpyrifos, reduced mental and psychomotor development, and reduced Intelligence Quotient (IQ) are unlikely to be due to important misclassification of or bias in assessing exposure or to uncontrolled confounding. [The Panel notes that in Appendix 5, some of the directional signs of effect, *i.e.*, betas, and their 95% confidence intervals (CIs), are missing or incorrect so that all of them should be double-checked and corrected.] Finally, the review might additionally note that the timing of exposure may be important, although the critical time window of exposure is not known with certainty.

Although, the other two studies only used metabolites as markers of chlorpyrifos exposure, all three studies used TCPy, a metabolite specific to chlorpyrifos, in relation to at least some of the outcomes assessed. (Even though TCPy is a better measure than dialkylphosphates (DAPs), the other indirect biomarker, the TCPy measure has limitations as noted by the review. The potential misclassification of exposure should be more explicitly stated. The value of using TCPy as a biomarker also hinges on the mode of action, which is not established with certainty.) As the epidemiology review appropriately notes, all three studies were strengthened in design by using biomarkers of exposure instead of relying on self-reports of exposure, which would be likely to result in much greater misclassification of exposure. All three studies also used similar standard, validated measures of non-verbal and general intelligence, behavior and home environment, which enhance the quality of the outcome assessments and the ability to assess the consistency of the findings regarding these outcomes across studies. In addition, as appropriately noted in the Agency's review, all three studies collected extensive data on potential confounding variables, used appropriate multivariate statistical techniques to control for confounding effects of socioeconomic factors, lifestyle and behavioral factors as well as additional environmental exposures, and conducted sensitivity analyses to determine if assumptions made about missing data, for example, were appropriate.

While agreeing that chlorpyrifos could have played a role in the neurodevelopmental outcomes observed in the Columbia cohort, some panel members raised concern about associating the observed deficits in neurodevelopmental outcomes in children with a single chemical, given that this was a multi-chemical exposure spanning a multi-year period that encompassed an important period of sequential developmental processes necessary for brain maturation. Rauh *et al*. (2011) reported that decreased working memory and full-scale IQ in 7 year-olds were statistically significantly associated with prenatal chlorpyrifos exposure. In an earlier examination of the same cohort, Perera *et al.*

(2009) reported an association between a decrease in full-scale IQ and verbal IQ in 5 year-olds with prenatal polycyclic aromatic hydrocarbons (PAH) exposure rather than chlorpyrifos, thus, raising an issue of the shift in chemical exposure association with increase in age. In each of these analyses, statistical modeling showed that the exposures were independently associated with IQ, and no significant interaction was observed with the other chemical. While this is a statistically sound approach to determine independent responses, panel members noted that it is very difficult to identify the independent physiological effects of a single chemical in this type of multi-chemical exposure scenario. This identification is further complicated by limitations in exposure assessment with respect to on-going and post-natal exposures and the potential for chemical interactions during the exposure period. In addition, developmental progression of the children and the level of skills examined by the tests employed may have been confounding factors. Maturation of the brain is a critically timed sequence of events with each subsequent event dependent upon the successful completion of the previous one. Thus, appropriate brain function at age 7 is dependent on completion of maturation processes that occur at earlier ages. Panel members noted that, while this statistical approach could be used in studies examining the exact same endpoint at a single age, this brain maturation process would need to be taken into consideration prior to determining that at 5 years of age the cognitive deficit was due to one exposure and at 7 years of age it was due to a different chemical. The ever-changing aspect of any developmental study is further demonstrated in the assessments of this cohort of children at earlier ages. At 36 months of age, the deficits in the Bayley Mental Development Index scores were associated with exposures to prenatal chlorpyrifos (Lovasi *et al*., 2011), prenatal phthalates (Whyatt *et al*., 2012), prenatal PAHs (Perera *et al*., 2006), and prenatal piperonyl butoxide (Horton *et al*., 2011). Thus, panel members cautioned about identifying any one specific chemical as the main one associated with the cognitive deficits observed at 7 years of age in the Columbia cohort.

One additional concern is that in general, the sample sizes of the three studies were only moderately large, ranging from just over 100 to slightly under 500, depending on which subset of data from mothers and children were analyzed. The more recent papers had fewer participants, ranging from just under 200 to just over 300. The epidemiology review correctly notes that the modest samples sizes were a limitation in having sufficient statistical power to detect as statistically significant possible modest relations of exposure to outcomes or interactions with other variables. Thus, modest sample sizes were one of the most important limitations of these studies, which is reflected in the wide confidence intervals for some of the effect estimates and the use of moderate (*e.g.* 1 standard deviation) or large (*e.g.*, 10-fold) increases in exposure measures (which did not seem to be mentioned in the Agency's epidemiology review) to see statistically significant effects, *e.g.*, in IQ (Rauh *et al.* 2011; Bouchard *et al.* 2011). However, some evidence of interaction with paraoxonase 1 (PON 1) genotype and/or phenotype was provided in some of the studies (Berkowitz *et al*. 2004, Engel et al 2007; Engel *et al*. 2011; Harley *et al*. 2011), and some examinations of interactions with other exposures were presented in the studies and summarized in the review. Future examination of potential epigenetic effects might also be informative. The Panel also recommended that investigators of the

three studies consider possible pooling of samples and data to enhance the ability to investigate effect modification and possible roles for other agents.

Two other items that might be added to the review are: 1) replacing "null" and "positive (ns)" with point estimates and 95% CIs for effect estimates (to the extent possible, realizing that quantiles or betas might have to be used) to Table 10 on page 59; and 2) noting in the text that other interactions (*e.g.*, with sex of the child, gestational age at measurement of exposure, length of breastfeeding, use of alcohol, *etc*.) were not consistently described in the three studies, and in most cases sample sizes were inadequate to have sufficient statistical power to detect meaningful effect modification as statistically significant. Providing the point estimates and 95% CIs for effects in Table 10 will permit assessment of the magnitude, variability, and direction of the effects, which are more important in assessing consistency than statistical significance. The second point concerning interaction is important because it means that the potential for stronger associations (larger effects) in subgroups with potentially enhanced susceptibility could not be or were not adequately examined or reported. In addition, the Agency's epidemiology review mentions that the restriction of some of the study samples by race/ethnicity and/or to low risk pregnancies (*e.g.*, nonsmokers, women without comorbidities) reduced the potential for confounding (which was a plus) but also reduced the generalizability of the results. However, the review perhaps did not sufficiently emphasize that this limitation also meant that modification of effect by race/ethnicity or other risk factors could not be examined with these study sample restrictions, and the sample sizes in general were inadequate to examine interactions with such factors. Thus, differential effects for subgroups with other risk factors or characteristics could not be determined.

The Agency's epidemiology review also examined the potential for misclassification and bias in each of the studies and mentions the likelihood that any such misclassification and/or biases that operated were non-differential and thus likely to result in an under-estimation of effect. For example, chlorpyrifos exposures, particularly when the parent compound was not measured, could have been misclassified, especially because some analyses indicated greater within-person than between-person variability in exposure measures. However, this was unlikely to be differential with respect to the neurodevelopmental outcomes measured and thus would likely have resulted in bias to the null or under-estimation of effect measures. Similarly, although not explicitly stated in the publications, it was unlikely that those who were assessing outcomes using standardized measurement instruments knew the exposure levels of the participants, which could have biased their assessments. Thus, again, misclassification of outcomes could have occurred but were likely non-differential with respect to exposure levels and thus were likely to have resulted in bias to the null or under-estimation of effects.

The Panel also felt they should respond to the issue of multiple comparisons that was raised in the public comments. The Panel feels it is important to note that all the comparisons made in the three studies were hypothesis-driven and dealt with related outcomes, rather than reflecting "fishing expeditions" that would have been likely to result in significant findings by chance due to multiple comparisons. The Panel thus

believes that the multiple comparisons issue is not an important concern regarding the findings of the three studies over the years.

The Agency's epidemiology review reflects the authors' views from the three studies that among the statistically significant effects seen, most appeared to have a linear relation with exposure with no evidence of a threshold. However, upon examination of some of the graphs and other results presented in some of the papers, it would appear that this point requires some further data and examination. For example, the graphs in the Columbia study seem to suggest no threshold for the effect on working memory but do suggest a threshold for the full-scale IQ (Rauh *et al*. 2011). In the California sample, the graphs presented in the most recent paper (Bouchard *et al*. 2011) suggest a drop in IQ beginning generally with the second quintile of exposure level (depending on which outcome is examined) and seem not to worsen greatly in higher quintiles of exposure levels, which is also suggestive of a threshold effect. The graphs presented in the recent publication from the Mt. Sinai study (Engel *et al*. 2011) seem to indicate no threshold when using tertiles, but the confidence intervals were quite wide. Thus, the Agency's epidemiology review appropriately notes that, due to the modest sample sizes, statistical power may have been inadequate to detect departures from linearity with log transformed exposures or outcomes. It might also be mentioned that modest sample sizes limited statistical power to assess dose-response adequately, which is one of the key postulates promulgated by Bradford Hill, so that such attempts sometimes resulted in wide confidence intervals around effect measures in each quantile, making adequate assessment of dose-response difficult, and that different quantiles were used across the studies, making direct comparisons difficult. Additional analyses of dose-response in both animal and human data and particularly at lower levels of exposure would be very helpful in informing inferences from the epidemiologic studies.

In conjunction with the modified Bradford Hill criteria, the epidemiology review states that a biologically plausible role for chlorpyrifos in relation to adverse neurodevelopmental outcomes is believed to involve inhibition of AChE. While this is a reasonable assumption, the recent papers from the epidemiologic studies noted that noncholinergic mechanisms may play roles in the associations of exposure with the neurodevelopmental outcome measures (Bouchard *et al.* 2011; Rauh *et al*. 2011). In addition, the observed effect modification also suggests other mechanisms, including oxidative stress and lipid peroxidation. The mode of action is discussed in more detail elsewhere in the draft issue paper, but these additional potential mechanisms might be added in the epidemiology review. As noted above, just because the significant effects are observed at exposure levels below which (acetyl cholinesterase) AChE inhibition occurs does not mean that the observed associations are not real, but rather that the mechanism(s) in humans may be other than by AChE inhibition. Further mechanistic work needs to be done to clarify this issue.

In summary, the epidemiology review contained in the draft issue paper is very clearly written, accurate and generally provides a very thorough review in the context of the modified Bradford Hill criteria. As noted above, a few additions would enhance the completeness of the review.

Question 4.2

Similar to the initial conclusions from 2008, the Agency has preliminarily concluded that, qualitatively, chlorpyrifos likely played a role in the neurodevelopmental outcomes reported in the epidemiologic studies, and that information available since 2008, including both new etiologic investigations as well as epidemiologic methods papers, strengthens this conclusion. *Please comment on the Agency's preliminary, qualitative conclusion that chlorpyrifos likely played a role in the neurodevelopmental outcomes observed in the epidemiologic studies. Please include in your comments a discussion of the strengths and uncertainties associated with this preliminary conclusion.*

Response

Overall, the Panel reiterates the 2008 SAP's conclusion and the Agency's concurrence with the statement that chlorpyrifos likely plays a role in neurodevelopmental outcomes in the three cohort studies. The qualitative conclusion of the epidemiology review seems well-justified. The Panel agrees with the Agency that although exposures to other AChE-inhibiting compounds cannot be ruled out as contributing to neurodevelopmental outcomes, the potential combination and/or additive effects of these compounds do not rule out the role of chlorpyrifos. However, it should be noted that it cannot be stated that chlorpyrifos is the sole contributor to the observed outcomes.

The conclusion is enhanced by the strengths of the three studies reviewed, specifically:

- the longitudinal designs which permitted clear indications of the temporal relation of chlorpyrifos exposure to adverse neurodevelopmental outcomes;
- the inclusion of biomarkers of exposure as well as self reported exposure;
- the relative consistency of findings in different populations but using similar standardized exposure and outcome measures;
- the strength of the associations found;
- the use of objective measures of exposure and standardized, validated measures of outcomes;
- the control of multiple confounding variables including other environmental exposures and other pesticides;
- the suggestion of a dose-response effect;
- minimization in bias in assessing outcomes and exposures and the likelihood that biases and misclassification of exposures and outcomes resulted in a bias to the null, *i.e.*, under-estimation of effect; and
- attempts to investigate genetic and phenotypic effect modification and dose-response effects.

The conclusion is further supported by the following details of strength and consistency of association and a crude exposure response relationship. Some of these details were previously presented in the 2008 SAP report, but they are reiterated here to include all epidemiologic evidence in one place. It should be noted that studies published since 2008 have continued to show associations between neurodevelopmental outcomes and potential exposure to chlorpyrifos and have strengthened the available epidemiologic

evidence. Recent analyses have looked at neurodevelopmental outcomes in older children and addressed some of the issues of confounding by socioeconomic status, other pesticides, and issues of exposure measurement validation.

Strength of association: This criterion focuses on the Columbia cohort because this cohort specifically measured chlorpyrifos directly from cord blood and therefore has the most robust exposure measurement. Although the results from the other cohorts are useful even if these studies were negated due to non-specific exposure measurement, the Columbia cohort provides a number of strong associations. The effects as described below are seen as early as fetal growth and continue through early childhood with recent evidence of neurodevelopmental effects until age seven.

a) Fetal growth: Statistically significant deficits of birth weight of 186 grams when comparing high exposure to lowest quartile of exposure and decreases of 43 grams in birth weight per log increase in chlorpyrifos in cord blood (Whyatt *et al.*, 2004).

b) Infant neurodevelopment: Statistically significant deficits of 6.5 points on Bayley Psychomotor Development Index (PDI) at 3 years of age when comparing high to low exposure groups (Rauh *et al*., 2006). Notably these decrements in PDI persist even after adjustment for group and individual level socioeconomic variables (Lovasi *et al*., 2010).

c) Increased odds of mental delay (OR=2.4; 95% CI: 1.1-5.1) and psychomotor delay (OR=4.9; 95% CI: 1.8-13.7) at age three when comparing high to low exposure groups (Rauh *et al.*, 2006). When controlling for diazinon and propoxur exposures, chlorpyrifos still showed significant increased odds of mental (OR=3.2; 95% CI: 1.3-8.2) and psychomotor delay (OR=7.9; 95% CI: 2.1-29.1) (Appendix 4, Whyatt & Rauh, 2011 unpublished)

d) Attention problems: Extremely large odds ratios for attention disorders (OR=11.26; 95% CI: 1.79-70.99), ADHD (OR=6.50; 95% CI: 1.09-38.69), and PDD (OR=5.39; 95% CI: 1.21-24.11) were seen when comparing high to low chlorpyrifos exposure groups (Rauh *et al.*, 2006). The magnitude of these results as so large that they are unlikely to be affected by residual confounding although limited sample sizes resulted in imprecise estimates.

e) Intelligence measures: Statistically significant decreases of 1.4% in full scale IQ and 2.8% in working memory among seven-year olds for each standard deviation increase in chlorpyrifos exposure (Rauh *et al.*, 2011). These results persist even when performing sensitivity analyses including only those with detectable chlorpyrifos levels. In addition, no evidence was provided of mediation by child behavior on the measure of working memory instrument.

Consistency of association: This criterion outlines the results from the Berkeley and Mt. Sinai cohorts which were consistent with or supportive of the conclusions

of the Columbia cohort. It should be noted that the Berkeley and Mt. Sinai cohorts did not replicate the effects on fetal growth that were seen in the Columbia cohort. Although the cohorts had similar composition and study design, it should be noted that the Berkeley and Mt. Sinai cohorts used non-specific measures of general organophosphate exposure (TCPy and DAPs). However, the internal validity across cohorts gives confidence in the consistency of the results for the neurodevelopmental outcomes. It should also be noted that neurodevelopmental effects are seen in both of these cohorts beginning at neonatal development and extending to early childhood.

a) Neonatal neurodevelopment: Increased abnormal reflexes in neonates were significantly associated with maternal and urinary DAPs in both the Berkeley and Mt. Sinai cohorts (Young *et al.*, 2005; Engel *et al*., 2007).

b) Infant neurodevelopment: In the Mt. Sinai cohort, prenatal DAP was significantly associated with deficits in Bayley mental development index (MDI) at 12 months among blacks and *Hispa*nics. This as*socia*tion was enhanced among children with maternal carriers of PON1 QR/RR, *i.e*. fast metabolizers (Engel *et al.*, 2011). In the Berkeley cohort significant decreases in MDI at 24 months were associated with increased prenatal and infant urinary DAP measures (Eskenazi *et al.* 2007). Examination by PON1 status also showed evidence of poorer MDI scores at 2 years among those children with the PON1-108T allele (Eskenazi *et al., 20*10).

c) Attention problems: In the Berkeley cohort, total urinary prenatal and postnatal DAP measures were associated with significantly increased odds of PDD at 2 (Eskenazi *et al.*, 2007). In addition, prenatal DAP was associated with ADHD and Child Behavior Checklist attention problems at 5 years. Child concentrations of diethylphosphate (DEP) were also adversely associated with a composite measure of attention (Marks *et al.*, 2010).

d) Intelligence measures: In the Berkeley cohort, a significant deficit of 7 points in full scale IQ was seen among seven year olds when comparing the highest quintile of maternal DAP to the lowest level (Bouchard *et al.*, 2011). In the Mt. Sinai cohort, there were slight but not significant decrement in full scale IQ, perceptual reasoning and working memory associated with prenatal maternal urinary DEP in 6 to 9 year olds. Increased p*renata*l maternal urinary DAP was also associated with decreases in perceptual reasoning in maternal QQ carriers. This association showed a monotonic trend (Engel *et al.*, 2011).

e) Crude exposure response relationship: This was demonstrated in the pre-post residential cancellation analyses in the Columbia cohort in the outcomes of birth weight, birth length, and three year MDI and PDI sco*res (W*hyatt *et al.*, 2004; Rauh *et al*, 2006). In addition a significant reduction in cord blood chlorpyrifos and maternal personal air samples was seen when comparing pre and post cancellation levels (Whyatt *et al.*, 2004). The effectiveness of a

prevention measure can often be shown when reductions in effect can be measured subsequent to a reduction in exposure. This was the case in the natural 'experiment' that occurred during the course of the Columbia cohort. Although the study was not designed to test an exposure-response relationship, decreases in both outcomes and exposure following the residential ban argue for a crude dose-response relationship.

The following uncertainties should be noted:

- Relatively modest sample sizes which limited the statistical power to classify some meaningful differences as statistically significant and to examine the effect of modification by race/ethnicity and other characteristics.
- Relatively moderate to large exposure differences needed to see significant effects, likely due to the modest sample sizes used.
- Exposure at one point in prenatal time with no additional information regarding postnatal exposures.
- Lack of clarity regarding a linear dose-response instead of a potential threshold effect.
- Use of a single or average sample for exposure. Although Whyatt *et al.* (2009) noted moderate but significant correlations between meconium and cord and maternal blood and average urine TCPy, the representativeness of a single point exposure is still unclear. Time-varying exposures or the ability to define cumulative exposures would be preferable.
- Lack of specificity of a critical window of effect and the potential for misclassification of individual exposure measures.
- External generalizability of the cohorts given their unique racial/ethnic and socioeconomic characteristics. However, it should be noted that their exposures were within the range of those seen in NHANES.
- Questions about biologic plausibility due to lack of clarity on mechanism of action, particularly at the low exposure levels seen in the cohorts and the limited and mixed results of animal studies showing neurodevelopmental effects.

One panel member suggested that before the Agency could conclude that chlorpyrifos is likely to play a role in the neurodevelopmental outcomes observed in epidemiologic studies, particularly in the Columbia study, additional analyses need to be conducted.

In order to eliminate the possible causes of neurodevelopmental effects by other pesticides in the Columbia study, it is suggested that EPA should repeat the pre-post residential cancellation analysis done for chlorpyrifos using other pesticide measurements, such as malathion diacid (MDA), a specific metabolite of malathion. The outcomes from those additional analyses will either confirm or reject EPA's preliminary conclusion that chlorpyrifos is likely to play a role in the neurodevelopmental outcomes.

While one panelist agreed with the overall statement, the Panelist also endorsed changes in the phrasing from "chlorpyrifos likely played a role ..." to "chlorpyrifos may [or could] have played a role ." That Panelist noted that TCPy has some serious limitations as a

quantitative indicator of exposure to chlorpyrifos due primarily to its common occurrence in foods. In addition, triethylphosphate (TEP) has some similar limitations particularly within the Berkeley cohort because the usage rate of diazinon in Monterrey County is at least10 times more than the use rate of chlorpyrifos; and diazinon also produces TEP. This ratio presents a dilemma between the characterization of this cohort as farm laborers and the attribution of their higher levels of urinary TCPy to exposure to chlorpyrifos.

In conclusion although the three studies were not comparable in all regards, more similarities than discrepancies were found across them. The Panel concludes that the additional literature since the 2008 SAP continues to support and strengthens the evidence for the conclusion that chlorpyrifos plays a likely role in the adverse effects in child neurodevelopment.

Question 4.3

As discussed in Question 2.0, a mode of action/adverse outcome pathway has not yet been fully elucidated for the potential neurodevelopmental outcomes as a result of prenatal chlorpyrifos exposure. Although this does not undermine the qualitative interpretation of these studies, and the preliminarily conclusion stated above (Question 4.2), the identification of the dose-response for neurodevelopmental effects based on mode of action is not possible. Further, given the urine and cord blood sampling frequency in the study there is a large degree of uncertainty in estimating absolute exposure-response relationships, as opposed to establishing relative exposure groups for evaluating associations. With respect to dose-response, critical durations of exposure, and windows of susceptibility are unknown. In 2008, the SAP cautioned against using the Columbia cohort data for deriving a point of departure due, in part, to only measuring biomarkers (3rd trimester maternal, cord blood, meconium) at one point in time, and because they cannot exclude possibility that the effects seen were due to chlorpyrifos in combination with other pesticides. In 2008, the SAP advised against using data from the epidemiology studies (including the Columbia Mothers and Newborn study which measured chlorpyrifos directly) for deriving a point of departure due to limitations of the exposure assessment in these epidemiology studies for the purpose of risk assessment, *e.g.*, lack of repeated exposure estimates to ascertain more specifically the variability and periodicity of exposure over time (*i.e.*, predominant use of one-time exposure estimate).

a. Due to the limitations of exposure assessment performed in the epidemiologic investigations for the purposes of quantitative risk assessment, the Agency has concluded that the epidemiologic data are not sufficient for deriving points of departure for quantitative risk assessment. The Agency proposes that AChE inhibition data from laboratory animals remain the most appropriate data to use for dose-response modeling and the derivation of points of departure. *Please comment on the scientific evidence that does and does not support this conclusion, as well as the strengths and limitations of the evidence.*

Response

The Panel acknowledged the limitations in the three longitudinal children's cohort studies of estimating chlorpyrifos exposures (*i.e.*, the Columbia study, the Mt. Sinai study, and

the CHAMACOS study), based on the exposure measures collected, and was in general agreement that the data from these studies alone were not sufficient to derive a point of departure (POD) for purposes of quantitative risk assessment. As a panel member noted, these three epidemiologic studies were primarily focused on assessing health outcomes associated with a variety of environmental factors, and were not designed to conduct a quantitative exposure assessment for chlorpyrifos. In addition, the use by the three studies of different exposure matrices (urine, maternal blood, cord blood, and meconium) and different targeted analytes (TCPy, DAPs, and chlorpyrifos) makes the effort of deriving a definitive POD based on those data alone impossible.

Despite the exposure assessment limitations noted for these three epidemiology studies, the Panel recognized the value of these data and urged the Agency to find ways to use the epidemiology studies, and in particular, the data from the Columbia study, to inform the dose-response assessment of chlorpyrifos. Only the Columbia study provided data on measurements of chlorpyrifos in cord blood coupled with neurodevelopmental measurements. As noted by the Panel, if one assumes that cord blood measurements reflect exposure levels during the critical prenatal period for induction of neurodevelopmental effects, then in theory, these would be the ideal data from which to derive the POD for chlorpyrifos in humans. Specific Panel suggestions included using the Columbia data "as an exercise" to derive a POD for neurodevelopmental effects in infants, and analyzing the data from each of the cohorts to put some bounds on the range of chlorpyrifos doses associated with the observed neurodevelopmental effects.

The Panel also recognized the value in developing a functional PBPK model for chlorpyrifos for pregnancy and the prenatal lifestage. Such a model could be used to further characterize the dose estimates in the epidemiology studies, for additional dose-response analyses. Such a PBPK model will become even more important in the event that the Agency might, at some point in the future, decide to move from using AChE inhibition to another outcome. In particular, such a tool could not only relate a dose of chlorpyrifos to a non-AChE outcome but it could also link a dose to the chlorpyrifos or/and chlorpyrifos oxon concentration *in vitro* to a non-AChE target-site *in vivo*.

The Panel expressed concerns regarding the Agency's proposal to use the dose-response data on AChE inhibition in laboratory animals to derive points of departure for the chlorpyrifos risk assessment, and referred to multiple lines of evidence suggesting that adverse neurodevelopmental effects may be attributed to chlorpyrifos doses lower than those that elicit a 10% inhibition of AChE.

This evidence comes from the epidemiological data derived from the three longitudinal children's cohort studies *i.e.*, the Columbia study, the Mt. Sinai study, and the CHAMACOS study. A number of findings of neurodevelopmental outcomes associated with chlorpyrifos are consistent across these three cohorts. For example, there is a consistent association between chlorpyrifos exposure and deficits in mental development at age 7 as ascertained by decrements in full-scale IQ and Working memory using the Wechsler Intelligence Scale for Children (WISC-IV) (Engel *et al.*, 2011; Bouchard *et al.*,

2011; Rauh *et al*., 2011). (See responses to Charge Questions 4.1 and 4.2 for more detailed discussion and assessment of the findings from these studies.)
There are limitations to the exposure assessment in these three cohorts. The Columbia study has the most direct measure of exposure to chlorpyrifos, measuring the compound in cord and maternal blood at time of delivery (Rauh *et al*., 2011). This study also has 48-hr personal air measurements of chlorpyrifos for pregnant women, air chlorpyrifos measurements (stationary samples) collected during the last 8 weeks of pregnancy, urinary metabolite data (TCPy) during the last trimester (up to 4 measurements for some participants) and at delivery for mom and baby, and TCPy in meconium (Whyatt *et al*., 2007; 2009). In an exposure validation study conducted by the Columbia researchers, the levels of TCPy in meconium and maternal urine correlated with cord blood chlorpyrifos levels (Whyatt *et al*., 2009). This suggests that cord blood levels can be used as a representative measure of exposure. Overall, the estimates of chlorpyrifos exposure in the Columbia cohort (based on measured levels of maternal urinary TCPy) were slightly lower, but generally comparable with the levels of urinary TCPy measured in adults in the general U.S. population at that time, based on the NHANES data for 1999-2000 and 2001-2002 (CDC, 2009). The estimates of chlorpyrifos exposure (based on measured levels of maternal urinary TCPy) in the Mt. Sinai (Berkowitz *et al*., 2003) and CHAMACOS (Eskenazi *et al*., 2007; Castorina *et al*., 2010) cohorts were slightly higher, but generally also comparable to the 1999-2000 and 2001-2002 NHANES data for the U.S. adult population.

The Panel suggested that while there are no data on AChE inhibition in either the Columbia study participants (*e.g.*, Rauh *et al.*, 2006; Whyatt *et al*. 2007; 2009; Rauh *et al*., 2011)or the NHANES participants (CDC, 2009), the measured levels of chlorpyrifos exposure are not anticipated to produce AChE inhibition. Specifically, as noted in the Draft issue paper, neurodevelopmental effects seen in the Columbia cohort were associated with cord blood chlorpyrifos levels > 6.17 pg/g (Rauh *et al*., 2006). Based on AChE inhibition studies in adult men dosed with chlorpyrifos (Nolan, 1984), in which AChE inhibition was associated with peak blood levels of 0.01-0.03 µg/ml (more than 10^4 more), blood levels of 6.17 pg/g are unlikely to elicit AChE inhibition.

Additional evidence suggesting that adverse neurodevelopmental effects may be attributed to chlorpyrifos doses lower than those that elicit a 10% inhibition of AChE comes from the *in vivo* animal neurodevelopmental studies.

As discussed in response to Charge Question 3, the Panel concluded there are only 3 animal neurobehavioral studies that evaluated doses below 1 mg/kg and also assessed AChE inhibition—2 found no effects at doses below 1 mg/kg (Maurissen *et al*., 2000; Braquenier *et al*., 2010), and one reported effects at 0.3 mg/kg, but had serious methodological flaws (Jett *et al*., 2001). In addition, as discussed in the Agency's Draft Issue Paper and the SAP public meeting presentation entitled "Adverse Outcome pathway: Data for Chlorpyrifos at Varying Levels of Biological Organization", there are another three *in vivo* neurodevelopmental studies conducted in rats that report effects at doses below those at which acetylcholinesterase inhibition was detected (Ray *et al*., 2010;

Aldridge *et al*., 2004; Aldridge *et al*., 2005). These additional three studies are briefly summarized below.

The study, of Ray *et al* 2010, reported differential expression of oxidative stress genes in rat pup forebrain 24 hours after administration of chlorpyrifos, in the absence of AChE inhibition. Briefly, chlorpyrifos was administered via gavage to 7 day old rat pups at doses of 0, 0.1, 0.5, 1, or 2 mg/kg, and forebrain gene expression and AChE activity assessed after 24 hours. Gene expression changes, including differential expression of genes associated with oxidative stress, were observed at all doses, while inhibition of brain AChE was observed only at the highest dose tested (2 mg/kg). As noted on p. 30 of the Agency's Draft Issue Paper, it has been suggested that oxidative stress can result in dysregulation of signaling pathways controlling neuroprogenitor cell function.

The studies of Aldridge and colleagues (Aldridge *et al*., 2004; 2005) reported a number of molecular, biochemical, and functional changes associated with altered serotonergic tone in the brains of adult rats exposed prenatally to chlorpyrifos at doses shown in separate studies employing the same (Qiao *et al*., 2002) or similar (Mattsson *et al*., 2000) experimental designs to not result in fetal AChE inhibition. Briefly, in the Aldridge studies rats were administered chlorpyrifos (0, 1, or 5 mg/kg in DMSO) by subcutaneous injection on gestation days 17-20, and assessed in adulthood for a number of brain parameters on postnatal day 60. Developmental chlorpyrifos exposure at the 1 mg/kg dose level was associated with increases in serotonin receptors (5-HT_{1A} and 5-HT_2) (Aldridge *et al*., 2004; 2005), increases in serotonin reuptake receptors (Aldridge *et al*., 2004; 2005), increased serotonin turnover (Aldridge *et al*., 2005), and changes in the adenylate cyclase response to serotonin in the cerebral cortex and mid-brain (Aldridge *et al*., 2004; 2005). While neither of the Aldridge studies measured AChE, another study from this group reported that subcutaneous injection of 1 mg/kg chlorpyrifos on gestation days 17-20 had no significant effect on fetal rat AChE levels, which were measured 24 hours after the last administered dose (Qiao *et al*., 2002). The Panel noted that the measurement of AChE activity 24 hours after the last chlorpyrifos dose, and the use of DMSO as the vehicle, raises some concerns about the validity of the Qiao *et al*. 2002 findings regarding fetal AChE activity. These concerns are tempered somewhat by the study of Mattsson *et al*. (2000), in which pregnant rats were dosed with 0, 0.3, 1, or 5 mg/kg chlorpyrifos (in corn oil) from gestation day 6 through gestation day 20, after which fetal rat AChE activity was assessed 4 hours post-gavage. No inhibition of fetal AChE activity was observed at either the 0.3 or 1 mg/kg dose levels (Mattsson *et al.*, 2000).

Evidence that adverse neurodevelopmental effects may be attributed to chlorpyrifos doses lower than those which elicit a 10% inhibition of AChE also comes from the several *in vitro* mechanistic studies that have been summarized in the Draft Issue Paper, demonstrating a variety of effects at the molecular and cellular level, including interference with neurite and axon outgrowth (Das and Barone, 1999; Howard *et al.et al.*, 2005; Yang *et al.et al.*, 2008), reduced axonal transport (Middlemore-Risher *et al.*, 2011), and increased oxidative stress (Crumpton *et al.*, 2000; Qiao *et al.*, 2005; Giodano et al ., 2007; Saulsbury *et al.*, 2009). Briefly, the study of Das and Barone (1999) in PC12 cells

shows that chlorpyrifos interferes with neurite outgrowth at concentrations that do not inhibit AChE, and the studies of Howard *et al.* (2005) in rat sympathetic neurons and Yang *et al.* (2008) in dorsal root ganglion sensory neurons show that chlorpyrifos decreases axonal outgrowth at concentrations that do not inhibit AChE. The studies of Middlemore-Risher *et al.* (2011) show that incubation of rat cortical neurons with chlorpyrifos or chlorpyrifos oxon reduces axonal transport of mitochondria at concentrations that do not inhibit AChE. The studies of Crumpton *et al.* (2000), Qiao *et al.* (2005), Giodano et al. (2007) and Saulsbury *et al.* (2009) demonstrate that exposures of a variety of cell types (*i.e.*, primary cerebellar granule cells, oligodendrocyte progenitor cells, PC12 cells) to chlorpyrifos at concentrations thought to be so low as not to inhibit AChE result in increased levels of reactive oxygen species and oxidative damage (measured as lipid peroxidation).

As mentioned in the response to Charge Question 2.2, there are additional effects that should be included in the EPA review, namely, the effects of chlorpyrifos on nerve growth factors (Pope *et al.*, 1995; Slotkin *et al.*, 2007; Betancourt and Carr, 2004; Terry *et al.*, 2007) and mitochondrial morphology (Middlemore-Risher *et al.*, 2011). Many of these effects have been observed in the absence of AChE inhibition, or at concentrations below which acetylcholinesterase inhibition would be predicted.

In summary, these lines of evidence suggest that chlorpyrifos can affect neurodevelopment at levels lower than those associated with AChE inhibition, and that the use of AChE inhibition data may not be the most appropriate for dose-response modeling and derivation of a point of departure for assessment of the neurodevelopmental risks of chlorpyrifos.

The Panel suggested additional research that could answer the critical question of whether chlorpyrifos induces neurodevelopmental effects in humans at doses that do not cause AChE inhibition. This suggestion was to test whether the chlorpyrifos levels measured in cord blood that were associated with neurodevelopmental effects in the Columbia study would result in either red blood cell or brain AChE inhibition. This study could be easily performed by EPA researchers, or by others.

Additional concern about the use of AChE inhibition dose-response data to protect against neurodevelopmental effects was based on the potential for the outcomes of AChE inhibition and adverse neurodevelopmental effects to be two separate observations, in which the former is the result of an acute exposure scenario and the latter is likely to be caused by chronic low level exposure to chlorpyrifos *in utero*. All 3 cohort studies report neurodevelopmental outcomes associated with maternal or *in utero* chlorpyrifos exposure measures, which are considered to be representative of chronic exposures during the prenatal period. None of these studies assessed AChE inhibition or other acute responses to recent chlorpyrifos exposures.

Additional questions and concerns about the use of the rodent AChE inhibition dose-response data were raised. The AChE inhibition study that serves as the basis for selecting 0.03 mg/kg/day as the POD (BMDL10) for a benchmark response of 10%

AChE inhibition is Maurissen et al (2000). In this repeat dosing study pregnant dams received daily doses of chlorpyrifos by oral gavage from gestation day (GD) 6 to 20, and red blood cell AChE inhibition was measured 4-5 hours after the last dose of chlorpyrifos was administered. One question raised by the Panel is whether the time of AChE assessment was optimal to detect the peak inhibition effect. The Draft Issue paper provides no information on how the time of AChE assessment in this study was justified by the study investigators, although a general statement on p. 17 indicates that the peak inhibitory effect on AChE activity is typically within one to several hours after dosing.

A second question broached by the Panel regarding Maurissen et al (2000) was whether inhibition of AChE had reached steady state in this study. The Panel noted that a similar BMDL10 for RBC AChE inhibition of 0.044 mg/kg/day was obtained from a companion study (Mattsson et al , 2000) that dosed dams for a longer period of time, *i.e*., from GD6 to lactation day 10. This comparison suggests that steady state inhibition of RBC AChE likely had been reached in the Maurissen et al (2000) study.

A more important question is whether the dose-response for AChE inhibition in the pregnant rat is predictive of AChE inhibition in the human fetus. The Panel cautions the Agency on using pregnant rodent and rodent neonatal/juvenile data as the basis for deriving a point of departure for quantitative calculation of dose-response and risk assessment in human pregnancy and human children for the following reasons:
The AChE inhibition is caused by an oxon of chlorpyrifos that is produced metabolically by CYP450 (P450) metabolism. The isoforms involved include P450 1A2, a 2B isoform, 3A4, 2C9 and 2C19 (there may be others). This presents the following problems with extrapolation from rodents to humans:

- Several of these P450s are highly polymorphic in humans, which will cause considerable variation in human responses.
- The polymorphisms existing in humans may be different from those in rodents.
- Since rodents have different homologues and orthologues, metabolic activation rates and extents may differ between rodents and humans based on differing enzyme affinities for chlorpyrifos.
- Several of these P450s are not active (or only active at very low levels) in the human fetal liver and arise in months-to-years after birth, yet their corresponding rodent P450s are commonly present in the fetal rodent liver.

A positive suggestion in this respect is that much of the ontogeny work in humans and rodents has already been performed and ontogenetic differences are known. For human pediatric CYP ontogenies, the Panel recommended that the Agency explore the work of Professors Ron Hines and J. Steven Leeder to determine qualitative and quantitative differences ((e.g., de Wildt et al, 1999; Pearce et al, 2001; Koukouritaki et al , 2004; Nong et al , 2006; Blake et al, 2007, Hines, 2007; Hines, 2008; Stevens et al, 2008)

Question b. The Agency does, however, believe that the epidemiologic data are useful to informing other key aspects of the chlorpyrifos risk assessment including hazard characterization, exposure characterization, and quantitative uncertainty characterization and analysis. *Please suggest approaches/analyses for potentially using the epidemiology*

data in different parts of the chlorpyrifos risk assessment including those noted above. (Note: Some of these may also be covered in Question 5.4 below.)

Response

The framework for integrative analysis to evaluate multiple lines of evidence in the context of understanding the AOP/MOA proposed by the Agency is extremely helpful as a basis for framing thoughts on the weight of evidence and the integration of increasingly varied types of information, including epidemiological data.

In relation to the specific use of the epidemiological data to inform key aspects of the chlorpyrifos risk assessment, this is likely best expanded beyond the scope included in the question – – *i.e.*, "hazard characterization, exposure characterization, and quantitative uncertainty characterization and analysis," since the epidemiological data are also informative in the context of dose-response analysis.

Although the panel was not explicitly charged with making a FQPA safety factor determination, one panel member suggested that the epidemiologic data, which represent a significant portion of the evidence base demonstrating increased sensitivity of early lifestages to the neurodevelopmental effects of chlorpyrifos, be used in selecting the Food Quality Protection Act (FQPA) factor to be applied in the risk assessment. The Panel recognizes that it is constituted as a technical advisory body, not a panel intended to provide policy advice. However, the choice to apply particular FQPA safety factors in the EPA's risk assessment involves both policy and science. The FQPA safety factor recommendation is based on the scientific evidence provided to the panel. As discussed in detail in the responses to Charge Questions 4.1, 4.2, and 4.3.a, the strengths of the three longitudinal children's cohort studies, the consistency of associations of chlorpyrifos with neurodevelopmental outcomes across these studies, and the large effect measures observed for serious long-term neurological effects (*e.g.*, attention problems), coupled with data indicating that chlorpyrifos exposures in these cohorts were generally comparable with those of the general U.S population and unlikely to be associated with AChE inhibition, all suggest that in the event that the Agency continues to use dose-response data for AChE inhibition to derive a point of departure, a FQPA factor of 10=fold is recommended to protect sensitive early lifestages,

<u>Exposure Characterization:</u>

Environmental monitoring and biomonitoring data in the epidemiological studies contribute to the overall database on estimation of exposure, including (particularly) population variability and (to some degree) inter-individual variability in the study populations. They also provide insight into more generalizable observations on temporal trends in exposure of the general population – *e.g.*, following the impact of withdrawal of domestic (nonagricultural) uses of chlorpyrifos.

The biomonitoring and environmental monitoring data from the three children's cohort studies should be used, then, along with exposure information from other studies and sources, to characterize the levels of exposure to chlorpyrifos experienced in different

populations (production workers, agricultural workers, individuals exposed via residential use, general population, *etc.*), and in similar populations over time (*e.g.*, before and after cancellation of residential uses).

Data available from the epidemiological studies also provide unique opportunity to investigate the relationship between environmental levels and results of biomonitoring (*e.g.*, dose reconstruction as described by public commenter, Dr. Dale Hattis) since for some of the studies, both types of data (including air monitoring in the Columbia study) are available.

To some degree, the epidemiological studies can also provide sources of data to consider the suitability of the various biomarkers as measures of short and/or long term exposure to chlorpyrifos.

Toxicological Hazard Characterization:

The epidemiological data contribute to an evolving database on potential toxicological hazards to humans. They have contributed and continue to contribute to hypothesis generation for targeted investigations of developmental neurotoxicity in animal studies. To (limited) degree, they also confirm expectations concerning potentially susceptible subgroups based on mode of action – *i.e.*, the PON 1 genotype. They also provide some information on the extent of impact of other factors, which in combination with chlorpyrifos, may have an impact on the observed effects.

These studies represent the key datasets that support the identification of chlorpyrifos prenatal exposures as causing neurodevelopmental effects in humans. Important elements to discuss in their evaluation include i) consistency in the findings of neurodevelopmental effects across these three studies, and ii) comparison of the levels of chlorpyrifos exposure experienced in these cohorts based on biomonitoring (blood and urine measurements of chlorpyrifos, metabolites, *etc.*) and environmental monitoring measures (*e.g.*, personal air monitoring in the Columbia study) with data collected in other studies of the general U.S. population (*e.g.*, NHANES), for similar time periods (*i.e.*, pre- and post-cancellation of residential uses).

Quantitative uncertainty characterization and analysis:

It seems important to address this aspect in the context of dose-response analysis, given particularly, that EPA has concluded that the current epidemiological database strengthens the 2008 SAP conclusion that "chlorpyrifos likely plays a role" in observed adverse effects on child neurodevelopment (specifically those reported by Columbia University). There is also a need to address the consistent epidemiologic findings of significant, long-term neurodevelopmental effects across the three cohorts at levels within the same range as those in the general population since this would seem to suggest that these effects occurred at exposures below those associated with AChE inhibition.

As a minimum, then, it seems important to maximally utilize available data on dose-response from these studies to at least "bound" reference doses developed on the basis of animal data (Given that this was also recommended by the 2008 SAP, prioritization of this work seems critical.). However, the scientific weight given to the different measures of dose and of response necessarily needs to take into consideration that most of the effort in the epidemiological studies has been directed to the assessment of outcome rather than exposure. In addition, the use of different exposure matrices (urine, maternal blood, cord blood, and meconium) and the difference in the targeted analytes (TCPy, DAPs, and chlorpyrifos) complicates derivation of the POD based on epidemiological data, uncertainties which need to be assessed in dose-response evaluation and risk characterization.

In addition, given the potential significance of the observations in the epidemiological studies, it is also clearly desirable to consider at least semi-quantitatively the potential impact of factors of study design and interpretation that bound the dose-response relationship from the human studies. It would be helpful, for example, to consider systematically (and at least semi-quantitatively) the potential impact on the reported dose-response analysis of exposure measurement error, outcome ascertainment, confounding variables and statistical analysis.

For example, in relation to limitations of data on exposure in the epidemiological studies, a Panelist noted that despite a fairly high portion of the samples whose results were below the limit of detection or quantification for whatever was being analyzed, little use was made of techniques to integrate non-quantified samples into the statistical test. [One of the studies utilized a method described by Richardson and Ciampi (2003).] Various methods were reviewed by the July 2010 SAP that can be applied to either normally or lognormally distributed data that include a significant (even a majority) of non-detectable sample Specifically, the use of "probability plots" was described that can yield an estimate of the geometric mean of the distribution [GM], the geometric standard deviation [GSD], and corresponding percentiles. Various aspects of the technique are described in publications such as Cunane (1978), Haas and Scheff (1990), Travis and Land (1990), Helsel (1990), Hattis and Burmaster (1994), and Hattis *et al.* (1999). Another method called the "maximum likelihood estimate" is not recommended for data sets with a large number of measurable values (Cohen, 1961; Perkins *et al.*, 1990).]

As a basis to increase the confidence in the selected point of departure, a relatively simple experimental protocol to determine whether chlorpyrifos levels measured in the cord blood in the Columbia study inhibit either red blood cells or brain AChE inhibition would be helpful. This seems to be an important priority, given that human data (*e.g.*, coupling of chlorpyrifos measurements in cord blood with neurodevelopmental measurements from the Columbia study) would typically be preferred in estimating dose-response relationships (and particularly for potentially susceptible age groups, such as infants).

The outcomes from the above exercise should contribute to consideration of the critical question of whether or not "a causal association between chlorpyrifos and neurodevelopmental effects in the absence of AChE inhibition is plausible for humans."

Given that AChE inhibition results from acute exposure and adverse neurodevelopmental effects are likely to be caused by chronic low levels of chlorpyrifos, it is important to verify whether or not maintaining long-term exposure to levels below those likely to cause AChE inhibition is likely to be sufficiently protective to prevent neurodevelopmental effects.

With regard to quantitative uncertainty characterization, the results and the uncertainties associated with the dose-response analysis of the neurodevelopmental epidemiology findings should be taken into consideration, along with uncertainties in the dose-response assessment for acetylcholinesterase inhibition, data gaps and database uncertainties regarding whether neurodevelopmental effects or acetylcholinesterase inhibition is the most sensitive endpoint in humans, and uncertainties associated with pharmacokinetic differences due to lifestage and genetic polymorphisms in metabolic enzymes.

Question 5.0 Exposure Profile & Biomonitoring Research

Question 5.1

c. Section 5 of the draft issue paper presents an overview of the principal chlorpyrifos biomarkers and a comparison of biomonitoring studies that measured urinary TCPy levels in a range of study populations involving both the general population and potentially vulnerable populations, including children, workers, and farm families. *Please comment on the degree to which the Agency identified the primary chlorpyrifos biomarkers of exposure, appropriately discussed the strengths and limitations of such biomarkers, and how the strengths and limitations affect the interpretation of the chlorpyrifos biomonitoring data.*

Response

The draft paper was thorough in its coverage of the literature on chlorpyrifos and its biomarkers of exposure. Considering the availability of standard methods, the specificity of the biomarker, the number of laboratories capable of making the measurement, and the relevant concentration levels, the first choice for a biomarker would be chlorpyrifos in blood. The Panel recognizes that this is the most difficult assay and represents only a small percentage of the literature, but it s deemed to be the highest priority because of its specificity.

The next biomarker of choice is TCPy, then DETP/DEP in urine. These have roughly the same equivalence and neither is close to the validity of measuring chlorpyrifos directly in blood because they are both present in the environment as degradates of the active ingredient. Total DAPs (as DMP and DEP) are not selective enough to be a useful biomarker for chlorpyrifos although it may be more appropriate in a global risk calculation model because all AChE inhibiting chemicals should be considered together when evaluating risk. The Panel recognizes the inability of urinary TCPy to distinguish between exposure of chlorpyrifos, chlorpyrifos-methyl, trichlopyr as well as direct exposure to TCPy (a chlorpyrifos degradatein the open environment). However, TCPy is more selective than any of the DAPs and currently is the most selective of the urinary metabolites but questions remain about its efficacy because the Panel believes there could

be significant contribution from environmental and dietary TCPy as a chlorpyrifos degradate.

More emphasis should be placed on the direct intake of the environmental degradate TCPy, mainly present in foods. As early as the late 1990s, the Ryan group (See MacIntosh, *et al.*, 1999) had identified an anomaly in that the amount of TCPy found in urine was substantially greater than the measured likely intake of chlorpyrifos. This work has continued with the papers by Morgan, *et al.*, Wilson, *et al.*, and Lu, *et al*. 2005, 2008, indicating the presence of degradates in foods. Radford *et. al*., 2012 have continued this work on the kinetics of this process. While this work has not yet been published, and the other works are mentioned in the Issues Paper, insufficient emphasis has been placed on the presence of TCPy in food or other exposure media (dust, air particulate, *etc*.) putting into question the utility of urinary TCPy as a useful measure of exposure to the parent compound. During the discussion it was pointed out that Lu et. al., 2005 found that roughly 30% of the TCPy measured in urine could be coming from TCPy directly, present in foods.

The Panel also recognizes the ability to measure AChE and BuChE as biomarkers of exposure, but they are even less specific than DAPs. They are however more indicative of potential health risk and are more than just a biomarker of exposure. Unfortunately the ability to measure these enzymes is likely to vary widely from lab to lab and method to method as they are difficult to calibrate. Changes in cholinesterase activity after an exposure should probably be evaluated more within a laboratory (especially via the use of an unexposed control group) than across laboratories or from study to study.

From its earliest years, measuring AChE has been subject to unresolved inter-day variability (Gage, 1967). For instance, Grob and Harvey (1958) could measure AChE in replicate samples on one day with a standard deviation of ±3% but only to within ±5% on separate days following storage of hemolyzed RBC. The literature dating as far back as Gage (1967) has recommended that in order to measure small changes within an exposed group's cholinesterase activity, researchers should collect blood from an unexposed group of controls, measure the cholinesterase activity in their blood at the same time as the exposed group, and apply a correction factor based on the daily change in the mean of the measured activity in the unexposed group of controls. Yager *et al.* (1976) collected 10 blood samples from 10 unexposed people over five weeks and found that the measured intra-individual coefficient of variation for RBC enzyme activity of ±10% could be reduced to ±6% by controlling the day-to-day component of the variance (*i.e.*, accounting for a shift in the average laboratory results from one day to the next). They also found that plasma activity is more variable between individuals but less variable day-to-day. Similar findings for plasma ChE were reported by Trundle and Marcial (1988) and Brock and Brock (1990).

In an occupational (or other repeated exposure) dose-response study, it is generally cost-effective to adjust the blood ChE results of each member of the exposed or "test" group for the change in the laboratory's reported mean blood ChE of an unexposed or "control" group analyzed at the same time (typically the same day as the post-exposure group or in

the same batch if they were stored). This adjustment has traditionally been made in proportion to the change in the mean AChE of the unexposed controls; however, this form of adjustment could also be applied to plasma ChE except using ChE values. The more accurate fraction of inhibited enzyme [ΔAChE] would be calculated for each subject using this adjusted activity. The final variance of the group would decrease in proportion to the square-root of the number of subjects within the study.

In the future the phase II conjugation products of chlorpyrifos (namely, glucuronides and sulfonates) should be considered. Quantifying conjugative metabolism will ensure that levels of biomarkers are correctly interpreted with respect to biomonitoring data and for performing reverse dosimetry. Even though the AChE adducting oxon is not conjugated, the TCPy and DETP metabolites are extensively biotransformed by the glucuronosyl transferases and sulfotransferases, although the precise isoform pathways are not yet known. Therefore, particularly in the fetus and child, if glucuronidation or sulfonation are saturated and/or ontogenetically deficient, then TCPy and DETP may accumulate. This would almost certainly cause error in biomarker analysis through overestimates of exposure. Moreover, accumulation of these metabolites may present the opportunity for direct metabolite toxicity. Panelists noted there has not been significant effort to look at either the glucuronide or sulfate metabolites possibly because these metabolites have only recently been evaluated both from a physiological and analytical perspective.

The oxon is believed to be the most toxic of the metabolites of chlorpyrifos and is not an environmental degredate. While the oxon does not exist long in the blood, a method to directly measure the oxon in blood is likely to be available in the near future. As the most toxic form and an exclusive measure of exposure to chlorpyrifos, the chlorpyrifos-oxon may be the most predictive biomarker of risk, once a method is published.

When evaluating any of the biomarkers in blood, the EPA will need to consider that some of these biomarkers will differ in concentrations between cord blood and maternal blood as they will have different lipophilicity.

Measuring multiple metabolites simultaneously and then taking ratios of metabolites such as TCPy/DETP represents an untested route to provide greater discrimination between exposure to chlorpyrifos and its degradation product TCPy. However, the Panel could find no direct studies on the stability of these two degradates in the environment. By using this ratio and assuming that the ratio of degradates-to-active in the environment is different from the 1:1 ratio that results from metabolism, it may even be possible to do a source apportionment and separate exposure to the degredate from exposure to the active ingredient.

<u>Other considerations:</u>

The Agency suggested in their public presentation that meconium could be used as a biomarker of fetal chlorpyrifos exposure throughout pregnancy. chlorpyrifos in meconium represent the unmetabolized pesticide. The metabolized form oxon or TCpy may have arrived at the fetus in that form rather than having been metabolized by the

fetus itself. Although there should be chlorpyrifos or metabolites in amniotic fluid if it is found in meconium, no studies exist as to the residence time, flow or amount. It is conceivable (if not totally likely) that chlorpyrifos and metabolites are sent directly into the fetal blood (across the placenta) and that meconium picks it up from sloughed cells. More importantly, the utility of meconium as a cumulative biomarker is uncertain. More specifically, the Panel suggests that this is not (currently) a good idea for the following reasons:

1) No studies of chlorpyrifos in amniotic fluid have been performed. The ratio of aminotic fluid chlorpyrifos to metabolite may add evidence that the fetus is actually metabolizing the chlorpyrifos (if they are developed enough to metabolize the chlorpyrifos).

2) The diffusion and/or transport of chlorpyrifos across the placenta (in either direction) is unknown, but since it is rather fat soluble, equilibration with maternal serum might be postulated. This does not seem to be the case with at least one umbilical: maternal serum study presented at this meeting (Yan, 2010).

3) Metabolism across the placenta is unknown. Are the metabolites passed or only the chlorpyrifos, possibly the oxon?

4) The contribution of umbilical tissue, including any adducting of cord tissue for example by the oxon is unknown. However, since umbilical tissue is so well perfused, it may be expected to be a target for oxon binding. Umbilical cord tissue consists of a polymatrix of Wharton's jelly, which is made up of mucopolysaccharides. (Kliman, 1998). Based on these characteristics, the very fat-soluble nature of chlorpyrifos and its relatively fat soluble metabolites, the umbilical cord would not be expected to function as a good reservoir of the parent compound or metabolites, but may be a target for oxon binding and deregulation of pregnancy homeostasis.

5) Meconium, being composed mostly of intestinal epithelia, lanugo, mucus, amniotic fluid, bile, and water, is reasonably hydrophilic and thus should also be considered a poor matrix (reservoir) for chlorpyrifos and other fat-soluble xenobiotics to accumulate. It may be marginally better for the metabolites TCPy and DETP, but these molecules would also, in addition to diffusion, have some net flow in the paracellular pathway. Thus, they may over represent the exposure to chlorpyrifos (as described above).

6) Human placental studies of chlorpyrifos metabolism and transport have not, to the best current knowledge been published, and this is a limitation of data available to the Agency. Several important points are already known from the illicit drug literature and should be considered when attempting the same type of monitoring for chlorpyrifos (or other xenobiotics). For example, antipyrine (an amphetamine derivative) is used in placental perfusion experiments as a marker of pure diffusive transport with effectively no barrier (Schneider et. al. 1972). In contrast, cocaine and cotinine show differing and, slightly less fat-soluble profiles. For example, cocaine is transferred across the placenta

at only 80% the rate of antipyrine (Schenker et. al.,1993) and some studies have suggested that the placenta acts as a depot for cocaine accumulation preventing transfer to the fetus (Simone et.al. 1994). Additionally, previous studies have indicated that while nicotine (again highly fat soluble) is transferred into the fetal compartment up to 5 times the concentration in the maternal blood, cotinine concentrations in the fetal compartment were considerably lower than corresponding maternal serum levels (Luck et. al. 1985). Again, it has been suggested that cotinine adducts the placenta, preventing equilibration of concentrations between maternal and fetal systems. These studies support the need for greater consideration of the trans-placental characteristics of chlorpyrifos, and since placental characteristics change drastically by term (the placental barrier becomes increasingly "leaky" after ~36 weeks), placental studies need to consider each trimester. In the first trimester, the placenta is perfused only after ~8 weeks; prior to 8 weeks only active transport or diffusion across the placenta can occur because villi are being blocked. Analgous studies for chlorpyrifos are recommended before extrapolating fetal exposure and may be included as part of a longitudinal study in pregnancy. Such a longitudinal study may present additional problems.

The real question for the Agency is almost certainly not related to the fetal load of chlorpyrifos or its metabolites at birth or even at discrete pregnancy time points. The exposure information (fetal load) needs to be correlated to a time in fetal development when the fetus is susceptible to effects of chlorpyrifos, perhaps during critical points of neurodevelopment. Unless the time of these exposures can be definitively correlated with specific adverse health effects, then consensus on how to relate fetal effects to a biomarker concentration is unlikely. Rather, the Agency seems to be seeking to quantify the amount of maternal chlorpyrifos ingestionor/exposure is subsequently experienced by the fetus. In general, the half-lives of chlorpyrifos and its metabolites are rather short. This means that even in the case of TCPy, which has the longest systemic residence time, the terminal half-life (*i.e.,*. complete clearance of TCPy from the fetal compartment) would occur within several days. Therefore, unless the pregnant woman is exposed to chlorpyrifos either chronically or acutely exposed but within a few days of testing, quantifying the chlorpyrifos exposure of the fetus would be difficult. It would require collection of samples from pre-term as well as [full or near full term fetal tissues or sampling directly from placentas (such as chorionic villus sampling), amniotic fluid (amniocentesis), or umbilical blood. A longitudinal study would almost certainly be needed to determine exposure over pregnancy, which may not be cumulative but pulsatile.

These points highlight the uncertainty of using meconium as a measure of exposure over the course of pregnancy at this time. Essentially, production of meconium is from fetal swallowing of amniotic fluid as well as some sloughing of intestinal epithelia, and meconium should not be thought of as a matrix into which chlorpyrifos or its metabolites may accumulate by simple diffusion through fetal tissues.

d. Section 5 of the draft issue paper compares biomonitoring findings from the three children's health cohorts with other major observational exposure studies in the United States. Based on comparison with NHANES 2001-2002, median TCPy levels in the

CHAMACOS and Mount Sinai cohorts were slightly higher than in the general population. It should be noted that the exposures experienced by the CHAMACOS and Mount Sinai cohorts overlapped the start of the residential chlorpyrifos phase-out. By contrast, median TCPy levels in the Columbia cohort, for which sampling occurred when chlorpyrifos use should have rapidly declined due to the voluntary cancelation, were slightly lower than the levels measured by NHANES in the general population. *Please comment on the adequacy of the Agency's comparison for the purposes of evaluating chlorpyrifos exposure levels in the three children's health cohorts. Are there any additional biomonitoring studies that should included in the Agency's comparison?*

Response

The human studies discussed in this section are the best available. They were carefully designed and well implemented. They do, however, look at specific types of exposure: agriculturally based exposure and exposures in city dwelling units likely treated for insects on a regular basis. Further, they span a range of times from when chlorpyrifos use was ubiquitous through the phase-out of indoor uses of the insecticide. Because of this, there are "inconsistencies" in the data that are indicative of changes in use patterns. Current use in indoor settings is dominated by pyrethroids rather than chlorpyrifos. Agricultural settings are still likely to see large exposures to chlorpyrifos (although apparently not in the county surrounding Salinas, CA). There appears to be inconsistent recognition of this change, especially in light of comparisons with "group norms" via, for example, the NHANES studies. It would be to no one's surprise if the 1990-2000 NHANES data indicate higher exposures to chlorpyrifos in residential settings than the later data. Among these three studies the Panel believes the Columbia study has a particular importance because it has data collected before and after the indoor use "ban," and the results reflect the pathway from exposure to biomarker concentration and health outcome.

The Panel recommended the following order in which the studies should be considered. They believe that the next NHANES data set may be the most important as it is likely to reflect the decrease in exposure caused by the voluntary removal of chlorpyrifos from the home market. If the levels progress in a manner similar to those predicted by the decrease demonstrated in the Columbia study, the risk from chlorpyrifos might also decline as rapidly. Even if this is true, chlorpyrifos as a model compound for a risk paradigm that includes epidemiological, dose reconstruction, PBPK modeling, and exposure dosimetry, requires a much broader consideration of studies. The Agency seemed to concentrate on studies that include a reported health outcome, and the Panel wonders why these studies were the principal focus as many studies provide data on exposure and dose. For example to be protective the agency should consider the National Human Exposure Assessment Survey, (NHEXAS-Az, summarized in Egeghy et al. (2011) study many of the participants from Arizona were exposed through agricultural application of chlorpyrifos and this represents the highest non manufacturing level of exposure and may continue to represent direct or indirect agricultural exposure.

Many of the studies listed in the draft paper but not directly discussed should be considered when estimating dose and subsequently risk. The NJ studies where cord

blood measurements were used as the principal sample type are important because that is likely to be the desirable biomarker and used more frequently in future studies. Farm-workers studies are important because their families are likely to be one of the remaining populations that continue to see significant exposure, again an expectation to be validated by the next round of NHANES data. They should however probably be considered primarily in relation to farm workers' families. The Children's Pesticide Exposure Study (CPES) by Lu, 2009 and Children's Post-Pesticide Application Exposure Study (CPPAES) studies are important because they provide data on multiple exposure vehicles/media and will be especially useful in dose reconstruction. Dose reconstruction will be paramount in validating PBPK models using media (dust, food, air particulate) measured concentrations and estimated exposure levels, to be subsequently discussed sections 5.3. Among the current studies those that look at both the urinary concentrations and the media where the exposure is likely to occur, will provide the best models for closing the knowledge gap between exposure and dose; and studies where urine was collected within one half-life after a fresh exposure may provide the most useful information.

Although not ready for this report, studies now underway that are longitudinal in design will afford a better understanding of actual exposure profiles when compared to cross-sectional approaches. Due to the short biological half-lives of the metabolites of chlorpyrifos in the body, a spot check of a relatively small number of people may not be enough to represent the exposure of a vulnerable population at key time periods. Only a longitudinal investigation can get at these important data.

Several new studies of interest to this group have been completed and will be published in the near future that are. The Children's Pesticide Exposure Study, led by Dr. Alex Lu of Harvard School of Public Health, focuses on dietary intake of children and related pesticide exposures. The Children's Exposure to Environmental Pesticides, led by Dr. P. Barry Ryan of Emory University, evaluated the utility of biomarkers of pesticide exposures, *e.g.*, DAPs and pesticide-specific markers of OP and pyrethroid exposures, and environmental levels measured in soil, house dust, and food. The target population is children ages 3-6. The SAWASDEE study, led by Drs. Dana Boyd Barr and Ryan, and Dr. Tippawan Prapamontol of Chiang Mai University in Northern Thailand, examined pesticide biomarker concentrations in pregnant mothers, and similar markers in their newborn children. Multiple measurements in both urine and serum have been made throughout pregnancy giving a better longitudinal picture of exposure. Several smaller investigations are underway designed to evaluate the direct intake of pesticide degradates and to evaluate the kinetics of the degradation process in environmental media, including food.

In comparing the results from study to study, it is important confirm that analytical results are directly comparable. Data quality of some studies has been called into question due to apparent changes in limits of detection associated with two analytical methods developed by Center for Disease Control (CDC) used to evaluate serum chlorpyrifos concentrations. The questions arose due to a misunderstanding of the methods. There is an apparent 20-fold difference in the limits of detection (LOD)

between the two methods. This can be accounted for in three ways. First, the "newer" method uses a sample size one-half as large as the "older" method, and injects one-half as large an aliquot thereby accounting for a factor of four difference in LOD. Second, although both methods are multi-contaminant, the newer method spans a much larger range of analyte polarities. In order to obtain adequate recoveries for some of the less polar compounds, there is some sacrifice in sensitivity toward more polar compounds, such as chlorpyrifos.

Third, the newer method was developed with the expectation that higher concentrations would be evident in the samples analyzed, hence precluding the need for a lower limit of detection; a listed limit of detection of 10 ppb was adequate for the purposes of the study. Attribution to the new, higher limit of detection to samples analyzed by the older, more sensitive method, is therefore not warranted. The value for the LOD determined for the earlier method should be viewed as appropriate for the samples analyzed by that methods and deemed useful for presentation in any other work.

In addition to analytical differences these studies (Columbia, CHAMACOS and Mount Sinai) are all cross-sectional in design with some repeated measurements during the pregnancy period. Because of the cross-sectional design coupled with the short biological half-life of chlorpyrifos, the spot urine measurement would be highly affected by daily chlorpyrifos exposure, as well as the timing of sample collection. It would be great if all three epidemiologic studies were using the identical sampling protocol so the outcome measurements could be compared across the board they weren't. Generally speaking, it should not be surprising either to see the similarities in the CHAMACOS and Mount Sinai cohorts during the period they overlapped the Columbia study, before the residential chlorpyrifos phase-out. It is likely that dietary exposure to chlorpyrifos in these two cohorts may represent a portion of the overall exposure. However, the Columbia cohorts may differ from the CHAMACOS and Mount Sinai cohorts because the Columbia study reported the reduction of chlorpyrifos in the indoor air after the phase-out.

Question 5.2

In Section 5.0 of the draft issue paper, the Agency summarized the 2008 preliminary findings on the association between urinary TCPy levels and AChE/BuChE inhibition and discussed two recent studies involving manufacturing workers in the US and Egypt. *Please comment on the scientific quality of these studies and their findings. Please include a discussion of their strengths and limitations. Please comment on the strengths and limitations of the evidence from this research to show an association between TCPy and AChE/BuChE inhibition at exposure levels experienced by occupational populations.*

Response

Both of the occupational exposure studies were observational in nature. Garabrant *et al*. (2009) involved 53 workers manufacturing chlorpyrifos in Michigan, while Farahat *et al*. (2001) involved 38 field workers applying chlorpyrifos onto cotton plants in Egypt. Both of these studies contain data that have multiple sources of imprecision (as will be detailed below), but they both included enough participants that their overall results match PBPK

model predictions quite well. In many ways both studies were well designed and implemented. Both studies had sufficient power to show an association between TCPy and AChE or/and BuChE inhibition at exposure levels experienced by occupational populations. In fact, the PBPK model and cholinesterase data confirm that chlorpyrifos once absorbed interacts first with BuChE and only starts to inhibit RBC AChE and AChE in the central nerve system after BuChE is more than 50% inhibited.

Perhaps the most unique feature of the Farahat study was the extremely high levels of TCPy found in urine from these field workers after applying chlorpyrifos to the target cotton fields. For example, the mean post-exposure values of urinary TCPy were about 25× more than the TCPy from the manufacturers reported by Garabrant *et al*. and over 1000× more than those in the women and children cohorts discussed in Section 4. On the one hand, the Panel pointed out that this contrast made this study less relevant to our discussion. On the other hand, a major strength of the study is that not only were the qualitative patterns of both BuChE and AChE activities when paired to urinary TPCy from the same individuals qualitatively similar to the patterns predicted using the PBPK model described by Timchalk *et al*. (2002) and used by Garabrant *et al*. (2009), but the "inflection points" within the paired data closely match those predicted by the PBPK model. This correspondence between the measured and predicted TCPy excretions and cholinesterase inhibitions is strong evidence for the robustness of the PBPK model over a wide range of exposures.

The Panel pointed out five weaknesses within the Farahat study for use within the weight of evidence. First, virtually all of these field workers had high levels of TCPy in their pre-exposure urine samples. These background concentrations (with sub-group means ranging from 10 to 2000 µg TCPy/g creatinine) were up to three orders of magnitude higher than the levels in the women and children epi cohorts. The source of this background is unknown but seems likely to have been due in large part to these workers' prior use of chlorpyrifos outside of the jobs being studied and possible contributions from TCPy on chlorpyrifos treated food and from TCPy or/and chlorpyrifos within homes treated with chlorpyrifos. Second, the urine samples were collected from morning voids that a study by Lu *et al*. (2006) found to be less reliable than evening voids. Nonetheless, these high background TCPy levels jumped about 30× after the applications began. Thirdly, the cholinesterase values were measured by the battery-powered kit based on the Ellman method. Prior publications (including one by the same researchers who participated in the Farahat study) concluded that cholinesterase activities measured by those field test kits are not as reproducible either from kit-to-kit or as a function of temperature as those using more robust clinical methods (Oliveira *et al,* 2002; Hofmann *et al*., 2008). Fourth, the study was not designed to analyze blood samples from an unexposed control group concurrent with blood from their field workers; the importance of such control was discussed in the Panel's response to Charge Question # 5.1. This deficiency further weakens the precision of their cholinesterase results which was offset somewhat by having 38 participants. Lastly, for reasons not stated, the authors chose to report (and plot) individual cholinesterase activities rather than inhibitions in comparison to individual baseline values. Thus, the reader is led to believe that the ratio of an activity of 2 U/g Hgb for the individual with the lowest AChE and the highest TCPy to

about 25 U/g Hgb for the cluster of individuals with the highest AChE and lowest TCPy measurements (in their Figure 3) represents an inhibition of almost 90%. While some found this degree of inhibition incredulous, Grob *et al.* (1947) and Grob and Harvey (1958) showed that a sequence of small oral doses of an OP (DFP) delivered over three to five days can cause someone's AChE to be reduced down to about 1% of their normal level or to be 99% inhibited) but still not cause symptoms if delivered slowly enough. But of course, a fractional ΔAChE inhibition of 30-50% in one day can cause acute symptoms (*e.g.*, Gage, 1967; Reigart and Roberts, 1999). Thus, the idea that any particular level of ΔAChE either is or is not clinically important depends on more than just its numeric value. Another troubling observation in Farahat *et al*. (2011) is the persistent elevated TCPy measurements in some of the workers and the persistent depressed RBC AChE 14 days post-application; perhaps these lingering effects are linked to the high preexposure levels or the inhibition may have "aged." With these caveats, not only does the pattern of paired levels of AChE activity and concentrations of TCPy in urine qualitatively match the pattern predicted by the Timchalk PBPK model, but also the value of the mean of four measured AChE inflection points at 3161 µg TCPy/g creatinine quantitatively matches the inflection point predicted for AChE by that model.

The study reported by Garabrant *et al.* (2009) has some broadly similar and some different weaknesses for use within the weight of evidence. One different weakness is the greater potential for a proportion of the chlorpyrifos employees' urinary TCPy to have come from doses of residues of TCPy that might have accumulated within the manufacturing workplace (Burns *et al*., 2006). This study added urine collection to an on-going occupational health monitoring program that involved monthly blood samples that were analyzed for cholinesterase via a proprietary system (*Vitros* by Johnson & Johnson) with which the Panel was not familiar. The time at which the pre-exposure cholinesterase was measured was not stated but could have been some years earlier. Despite the study having a "referent group," there is no indication that the cholinesterase results for the chlorpyrifos workers were adjusted for variations in the results of blood samples from an unexposed control group (again see CQ#5.1). The three urine samples per person collected in this study were also collected in the morning (first voids in this case); however, an additional source of uncertainty was introduced into the results of Garabrant *et al*. because the blood and urine samples were collected between 5 and 14 days apart. The authors concluded that conducting paired analyses using only the 48% of the urine results that were collected within 7 days of a blood sample was optimum; however, this interval spans several half-lives for TCPy within the human body. The range of ΔAChEs reported in this study slightly exceeded ±20% but, as predicted by the PBPK model, showed no correlation with TCPy. Only the BuChE inhibition could be attributed to chlorpyrifos exposures. Indeed, the inflection point for ΔBuChE found by this study (110 µg TCPy/g creatine)) not only matched that found by Farahat *et al.* (114 µg TCPy/g creatinine) but also matched that predicted by the PBPK model.

In the responses to Charge Questions 4.2 and 4.3, the Panel suggested that the Agency should separate scenarios for occupational exposures, as reported in these two studies, from exposures from environmental sources. Indeed, one panel member suggested that data from Farahat *et al.* (2011) should not be considered for any further uses. It should

be noted that the subjects in these two studies were adults. Although participants in Farahat's study were as young as 15 and roughly 25% of participants in Garabrant's study were females, none are directly comparable to newborn infants. Even the extrapolation of any working population to the population as a whole is subject to criticism. Such criticisms include the "healthy worker effect" and the idea that low-level exposure and high-level exposures are likely to be detoxified by differing mechanisms. Studies of agricultural workers and their families could offer a better avenue of investigation that compares "occupational-levels" exposure with other members of their families likely see slightly "elevated" but lower levels of exposure, and to study the potential impact on the offspring in such cohorts exposed either *in utero* or otherwise. In the future, the Agency should take into account the quality of ChE measurements prior to further uses in the exposure and risk assessments.

Question 5.3
Several approaches ranging from qualitative to the most sophisticated PBPK/PD modeling approach were introduced as potential options for analyzing the chlorpyrifos biomonitoring data. *Please comment on the strengths and limitations of these approaches. In addition, please suggest, if appropriate, alternative approaches or analyses not identified by the Agency.*

Response
The increasingly data-informed options for interpreting biomonitoring data presented by the Agency range from qualitative (non-comparative, looking at trends or comparative, taking into consideration controlled human studies data where ACN inhibition has been measured) to semi quantitative approaches (estimating biomarker levels associated with regulatory exposure guidelines or estimating exposures from biomarker levels using reverse dosimetry or a PBPK model).

Presentation of a number of options in an increasingly data-informed construct of this nature has potential to maximize the use of biomonitoring data for different applications (accounting internally for more factors contributing to variability in exposure than do external estimate), taking into account (relative) uncertainty depending on: availability and specific nature of biomonitoring data, and the intent of use (*i.e.*, what degree of uncertainty is acceptable for the intended purpose; what population; and what application?).

The selection of appropriate options is necessarily dependent on the extent of the data available on toxicokinetics relevant to the population subset and mode of action, and their integration, with a verified PBPK model having the potential to be the most informative, but being the most data intensive. In relation to intended application, for example, if the objective is media specific assessment or management, dose reconstruction (reverse dosimetry) from biomonitoring data is required.

As a minimum, currently, the biomonitoring data on chlorpyrifos should be helpful in "ground truthing" total external exposure estimates under various use conditions, which

are necessarily based on many more assumptions such as activity patterns and intakes and concentrations in various media.

Given the availability of biomonitoring data on chlorpyrifos in the general population, and as a basis to encourage its maximal consideration in a public health risk context, the Agency is also encouraged to seriously consider the development of a value akin to a "biomonitoring equivalent" concurrently with the derivation of a reference dose for chlorpyrifos. (A biomonitoring equivalent (BE), is a calculated level of a biomarker associated with exposures consistent with health protective guidance values for the general population). This BE would provide a valuable addition for interpretation of population biomonitoring data with limited additional effort, drawing efficiently on the existing process for review and consultation for the regulatory assessment (*i.e.*, BEs are based on similar considerations as the reference dose but incorporating toxicokinetic translation to internal doses).

Clearly, a verified PBPK model provides the most robust opportunity to integrate the considerable available data on external and internal exposure (*i.e.*, biomonitoring) to chlorpyrifos at different life stages under different conditions of exposure.

As indicated in the response to Q. 5.4a), prediction of excretion by the PBPK model can potentially be validated or verified with an accurate estimate of dose, through dose reconstruction based on data from the epidemiological studies on the concentration of chlorpyrifos in media such as house dust, air and water combined with market basket data on the concentration of chlorpyrifos on food. This would permit the effective prediction of exposure at the critical windows of maximum effect (*i.e.* AChE suppression) with measured urine concentrations. However, it's somewhat unclear currently based on input at the meeting from Dr. Bartels of Dow Chemical and Agency staff whether or not the developed PBPK model is life-stage specific. In the interest of addressing this need, the following recommendations are offered: If an adult PK or PBPK model is used, simple allometric scaling (3/4 power) or scaling based on Wang's modification of the Dubois and Dubois equation (Wang *et al.*, 1992) can be useful, relatively accurate and robust for extrapolating to children (Anderson *et al.*, 2009; Anderson, 2010). This is a simple way to improve prediction for pediatric populations. Moreover, plasma proteins differ drastically in infants (and in pregnant women); since chlorpyrifos is so highly protein bound, this should be taken into account, but may be less important for TCPy or DETP.

A sophisticated PB/PK model for children is also available that allows for flexible inputs (*i.e.*, SimCYP pediatric (SimCYP Company, Sheffield, UK). Although building a pregnancy PK or PBPK model is challenging and ambitious, it was extremely gratifying to see Dr. Hattis' progress on development of a multi-compartment model where the fetal compartment (including the fetus, amniotic sac/fluid and placenta) is separately considered. While it is acknowledged that this will affect outcomes from Dr. Hattis' current oral exposure model (but less so the inhalational) by altering first pass, it's important to recognize that at term, the placenta is perfused to ~600 mL/minute of maternal blood (*i.e.*, the equivalent of the entire mother's blood supply passes through the placenta in about 8 minutes) and has an average surface area of 11 m2. Moreover it

expresses significant CYPs, UGTs and SULTs that have been implicated in chlorpyrifos metabolism (Benirschke *et al.*, 2006). These considerations are relevant to the importance of the feto-placental unit as a separate compartment which is both well perfused and metabolic.

Additionally, while passage from maternal blood, to placenta and fetal blood may be bidirectional, distribution into amniotic fluid is uncertain; it would be helpful, then, to confirm whether or not placental effects might be negligible, retaining the placenta as part of the "liver metabolism." Based on similar scenarios for bisphenol A (BPA), this is not at all certain.

In response to a request from the Agency, it was clarified that the uncertainties in any PBPK model cannot be estimated at this time, since working model parameters [for the Agency's assessment] are not yet defined. In response to a further request from the Agency, Panelists suggest that a more expeditious path to attaining reasonable estimates of fetal exposure would be to generate an equation or algorithm that describes the relationship between maternal serum levels and cord blood levels of chlorpyrifos or its metabolites. Although less certain than the output of a verified PBPK model, this would enable basic dose reconstruction that can then be validated or verified by comparison to parameters in urine and blood reported in epidemiological studies. This may also be a starting point for assessing fetal exposure by defining "flow" and for continuing to build a more sophisticated model. The main limitation in using an equation describing the maternal: fetal ratio (and hence the flow) of chlorpyrifos and/or its metabolites is their short systemic residence time (*i.e.* the blood may only reflect exposures up to a few days prior to blood sampling). As a result, this method will not necessarily reflect cumulative exposure or acute exposures in earlier prenatal periods.

It was also noted that reported relationships between chlorpyrifos in maternal and cord blood warrant reconsideration. In particular, the ratio of 1.05 between the mean values 3.9 pf chlorpyrifos / g maternal blood to 3.7 pf chlorpyrifos / g cord blood in Table 2 of Whyatt *et al.* 2005 differs widely from the ratio of 1.49 between the mean value of 5.96 pg chlorpyrifos/g maternal blood derived from the regression equation Cord = 1.03 Maternal 0.76 given in Whyatt *et al.* 2004 and 4.0 pg chlorpyrifos/g cord blood in Table 1 of that publication. Only the former ratio was referred to in discussions of current PBPK models. Independent of whether or not these ratios represent the same populations, the broad range of this relationship needs to be defined.

Question 5.4

Characterization of chlorpyrifos exposure experienced by women in the Columbia cohort, particularly during the pre-cancellation period, remains an important uncertainty in using these data in quantitative risk assessment. Exposure levels in the range measured in the cord blood data from the epidemiology studies (pg/g plasma) are probably low enough that is unlikely that the cohort mothers were experiencing AChE inhibition at the time of delivery; however, the biomonitoring data were taken after birth and not necessarily associated in time with an application of chlorpyrifos. As such, the actual level of such exposure particularly during any critical window(s) of susceptibility is not known, and a

better understanding of the range of possible exposures and the degree to which they may or may not have elicited inhibition of AChE, remains a key scientific question. *In light of Panel discussions of Questions 4.3 and 5.3, please suggest approaches and/or analyses which would inform the understanding of the degree to which exposure levels experienced by the Columbia cohort participants may or may not have been below doses which result in 10% inhibition of AChE in the most sensitive lifestage. Please discuss the strengths and uncertainties associated with such analyses. Please include in your discussions approaches involving chlorpyrifos and its metabolites and also chlorpyrifos plus other AChE-inhibiting pesticides (propoxur, diazinon) which the cohort participants were exposed too.*

Response

It is important to realize that the short half-life of chlorpyrifos and its metabolites in the body calls into question any" spot data" that might be used. Large cross-sectional investigations may "catch" some exposure, but do not put them in context. Only longitudinal investigations, with frequent sampling are likely to give results that are of real use.

What is called for in estimating the peak dose is prediction of the dose-response curve that would correspond to the vulnerable populations that were exposed. Understanding the limitations of the data available, a PBPK model having the potential to estimate dose given a fixed time since exposure, may provide some information. Additional information that would still be required for a reasonable estimation of maximum dose includes, whether the exposure/dose was steady state or bolus and approximately how long after the bolus exposure was the sample collected. With a very simple one compartment model and a time after exposure a reasonable estimate of the maximum dose can be calculated as well as whether the AChE inhibition threshold was reached. A more sophisticated PBPK model may provide even better data assuming that the PBPK model is applicable to the population being studied, specifically to pregnant women and small children.

Previous Panels have noted the decided lack of a realistic PBPK model for chlorpyrifos for all populations. An effective PBPK model that is applicable to target groups such as pregnant women and infants/small children should be used for these vulnerable populations. An effective commercial version has already been identified for infants/small children (SimCYP pediatric from the SimCYP Company, Sheffield, UK) and should be used for a more comprehensive risk assessment model. Children are potential targets for any developmental issues related to exposure and while there are effective PBPK models for children, they have yet to be discussed here. Utilizing PBPK models designed for the individual and unique demographic (e.g. children and pregnant women) means more than the adjustment of body mass within the model designed for adult males.

In assessing both exposure and dose, a significant data gap exists for the population as a whole but especially for pregnant women which should be addressed with a longitudinal study. A single dose PK study like Clement (1984) provides the foundation for like

populations (adult males) but does not address steady state (or the approximation that is our real world exposures) or populations with different metabolic conditions such as pregnant women or children. As discussed in Section 5.2, progress on this front has already been made as a compartment model with the fetus as a compartment currently exists.

A longitudinal study throughout the pregnancy rather than a few samples in the last trimester would fill many of the data gaps that currently exist for this group. The potential for neurodevelopmental affects on the fetus as well as the metabolic differences in pregnant women versus the workers from the 1984 study, necessitate such a study. Placental tissue might provide more information on the metabolism and the delivered dose to the fetus as the concentration of chlorpyrifos going into the fetus cannot be measured directly from the cord blood or from the difference between cord blood and maternal blood. The tissue concentration may provide information on the chlorpyrifos stores. This information will be vital in creating an effective PBPK model for pregnant women. For any PBPK model used in a comprehensive risk assessment, validation would add confidence to the predictions derived from its use.

Many of the studies discussed in Q 5.1 provide data on the concentration of chlorpyrifos in the media such as house dust, air and water while market basket data exists on the concentration of chlorpyrifos on food. These are the primary tools for generating an effective exposure assessment and a subsequent reconstruction of potential dose. Dose reconstruction can be used to evaluate the efficacy of the PBPK model since its prediction of excretion rates can potentially be validated with an accurate estimate of dose. This assessment of the PBPK model through reconstructed dose may bridge some of the data gaps in assessing risk by validating the PBPK model. A validated model allows for effective prediction of exposure at the critical windows of maximum effect (AChE suppression) with measured urine concentrations. More data exists on chlorpyrifos than other pesticides in the environment, and this may be the best opportunity for utilizing exposure data to evaluate a PBPK model. It is noted however that is there is a significant difference between the predicted urine or blood concentrations that both the PBPK modelers and those that produce the exposure estimate, will point to the other for using “bad” assumptions. In this case both models should be reevaluated for the assumptions used.

The effects mixtures of chlorpyrifos + Diazinon /chlorpyrifos + Propoxur or chlorpyrifos/Propoxur/Diazinon have not previously been considered. Like from all mixtures both constructive and destructive interference can occur. Questions will have to be addressed; do they affect each other’s half lives and distributions and clearance through metabolic competition (Coughli et. al. 2012)? Are their net AChE effects additive or multiplicative? Do they share mechanistic pathways? To address these questions, the Panel recommends further studies described in 5.3 to improve estimates of effects when mixtures of xenobiotics are used compared to single agents. In particular PK parameters such as distribution, half-life and clearance/elimination can be altered if admixtures of chemicals interfere with the absorption or metabolism of another component of the admixture. Using currently available data, other than improving in

silico PK or PBPK approaches (again described above in 5.3), the Panel is not sure there is more that the Agency can do to reanalyze or transform the available data into more meaningful studies. However, any estimation of effect should have an additive dose effect as a minimum and perhaps greater protective factors until mechanistic studies can be done.

Other considerations

A further criticism is in the focus on 10% AChE activity reduction. While certainly a benchmark, the fact that no mechanism has been proposed that would tie such a reduction to any specific outcome begs the question; what is the role of the 10% reduction of AChE in predicting negative health outcomes. The Panel noted that to their knowledge there is no proposed mechanism whereby a 10% reduction in AChE activity in a pregnant woman, even at a specific point in pregnancy, is responsible for cognitive deficit or neurodevelopmental delay of the fetus? The current proposed mechanisms focus on correlation; the deficit in AChE in the mother is assumed to be associated with some other activity, e.g., transport of parent chlorpyrifos (or TCPy for that matter) across the placenta and the nascent blood-brain barrier in the developing fetus? Since no one knows whether this occurs, the utility of the measurement of maternal AChE reduction is unknown. AS is often the case, "more research is needed."

Some on the Panel feel that the 10% figure is merely a marker of some level of exposure. This level may differ in its impact depending on the association of the AChE inhibition with the parent pesticide concentration in the serum. If the Panel assumes that each OP produces exactly the same level of AChE inhibition on a, say, molar basis, does that imply that there is an identical effect of each? Focusing again on chlorpyrifos, is the parent, the oxon, or some other metabolite that is responsible for some of the effects seen in the Columbia study? Only with a better understanding of exposure to chlorpyrifos at various gestational ages will the Agency be able to determine what exposures are causing the effects. The mouse studies do not seem to help all that much.

Question 6: Characterizing the range of potential risks.

The 2009 NRC report, *Science and Decisions*, focused on improving the *technical analysis* through the development and use of scientific knowledge and information to promote more accurate characterizations of risk, and thus improving the *utility* of risk assessment for risk-management decisions. The NRC report also pointed out that regulatory risk assessment does not routinely approach public health and environmental problems by arraying a wide range of options for dealing with them. *In the case of chlorpyrifos, in light of the discussions of Questions 1-5, please provide guidance for assessing and presenting the range of plausible responses at given doses, and the effect of the overall uncertainty and variability around that range.*

Response
Part of the value of the framework for integrative analysis to evaluate multiple lines of evidence in the context of the AOP/MOA is to enable us to draw inference on the weight of evidence from the totality of the data.

In characterizing the range of plausible responses at given doses, it seems important to draw maximally on the dose-response data, beyond a single or several points of departure. For example, for risk management purposes, characterization to the extent possible, of the nature of potential risks above the reference dose would be informative. Presentation, then, of an array of points of departure for various endpoints for different types of effects bounded by their relative uncertainty, would more meaningfully characterize that value selected for the Reference Dose in the context of the range of effects reported in the broader database. It should also promote reliance on more certain rather than the most conservative data. As a minimum, it would be helpful in communicating the relative degree of protection provided by the selected point of departure.

As indicated, in response to previous questions, the maximal use of the available dose-response data from the epidemiological studies is recommended as a basis to at least, "bound" reference doses developed on the basis of points of departure from animal data. To the extent possible, this step should take into account at least the semi-quantitative bounding of the dose-response relationship from human studies based on the impact of identified uncertainties. This would perhaps clarify the basis for (the seeming) conclusion that the uncertainties associated with the exposure–response relationship in the epidemiological studies are greater than those associated with the POD derived on the basis of the animal data (*i.e.*, the basis for relying on the latter for dose-response analysis).

Similarly, options for dose-response analysis for acute effects should be considered separately from those based on long term exposures – *i.e.*, measures representing acute adverse neurological outcomes (ChE inhibition) commonly associated with occupational exposure versus those potentially related to long term exposure in the general population, such as neurobehavioral disorders. This separation would underscore the significant variation in the range of exposures in the population associated with these different types of effects, as reflected in reported TCPy levels measured in the three birth cohort studies and the recent occupational studies. Reconciliation of variability and uncertainty for these different options will likely require additional, focused study (see response to Question 4.3b).

It is further suggested that the Agency focus on the data of chlorpyrifos levels in the cord blood samples as the base to develop the POD for chronic exposures to chlorpyrifos based on a PBPK/PD model. This preliminary work would not only identify priorities for the acquisition of additional data, but would also reduce overall uncertainty and variability.

It seems important, also, to consider comparability within the Agency across compounds for which epidemiological data on neurotoxicity have served as the basis of points of departure – *e.g.*, mercury and lead. How does the weight of evidence from epidemiological studies for these compounds compare with that for chlorpyrifos? For example, in the background paper, it is stated that: "There are a number of known developmentally neurotoxic chemicals with well established relationships between exposure and neurological disorders in humans for which a definitive mode of action has not been established: for example, lead, methyl mercury and ethanol." While documentation of an MOA is, then, not a prerequisite for basing points of departure on human epidemiological data, the nature of the weight of evidence that distinguishes chlorpyrifos from these cases, as a basis for reliance on animal rather than epidemiological data to characterize the point of departure, is unclear.

In relation to the databases of studies which underlie considerations related to weight of evidence including consistency, specificity and biological plausibility, it would also be extremely helpful to have *a priori* criteria (to be presented initially) as the basis for evaluation of the individual studies on, for example, neurodevelopmental effects in animals and humans. While it is recognized that these criteria cannot be prescriptive, an upfront discussion of the factors taken into account in judging the adequacy of individual studies and hence, the weighting of their contribution within the weight of evidence, would be valuable.

The most susceptible lifestage(s), populations that would be expected to be more vulnerable to the effects of the chemical, and the effects of background exposures on these risks need also to be addressed in risk characterization.

Background exposures would include:

- exposures to other sources of chlorpyrifos
- exposures to other chemicals that affect
    - key steps thought to be involved in chlorpyrifos' neurodevelopmental adverse outcome pathway, *e.g.*, exposures to other chemicals competing with chlorpyrifos for metabolism by the same enzyme system (see response to Charge Question 5.4).
    - the same apical neurodevelopmental endpoints, *e.g.*, decrements in working memory or full scale IQ.

In assessing the range of plausible responses at a given dose of chlorpyrifos, variability in response within the population of concern should be taken into account, and uncertainties in estimation of the response should be addressed.

Sources of variability in response to chlorpyrifos include:

- differences in biological susceptibility, such as differences in lifestage, health and disease status, and genetics (*i.e.*, polymorphisms in Phase I and Phase II metabolism)
- differences in background exposures

Significant uncertainty in the draft chlorpyrifos risk assessment, arises from multiple sources. These include uncertainties:

- estimation of chlorpyrifos exposures in the children's cohort studies,
- whether protecting against AChE inhibition is protective against neurodevelopmental effects,
- whether the dose-response data for AChE inhibition in the most susceptible animal model, pregnant rats, can be used to derive a dose at which AChE inhibition would not occur in humans exposed prenatally,
- fully characterizing the neurodevelopmental effects of chlorpyrifos,
- translation of in vitro concentration–response relationships for neurotoxicity to *in vivo* dose-response relationships,
- dose-response for neurodevelopmental effects,
- identifying the neurodevelopmental adverse outcome pathway(s) of chlorpyrifos,

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Appendixes

Appendix A to Question 3.1- Evaluation of Individual Studies

The strengths and weaknesses of the studies are outlined as are the principal findings and study designs that are also summarized in a different way in the EPA Draft Issue Paper in Appendix 3. In general, the studies fall into two major groups: (a) those that have adequate group sizes controlled for litter effects by sampling either only 1 offspring/sex/litter or over-sampled only slightly by testing 2 offspring/sex/litter and analyzed the data using appropriate statistical models, *e.g.*, ANOVA with factors of group and sex, optimally with litter as a block factor in a randomized block design in which treatment and sex are fixed effect factors within blocks and with within subject factor(s) for trial, day, or interval where the same subjects is assessed on the same parameter repeatedly; and (b) those that failed to control for litter effects, tested too few litters and/or offspring, were under-powered and prone to Type I error, and therefore should be given less weight.

Some studies included appropriate down-stream statistical analyses and some did not. In general, most studies used factorial ANOVA or MANOVA models, but the follow-up methods used varied significantly. Most ANOVA models should be further analyzed by some method that deconstructs interactions and all experiments should include a-posteriori group comparison methods than control for multiple comparisons while holding alpha-constant. There was much use of the Fisher Least Significant Difference (LSD) test for post hoc group comparisons among the above reviewed studies, which is only appropriate when there are not more than three groups. In a number of studies there were only three groups therefore this method is acceptable in those cases. However, there are a number of studies where there were more than three groups and the LSD or Protected Least Significant Difference (PLSD) tests were used despite their drawbacks. The use of Tukey tests in the absence of significant F-tests was also reported in several studies. This approach requires clearer justification and reporting of which results used Tukey T-Tests irrespective of a significant F-test and which were used after a significant F-test as a follow-up in experiments where both approaches are used.

The Radial-arm maze (RAM) has been used extensively in the experiments reviewed. A significant concern is that none of those reviewed above controlled for the response pattern known as chaining, *i.e.*, where the animal learns a strategy such as entering each adjacent arm successively or approximately successively by, for example, learning to always turn right or always turn left. When this occurs, working memory is not assessed. What is assessed is more likely habit formation, which is learning, but more rudimentary than working memory which is closely tied to higher cognitive functions such as attention and executive functions and hence assesses higher order processes. It is difficult, therefore, in all the RAM data in this group of reviewed experiments to determine what the chlorpyrifos-related effects were measuring. There is no doubt that chlorpyrifos has effects on RAM performance, but they may not be working memory effects unless it can be shown that more rudimentary forms of learning were not utilized. As for the reference memory effects, these are more likely to be as they appear but even

here one cannot be sure without better test procedures that have an inter-trial interval delay before each new trial to ensure that animals are not entering non-baited arms because the cost of doing so does not outweigh the overall retrieval of the rewards regardless of the small cost of running down an empty arm. One experiment used spatial delayed alternation in a T-maze (Maurissen *et al.*, 2000). This experiment appropriately imposed a delay between the sample and test trials, which is the appropriate way to test working memory. This paper also showed an appropriate short-term memory decay function that was dependent on the length of the delay interval. This is a valuable internal control to prove that working memory was assessed. Some delay between arm choices in the RAM is similarly needed, even if a full decay function is not demonstrated. This would ensure that trial-dependent memory is being assessed rather than some other strategy. Not one experiment that used the RAM in this group of experiments imposed this basic requirement rendering interpretations difficult at best.

There were many other test method issues among these studies that raise further concerns, including that methods were used that have no known neurotoxicological significance. The functional significance of increases or decreases in time in open arms of the elevated-plus maze is unclear as a toxicological end point. Which change represents an adverse outcome: an increase or a decrease? Or is any change from control regarded as an adverse effect? What is the meaning of greater or lesser social investigation of a stranger mouse in neurotoxicological terms? What does it mean that a female mouse has a more upright posture when presented with a male intruder? Is it more adverse that she stands more or stands less in such a defensive posture? With no validation as to the neurotoxicological significance of anxiety tests, depression tests, or social interactions, such outcomes should be regarded as exploratory, and hypothesis-generating, rather than evidence of toxicity.

The bidirectional neurodevelopmental changes that were found in many of the studies presents challenges to interpretation. These effects included a slower habituation trend in female animals in one study, but not another study with the same doses also administered during a prenatal period, and a few transient effects in some of the cognitive tests that were also observed to occur in opposite directions. The lack of specificity of the direction of the neurobehavioral findings is problematic. In some cases isolated markers of certain behaviors were determined to be statistically significant, but these findings were sometimes not supported by other studies reporting no effects (or effects in the other direction) in similar dose ranges using similar routes of exposure. It is difficult to reconcile effects that are bidirectional in given domains of neurodevelopment, or inconsistent across doses and sex, and in the absence of specific hypotheses. In addition, the lack of dose-response among test outcomes (especially those that took doses below 1 mg/kg/d such as Braquenier, *et al.*, 2010, that only found positive responses in the middle dose group) is not reassuring.

Prenatal Studies

This study (Abou-Donia *et al.*, 2006) was a prenatal study of chlorpyrifos. They used timed-pregnant Sprague-Dawley rats from Charles River (evidence of conception = E1).

The authors' indicate that dams were treated transdermally on E4-20 with chlorpyrifos in a 70% ethanol vehicle or were given ethanol vehicle alone. Dams were assigned to groups as follows: 5 mg/kg of chlorpyrifos, 5 mg/kg of nicotine, 5 mg/kg of chlorpyrifos + 5 mg/kg of nicotine, or saline. It is stated that 2 M/2 F per litter were sampled for testing. No statement of whether litter was included in the statistical model was provided; therefore, presumably it was not; hence there was no control for litter effects. Pairwise comparisons were performed using Fisher's LSD test, but they had 4 groups and this test is not appropriate for >3 groups. No litter culling to standardize litter size was done; hence, postnatal rates of growth between litters was not equalized. On P90, rats were tested for Beam walking, inclined plane, and forelimb hang time. Results: They report no CFP effects on beam walking; a female-only effect on the inclined plane (females slipped at lower angles than controls as the plane was tilted). Hang time: Both sexes in the chlorpyrifos group had shorter hang times than controls. Strengths: Used a transdermal route of exposure; they exposed animals from shortly prior to implantation to near-term. Weaknesses: Only one dose of chlorpyrifos was used (estimated to be equivalent to ~1 mg/kg/day). Used 2-way ANOVA but in the results they give no F-values; they provide no indication if reported effects were main effects or interactions; and they moved from ANOVA to LSD tests with no sorting of interactions (although which reported effects were from interactions and which main effects is unclear). They slightly oversampled per litter, but the most significant weaknesses are the small group sizes that results in an under-powered design and they did not analyze the data by litter. Additionally, they used timed-pregnant dams from the supplier for use in a prenatal study for which treatment started very early on E4. To do this, they would have had to have purchased rats that were plug-positive (without confirmation) and received them within 1-2 days of mating, leaving no more than 1-2 days to acclimate to their vivarium before treatment began.

This study (Icenogle *et al.*, 2004) used timed-pregnant SD Charles River dams for a prenatal study, again raising concerns about shipping stress. They dated pregnancy as counting evidence of conception = E1. They treated on E9-12 (the rationale for these days was not given, but seems to be a very narrow period of exposure). They randomized among dams on P1 and then again "every several days" thereafter, introducing unknown stressors in the experiment for both the dams and the offspring. They culled to 10 pups per litter then selected no more than 1 M and 1 F per (artificial) litter and assigned 10 litters to each treatment group. Groups: 0, 1, 5 mg/kg/day chlorpyrifos administered s.c in DMSO on E9-12. They tested offspring for T-maze spontaneous alternation. The T-maze was elevated with 1.5 cm curbs to prevent falling off the edge, and animals were given 5 trials/day with 30 seconds post-choice confinement. The test was given for 5 successive days. The apparatus is non-standard in the field and is designed more akin to the elevated plus maze (EPM), which is a test designed to induce anxiety so that it can be measured. The T-maze for spontaneous alternation is not intended to induce anxiety and for that reason is normally an enclosed maze. The elevated T-maze used in this study probably tests alternation AND anxiety, but one cannot determine how the resulting measurements can be attributed to memory versus anxiety. They also tested rats in a figure-eight locomotor activity monitor for 1 h with data recorded in 5 min intervals and repeated the test three times each spaced one week apart and in a 16-arm radial-arm maze

(RAM) with 12 arms baited daily, 4 arms never baited; the test sessions were 10 min or until 12 baited arms had been entered; they tested twice per week for 18 sessions; then gave a scopolamine (muscarinic antagonist) challenge at doses of 0.04, 0.08, 0.16 mg/kg or to separate animals a drug challenge of mecamylamine (nicotinic antagonist) at doses of 1.25, 2.5, 5.0 mg/kg. They also tested acoustic startle response with prepulse inhibition (ASR/PPI). They first conducted ASR-only trials and later intermixed ASR and PPI trials. Lastly, they tested animals in the elevated plus maze (EPM). For this they used a standard method (standard size apparatus for the typical 5 min. test). Note: this test was given AFTER all preceding tests whereas in most labs it is given before other tests based on the fact that this test is sensitive to prior experience. The statistical approach was MANOVA; interactions were further analyzed and pairwise comparisons were made by Fisher's PLSD. Findings: Spontaneous alternation: chlorpyrifos decreased shortened choice latency on early trials; no effects on alternation but failed to show whether they got alternation rates typical of this test to establish validity in their laboratory. Figure-eight test: They obtained a significant treatment x interval interaction. The chlorpyrifos 5 mg/kg showed faster habituation on two out of four of the last 5-min intervals, with one interval with higher activity in the chlorpyrifos 1.25 mg/kg group; they also note that the linear trend in this analysis was significant for treatment group, but it is noteworthy that the effects observed on this test were very small even if significant. RAM findings: The data were blocked into three sessions per block for analysis; hence, there were six blocks for the repeated measure factor. They found increased reference memory errors in block 1 and increased working memory errors in blocks 1 and 3 in the CFP 5 mg/kg group only with males and females combined. They found no effects on RAM performance after mecamylamine challenge. They found an effect of scopolamine challenge that was complex: Scopolamine increased errors with increasing scopolamine dose in controls but in the chlorpyrifos 1 mg/kg group it increased baited arm errors at lower doses more than in controls but less than in controls at the highest scopolamine dose. For the chlorpyrifos 5 mg/kg group scopolamine increased errors more than in controls after saline but less than in controls after scopolamine. They report no ASR/PPI effects and no EPM effects for time in the open (the principal index of anxiety in this test); the CFP 5 mg/kg group crossed center more than controls, however, suggesting a slight increase in activity. Strengths: Groups sizes were adequate and the data were analyzed by (artificial) litter. The factorial MANOVA models were appropriate. They tested two doses of chlorpyrifos and used many standard methods. Weaknesses: Used timed-prenatal females for a prenatal study thereby introducing prenatal shipping stress. RAM: they ran trials continuously with no intertrial interval (ITI) delay to ensure that working memory was being assessed. While there is no doubt they obtained RAM effects, it cannot be distinguished as to whether these were working memory or habit learning effects. The small reference memory effect they obtained is more likely to be a real reference memory change, but this cannot be certain without observation of the animals' performance or a method to ensure an ITI delay. In this test, a control for chaining (moving sequentially around the maze from one arm to the next or every other arm) or for random selection is important. Without a confinement period in the center between each arm choice, there is no way to rule out that rats obtained the food without relying upon working memory instead of another strategy. For reference memory findings, what one needs to determine is whether rats found the cost of entering even

empty arms insufficiently aversive so that is was more effective to check them all versus remembering which ones were unbaited. There is a basis for this concern in the data. Reference memory errors among controls improved from about 6.5 to about 5 across the 18 sessions. Given that there are 4 unbaited arms, the data suggest that even controls never actually acquired memory for the unbaited arms. If they had, one would expect well under 4 reference memory errors per trial block, assuming they blocked the data by averages rather than by sums; but the paper is moot on how the blocked data were formed. For these reasons, center confinement between trials is an important control especially for the working memory assessments. The authors would have greatly strengthened their experiment by either using center confinement between trials or having an observer map the problem solving strategy during testing to see that the animals were using alternate strategies. The chlorpyrifos vehicle used was DMSO, which was raised as a significant concern in response to another charge question.

In this experiment (Billauer-Haimovitch *et al.*, 2009), heterozygous HB/Igb mice were used and bred in-house (conception = E1). Mice were treated on E9-18 with chlorpyrifos at doses of 1, 3, 5, 10, 20 mg/kg given s.c in DMSO. Half the litters were fostered, half were not; they report no differences in outcome in preliminary analyses and state that the data were therefore pooled for subsequent analyses. They sampled 1 M/1 F for testing per litter. At P75, mice were tested in the Morris water maze (MWM). The pool was 87 cm diameter, the platform 8x10 which is a search ratio of 74:1 (Note: this is less than optimal for a test of spatial navigation in mice). They ran 2 blocks of 4 trials per day for 4 days. After completing hidden platform trials, they ran cued trials with the platform made visible above the water line. Data were analyzed by MANOVA, log transformed, with Tukey *a-posteriori* tests. They report no differential effects between males and females, so they combined sexes for presentation (but in the statistical analyses). They report that in the chlorpyrifos20 group all animals died. They found a chlorpyrifos main effect on MWM latency, which they report was significant for the chlorpyrifos 1 and chlorpyrifos 3 mg/kg groups but not for the chlorpyrifos 5 or 10 mg/kg groups. In a second experiment they used only the chlorpyrifos 3 mg/kg group and found somewhat larger MWM latency effects and these were reversed by nicotine treatment prior to each daily test session. In a third experiment with chlorpyrifos 3 mg/kg, they again found MWM latency effects and these were reversed by cell implantation from neonatal cells grown in neurospheres. Found no speed differences in MWM on trials where latency was significantly increased. They found no chlorpyrifos effect on developmental reflexes (surface righting, startle emergence, age of fur appearance, day of pinna unfolding, or day of eye opening). Strengths: They used the MWM which is one of the most well-validated spatial learning/reference memory tests in neuroscience; they used 14-34 litters per group. A major strength was that they replicated the MWM effect in three separate experiments. Weakness: It is unclear why effects were seen in the MWM at chlorpyrifos 1 and 3 mg/kg but not at 5 and 10 mg/kg. The vehicle used was DMSO.

In this experiment (Turgeman *et al.*, 2011), heterozygous HB/Igb mice were used and bred in-house; Conception = E1 and mice were treated on E9-18. The exposure was CFP 3 mg/kg given s.c in DMSO. Half the litters were fostered, half not; they reported no differences in outcome between fostered and non-fostered litters; therefore, they pooled

data across this factor for later analyses. They tested 1 M/1 F per litter. At P75, mice were tested in a MWM (with the same 87 cm diameter, platform 8x10, and 74:1 search area as in the study by Billauer-Haimovitch *et al.* (2009) and has raised the same concern as above). Testing was in two blocks of four trials/day for four days. After hidden platform trials they ran cued trials. Data were analyzed by factorial MANOVA, log transformed, with Tukey *a-posteriori* tests. They report no M/F differential effects, so they combined sexes for presentation (but not statistically, they note). They report that chlorpyrifos 3 mg/kg increased MWM latency which was reversed by stem cell transplantation, just as in the previous experiment by Billauer-Haimovitch *et al.* (2009) (the stem cell methods were also identical). They found no effect on swim speed, but this experiment included no cued trials to ensure the absence of proximal cue learning problems. They also ran no probe trial to assess reference memory. Strengths: Found clear MWM effects, used 13-25 litters per group, they tested only 1 sex per litter. Weaknesses: Tested only one dose of chlorpyrifos, used a small maze, and did not conduct a test of reference memory or include cued trials as a control or include a reversal component to verify that the effects were hippocampally-dependent. The vehicle used was DMSO.

This study (Laviola *et al.*, 2006) used heterozygous Reeler KO mice on a C57BL/6 background. The mice were bred in-house by het x het crosses and conception = E0. chlorpyrifos-oxon (chlorpyrifos-O) was tested, not chlorpyrifos. chlorpyrifos-O was given on E14-16 (the rationale for this embryonic period was not given but seems very narrow for a compound whose exposure would be expected to be chronic). The compound was delivered by implanted osmotic minipump at a rate equaling 5 mg/kg/d. Offspring were tested on P3, 7, 11 for ultrasonic vocalizations, wire mesh hang time when the mesh was gradually rotated 180 degrees until animal was hanging upside down, and surface righting. As adults (>P70), they tested locomotor activity (30x30 cm apparatus) for 45 min., then administered 2 mg/kg scopolamine and retested for another 45 min.; 1 week later they re-tested the same animals in another apparatus (40x30 cm) for 10 min then removed them and gave a high dose (10 mg/kg) of amphetamine and retested for 50 min.; movements were video recorded and scored later using the Noldus Observer system to rate specific behaviors. Within litters, they had 3 genotypes (KO, het, WT). Treatment and genotype were regarded as between subject factors and other factors as within-subject factors. They used factorial ANOVA with pairwise comparisons by Tukey tests. On P7, the lowest ultrasonic calls were found in KO mice; they were intermediate in hets, and highest in WT but no such pattern was found in chlorpyrifos-O-treated mice; in chlorpyrifos-treated mice they observed high call frequencies in all genotypes. For angle to fall on the wire screen grasping task, they found no genotype effect in controls but fall angle was higher in Reeler chlorpyrifos-O exposed groups and it decreased from WT to het to KO, but even KOs were better than Control KO mice. They reported a chlorpyrifos-O effect on righting only in KO mice on P 7 and 11 (longer latency) but this effect was seen only on the worst of the three daily test trials; no effect was seen on the best or intermediate test trial given each day. In the 30x30 cm activity chambers, they found control KO mice were hyperactive, an effect which according to the text chlorpyrifos-O exposure 'normalized' but they did not show these data. After they gave scopolamine, all groups showed the expected hyperactivity response, but for all

genotypes a main effect was seen of greater hyperactivity in the chlorpyrifos-O groups but only when the 45 min. test session was subdivided such that the difference occurred only in the first 25 min. of the test session and after subtraction of pre-drug activity data (in an effort to adjust for the innate hyperactivity of the KO mice). In the 30x40 cm test chamber, no differences during the 10 min habituation prior to amphetamine challenge were seen. Post-amphetamine, KO chlorpyrifos-O treated mice showed increased activity that was greater than in KO controls, but for stereotypy, the opposite occurred, *i.e*, chlorpyrifos-O exposed mice were less stereotypic than KO controls; similar but weaker trends were seen in WT and hets. Strengths: This is the only study among the neurobehavioral papers to test the oxon. Weaknesses: N's were given for progeny but not by litter; the number of litters used was vague: They state that 12 breeding pairs were used, so presumably 6 litters were treated with chlorpyrifos-O and 6 with the vehicle, but the Panel was given no information on how the genotypes were distributed among the litters within treatment groups. There does not appear to be any control for litter effects. Sex was not described as a factor in ANOVAs. Only one dose was tested. The study was under-powered given only six litters per group with each litter subdivided by genotype and sex. Given the expected Mendelian ratios, it would be expected that half of the offspring would be hets, leaving 25% as KO and 25% as WT. Given that C57BL mice typically have 6-8 pups per litter, the sample sizes per genotype per sex per litter would be quite small and not likely to be evenly distributed.

In this study (Venerosi *et al.*, 2010), they tested the effects of chlorpyrifos 6 mg/kg given by gavage on E14-17 to CD-1 mice (bred in-house), but the date of inferred conception was not provided. They treated 18 litters with oil and 16 litters with chlorpyrifos. Shortly after birth, they culled to 4M/4F. Testing began on ~ P90. They evaluated the offspring in a Light/Dark (L/D) test for 5 min and also did observer scoring. They tested the mice in the Forced swim test (FST) either with an injection of saline or of the selective serotonin reuptake inhibitor (SSRI) fluvoxamine (30 mg/kg) 30 minutesebefore the test. They scored immobility, struggling, and swimming and testing was during the dark cycle. Females were grown to adulthood and bred, and on P8, a stranger male intruder was put in their cage (pups were removed) and scored for aggression. Data were analyzed by ANOVA or Mann-Whitney using litter as a factor; ANOVAs were followed by Tukey tests but in some cases in the absence of a significant F-test. They do not define the reason for this but cite a reference that it may be used in the absence of a significant F-test. They also state that they used non-parametric tests in some cases including analyzing interactions by Mann-Whitney U tests. It is unclear how it is used to detect interactions. They report finding a significant U-test for females for time in the tunnel connecting the two sides in the L/D test, with chlorpyrifos-exposed females spending more time in the tunnel than controls. For the FST, they report an interaction between chlorpyrifos x fluvoxamine in which chlorpyrifos eliminated the increased swimming induced in controls given fluvoxamine; this pattern was repeated for immobility time, *i.e*, fluvoxamine reduces immobility in controls, but this effect was dampened in chlorpyrifos-treated animals enough that the change in this group was not significant. In the intruder test, they also reported a chlorpyrifos x fluvoxamine interaction. They found that fluvoxamine reduced the duration of attack and increased inactivity in controls, but this pattern was largely eliminated in the chlorpyrifos exposed

group. Strengths: chlorpyrifos was given in oil, not DMSO. They used litter in the analysis and tested 6-13 offspring per group. Weaknesses: They had only one dose of chlorpyrifos; they do not say which outcomes they assessed by direct Tukey tests and which by ANOVA followed by Tukey comparisons. The meaning of the intruder test outside of basic research or as an index of neurotoxicity is not known.

In this study (Haviland *et al.*, 2010), they used 0, 1, 5 mg/kg chlorpyrifos given s.c. in DMSO to Swiss-Webster on E17-20 where conception was termed E0.5 (but after stating this, they never used half days again, so it is unclear of E0.5 was rounded to E0 or E1). Mice were bred in-house and litters were culled to 8 and balanced for sex. Testing began on P60 using a modified 8-arm RAM with a T at the end of each arm with food always in the left arm of the T (they name it the Foraging maze) and they compared animals tested in this maze to a group tested in a standard 8-arm RAM. They tested animals for 3 sessions per week for 3 weeks for a total of 9 sessions. For some reason that they do not explain, in the RAM they baited 6 of the 8 arms on each trial, but for the Foraging maze they baited 4 of the 8 arms on each trial. Strengths: Testing two groups in two mazes that presumably assess the same functions is a strength (potentially). They analyzed the data by litter, but the results are not well described. They state that RAM data in standard terms of errors by type (entries into baited versus unbaited arms), but they did not use an ITI delay to ensure that working memory was being assessed. Data were analyzed by ANOVA, but no mention was given of how pairwise comparisons were done, but it was probably by LSD since they used the LSD for pairwise comparisons for their thyroid assay data. They report finding only reference memory errors in the RAM, but all groups showed poor learning; they do not show working memory data at all and the reference memory differences are scattered across the test sessions and between doses non-systematically. In the foraging maze, they report their findings differently. It is noteworthy that they state that the entrance of each of the baited arms were marked with a 0.5 cm radius peg (or 1 cm in diameter) (Note: this is an extraordinary procedure; the provision of an obvious cue to which arms are baited and which arms is not, defeats the purpose of the test which is to remember which arms are baited and which are not; cuing the rat at the entrance to each arm provides evidence only that the rat can recognize the cue, requiring little memory). They report the data as the proportion of correct choices out of total choices, such that perfect performance results in a score of 1.0. Clear improvement toward 1.0 was seen across test sessions and chlorpyrifos caused a slower increase toward 1.0 at both doses in females but not in males. In males, chlorpyrifos exposure cased a more rapid increase toward 1.0 at 1 mg/kg but not at 5 mg/kg. Strengths: Use of two learning tests and the effort to develop an improved version of the RAM are noteworthy. Weaknesses: The vehicle used was DMSO. Poor learning in the RAM, performance on the Foraging maze not shown in terms of working and reference memory errors, and the use of an in-maze cue to which arms were baited seems to defeat the purpose of the test; findings were not dose-dependent or consistent across sessions. No ITI delay was used. It is not clear what the foraging maze adds to a more standard RAM. Why would a mouse learn better if it goes down a cued arm and turns left rather than going down an unmarked arm to find food when the arm is straight?

This study (Levin *et al.*, 2002) used timed pregnant SD Zivic-Miller (ZM) rats; date of inferred conception was not stated. They tested 0, 1, 5 mg/kg chlorpyrifos given s.c. in DMSO administered on E17-20. Litters were culled to 10 and randomized to dams. The rationale for randomizing the pups is not stated, nor is there any published data this reviewer is aware of that establishes that this cancels out litter effects although it clearly randomizes within litter genetic factors, but whether that improves or confounds outcomes is unknown. Moreover, the degree of stress this induces in the pups or the dams is unknown. Testing was done during the dark cycle. Litters were culled to 10 M/10 F per group per artificial litter. Spontaneous alternation used an elevated T-maze with no walls (see above) and was tested on P28-42, 5 trials per session (with confinement) for 5 daily sessions. They tested rats in a figure-eight locomotor system on P28-42 for 1 h/session, and did this three times each spaced one week apart; also used a 16-arm RAM with testing on P57-91. The testing was for 3 days/week for a total of 18 sessions. Their procedure was to use 4 unbaited arms and 12 baited arms. Each session lasted up to 10 min or until all baits were taken. They continued RAM testing after this on P98-119 with either a scopolamine challenge (muscarinic antagonist) at doses of 0.04, 0.08, 0.16 or mecamylamine (nicotinic antagonist) challenge at doses of 1.25, 2.5, 5.0 mg/kg given prior to each test session. Data were analyzed by ANOVA with factors of treatment and sex as between factors and test interval or day as within-subject factors. Main effects were taken as significant if they occurred at $P<0.05$ but interactions were followed-up if $P<0.10$. Pairwise comparisons were by Fisher's PLSD. They found no effects on spontaneous alternation frequency but report finding that chlorpyrifos decreased latencies to choose one of the arms (Note: latency to choose is not a typical outcome measure in this test). For the figure-eight test, they found no effects on the omnibus ANOVA, but then analyzed the first session and report finding a treatment x sex interaction in females in which they habituated slower than both chlorpyrifos groups. They used a trend analysis in which the linear trend was different in both chlorpyrifos groups compared with controls, but the effect was small and linearity did not appear to be a good fit to the data. In the RAM, they report a significant main effect of treatment and a treatment x error type and a treatment x error type x sex interaction. They followed these up and found female working memory effects at chlorpyrifos 1 mg/kg but not at chlorpyrifos 5 mg/kg. They report the same pattern for reference memory errors but less pronounced. After scopolamine, it was again found that female chlorpyrifos1 animals showed differential effects on RAM on working memory errors (not on reference memory errors or latency). For females, they found the slope of improvement across trials was lower in the chlorpyrifos1 group than for controls or the chlorpyrifos5 group for total errors using a $P<0.07$ trend to justify the trend analysis. When only working memory errors were analyzed, the female chlorpyrifos 1 effect was not seen. No effects were seen in males. For reference memory errors, there was again a significant linear trend for females, but the effect was that the CFP 1 mg/kg females made fewer errors than controls but this was because scopolamine caused controls to make more errors whereas it did not cause this in the chlorpyrifos 1 mg/kg females, which might make sense except that a higher dose of scopolamine did not cause controls to make more errors, making these data largely uninterpretable. Strengths: Sample size was adequate and they used litter by selecting only 1 male and 1 female per (artificial) litter. They used two doses of chlorpyrifos. Weaknesses: They did not use an ITI during RAM testing to

ensure that working memory was being assessed. They use an unorthodox elevated T-maze to assess spontaneous alternation. They used ZM rats. They used timed-pregnant rats for a prenatal study introducing a potential prenatal stressor. They provide no rationale for the short E17-20 exposure. Given that environmental exposure to chlorpyrifos might be chronic this short exposure window does not appear to be a very appropriate choice.

In this experiment (Ricceri *et al.*, 2006), CD-1 mice were gavaged with chlorpyrifos at doses of 0, 3, 6 mg/kg/d in peanut oil on E15-18 (in-house breeding with date of inferred conception = E0). At birth, litters were culled to 4M/4F. Within each litter, one male/female pair was treated postnatally on P11-14 with 0, 1, or 3 mg/kg chlorpyrifos, creating permutations totaling 9 pre/post-natal treatment group combinations. On P70, they tested males for 20 min. in an open-field under red light, video recorded the animals, and later used the Noldus system to categorize behavior. On P75-80, males were tested for 20 min. in a novel cage with a stranger male. On P90, females were given 3 pups for 20 min. to observed induced maternal behavior. On P120, mice were tested for 5 min. in the EPM. Data were analyzed by litter using ANOVA models, but the statistical section is difficult to follow because the run-on sentence says "prenatal treatment as block with respect to postnatal treatment, sex, and repeated measure as within-litter treatment factors, postnatal treatment, and sex as fixed-effect factors within litter, and repeated measure as fixed factor within subjects." From this, it is somewhat difficult to determine whether sex was treated as a between or within factor, but the analysis does indicate that litter was handled as a blocking factor within the ANOVA and it appears that prenatal treatment was a between factor and postnatal treatment a within factor. Variables where the same subject was tested repeatedly were handled as repeated measure factors. Significant F-tests were followed up using Tukey with Bonferroni correction. Results: In the open-field, they found increased activity in the chlorpyrifos6 males (females were not tested). In the test with a stranger mouse, the principal finding was in the postnatal chlorpyrifos3 group that showed increased attack behaviors against the stranger; they also found that the prenatal chlorpyrifos6 group showed a significant increase "upright postures" during this test. In the test of induced maternal behavior, the postnatal CFP 1 & 3 groups showed decreased licking frequency but increased licking duration, along with increased crouching frequency over the pups and for longer intervals but decreased pup sniffing. In the EPM, the significant finding was in the postnatal chlorpyrifos3 group that showed increased time in open arms. Strengths: They controlled for litter effects, and then did factorial ANOVAs; they controlled for multiple comparisons; they used adequate numbers of litters; and they used sound behavioral methods. The complex prenatal x postnatal treatment design could be strength to the extent that it identifies critical periods of exposure but could be a weakness as it makes the experiment logistically complex and difficult to manage; the study included multiple doses, 2 of which were prenatal and 2 of which were postnatal. Weaknesses: The choice of the narrow exposure windows is not well justified and appears arbitrary. Changing the dose levels used for the prenatal and postnatal exposures adds another complication. The fact that significant outcomes appear only in some dose groups treated prenatally and in some treated only postnatally, but not in those treated both pre- and postnatally which cumulatively had greater chlorpyrifos exposure, is difficult to reconcile. The effects were

not generally dose-dependent even within the prenatal, postnatal, or prenatal-postnatal combination groups. More fundamentally, the interpretation of the anxiety and social interaction tests as indices of neurotoxicity is uncertain. While these tests are interesting and identify areas for further investigation, they are difficult to use in risk assessment until they have an established neurotoxicological basis. For example, is it more adverse to show a modest increase in anxiety or a decrease? It is known that in humans anxiety/stress is an inverted U-shaped function. Low stress and anxiety leads to poor performance whereas high stress and anxiety interferes with performance; moderate stress and anxiety produce optimal performance. This is true in all mammalian species. Chronic stress and anxiety follow the pattern; many studies have shown that moderate developmental stress in rodents leads to increased cortical thickness and greater arborization and improves learning. Anxiety tests, such as the EPM, have a well-validated basis in the context of antidepressants. They are valid when used as intended to assess the effects of acute or subchronic exposure to SSRI, tricyclic and atypical antidepressants. They are increasingly used in gene targeting studies where candidate genes suspected of involvement in fear, anxiety, and stress are being assessed, but they have never been validated in neurotoxicology. This is also the case with social interaction tests. These tests are still being developed and their meaning in basic neuroscience research, as for example, in genetic models of autism spectrum disorders (ASD), are not yet established. Trying to interpret such methods in neurotoxicology is premature. Again, one must pose the question: Is it worse or better that the females given pups in this experiment, lick them more or less if they were exposed pre- and/or post-natally to chlorpyrifos? Unfortunately, no one can answer this question based on currently available data.

This experiment (Venerosi *et al.*, 2006) is identical in design to that of (Ricceri *et al.*, 2006). The Ns are the same as are all the major experimental design features. In terms of test outcomes, they conducted a social recognition test in which females were placed in single cages for three days and then introduced to stranger females for three min.; 45 min. later, they were re-exposed to stranger mouse-1 for another 3 min. and 45 min. later exposure to stranger mouse-2 and ultrasonic calls were recorded. Vocalizations in controls went down on retest-same and up on retest-different, whereas vocalization in the prenatal CFP3 group changed slightly, and in the CFP6 group it changed dramatically, causing retest-same to go up and retest-different to go up more than in controls. Postnatal CFP largely reversed the pattern. Social investigation: prenatal CFP increased social investigation, had no effect on retest-same or retest-different (latter not shown in figures). Here again, neither social interaction induced vocalizations nor social investigation of other animals has a known neurotoxicology interpretation. Strengths: This experiment has the same strengths as Ricceri *et al*. (2006). Weaknesses: It has the same weaknesses as Ricceri *et al.* (2006).

Postnatal Studies

This study (Dam *et al.*, 2000) comes from the Slotkin lab and has most of the features of this lab's previous work. As before, Sprague-Dawley Zivic-Miller rats were used. As before, litters were culled after birth to 10 pups and randomized across dams on P1 and

every 3 days thereafter until weaning. chlorpyrifos was administered s.c. in DMSO on P1-4 at doses of 0 or 1, and other animals were treated with chlorpyrifos on P11-14 with 0 or 5 mg/kg/day. Prior to weaning, offspring were tested for surface righting on P3-4 and on the inclined plane (20 degree angle) on P5-8 for those treated on P1-4. Open-field activity was manually tested in a large 100x100 cm field on P21 and P30 for 5 min. each time. Results: They found delayed surface righting and inclined plane rotation times in the chlorpyrifos1 females treated on P1-4, but not in males. In the open-field, they report that males in the CFP1 group had decreased square crossings and rearing frequency with no change in grooming frequency. In the P11-14 groups, they report no change in P21 or P30 in open-field line crossings, but chlorpyrifos males showed increased rearing at P30 and no other changes. Strengths: They used adequate sample sizes and analyzed the data taking litter, albeit artificial litter, into account. Weaknesses: They sampled 2 offspring per sex per litter, so there was slight over-sampling. The artificial litter technique remains unverified as a technique, and it could introduce stress on the pups and the dams being shuffled every three days. The 5-minute open-field test is generally regarded as inadequate by current standards, including those from 2000 when this study was published. The Zivic-Miller rat is less than ideal for behavioral studies. The vehicle used was DMSO. The findings were not dose-dependent.

This study (Levin *et al.*, 2001) was in collaboration with the Slotkin lab and has many of the common experimental design features noted above from this group. They used Sprague-Dawley rats from Zivic-Miller. Litters were culled to 10 pups, randomized and re-randomized every several days. Offspring were treated with CFP on P1-4 with 1 mg/kg, or on P11-14 with 5 mg/kg s.c. as in the previous study by Dam *et al.* (2000) dissolved in DMSO. However, this experiment shows the influence of the Levin lab: Adult offspring were tested for spontaneous alternation in the elevated T-maze referred to previously, including significant concerns about using a non-standard way of conducting this test. Locomotor activity was tested in the figure-eight system and RAM testing was as reviewed above with pharmacological challenges given after initial learning. All testing was done during the dark cycle. Spontaneous Alternation was conducted on P28-42, 5 trials with confinement after an arm choice for five sessions; figure-eight testing was done on P28-42, 1 h per session, 3 sessions spaced one week apart. The RAM was the 16-arm system tested on P57-91 with testing conducted three 3 days per week for a total of 18 sessions (4 arms unbaited; 12 arms baited; 10 min per session). RAM with drug challenge was conducted on P98-119 with scopolamine at doses of 0.04, 0.08, 0.16 or mecamylamine at doses of 1.25, 2.5, 5.0 mg/kg. ANOVA models were treatment and sex as between factors, and interval or day as within factors at $P<0.05$ except interactions which were taken as significant at $P<0.10$; the method of doing pairwise comparisons was not indicated in this paper. Results: They reported no effect on spontaneous alternation frequency and a small effect on latency in chlorpyrifos-exposed males on this test. In the figure-eight test, no effects were seen in the chlorpyrifos P1-4 exposed group, but reduced habituation slope was noted in the P11-14 CFP group. In the RAM test, the P1-4 CFP group showed effects on working and reference memory errors in males in the first block of trials but not thereafter and in females across blocks; no effect of chlorpyrifos exposure was seen after P11-14 exposure on learning the task. Treatment with 0.16 mg/kg of scopolamine increased reference memory errors in P11-14

chlorpyrifos-exposed males, with a larger effect in females that occurred at the lower scopolamine doses but not at the highest dose (0.16 mg/kg) of. Strengths: They used adequate sample sizes, litter was accounted for in the analyses, there was no litter over-sampling, and the behavioral methods were mostly sound. Weaknesses: Not a dose-response study; the vehicle for chlorpyrifos was DMSO; the use of pup randomization; issues concerning interpretation of the RAM data given the absence of an ITI delay interval; and the use of the elevated T-maze for spontaneous alternation.

This study (Aldridge *et al.*, 2005) also comes from the Slotkin lab. This time they used Sprague-Dawley CD rats from Charles River. They obtained timed pregnant rats and culled litters shortly after birth to 10, randomized pups among dams and re-randomized them every several days. Pups were treated with 1 mg/kg chlorpyrifos on P1-4 by s.c. injection in DMSO. No more than 1M/1F per artificial litter were sampled for a total 9M/9F per treatment (hence 36 rats were used altogether). Rats were tested during the dark cycle starting. Tested consisted of the EPM on P52-53, a two-bottle sweetness preference test on P54, and starting on P64 RAM learning with ketanserin challenges on weeks 16-17 with the drug given 20 min prior to testing at doses of 0, 0.5, 1.0 or 1.5 mg/kg (5HT2 antagonist). RAM was tested for 18 sessions (as above). Results: In the EPM, they report that CFP-exposed males had increased time in open arms (indicative on reduced anxiety) with no effect in females. In the sweetness preference test, CFP exposure reduced preference for the sweet choice in both sexes from about 4:1 to about 3:1. In the RAM, chlorpyrifos caused treatment x sex interactions on both working and reference memory. For working memory, chlorpyrifos exposure increased working memory errors in males and decreased these errors in females; similarly reference memory errors were increased in chlorpyrifos-exposed males and were decreased in females. While several of these changes from the interaction were individually short of being statistically significant, when errors types were combined, the male and female changes in errors were significant. Ketanserin had no effect on errors of either type in controls, but increased both error types in chlorpyrifos exposed rats at all doses for working memory and at the high and low doses for reference memory. Strengths: Adequate sample sizes, analyses that took litter into account, well-conducted behavioral methods. The use of ketanserin to show an effect of a 5-HT2 antagonist tested at multiple dose levels of the challenge drug was a major strength of this study and is a finding worthy of future investigation. Weaknesses: They used only one dose of chlorpyrifos, they used DMSO as the vehicle, and they did not use a delayed ITI during RAM testing making it difficult to determine if working memory or habit learning was actually what was affected.

In this study (Ricceri *et al.*, 2003) CD-1 mice (bred in-house, conception = E0), were culled to 5/5 M/F per litter and 30 litters were used. chlorpyrifos was administered s.c. in DMSO at doses of 0, 1 or 3 mg/kg on P1-4 or P11-14. On P1, 5 and 11 ultrasonic vocalizations were recorded; on P10 homing behavior to home cage scent was tested; on P25 locomotor activity was tested; on P35 mice were tested in a box divided into white and black compartments; social interaction with stranger mice was tested at P45; and at P60, passive avoidance (males only) were tested for up to 10 trials to remain in light side for 2 min. with a 24 h retention test. They sampled only 1 mouse per sex per litter and

analyses took litter into account. Results: No effect of chlorpyrifos on ultrasonic vocalizations or pup homing to home cage scent were obtained, or on locomotor activity but the authors noted a p<.06 trend in the P11-14 chlorpyrifos3 mg/kg group to be more active. In the white/black box, effects of P1-4 chlorpyrifos exposure were significant on 1 out of 5 test intervals (interval-2), whereas in the P11-4 chlorpyrifos exposed animals activity changes were significant on 1 out of 5 test intervals (internval-4). Several borderline effects and several significant interactions on different social interaction measures were found in the CFP groups but a clear pattern was not evident. There were no significant CFP-related effects on passive avoidance acquisition or retention. Strengths: Sample sizes were adequate and litter was taken into account. Behavioral methods were appropriately conducted. Weaknesses: Dissolved chlorpyrifos in DMSO, effects were not dose-dependent nor exposure period-dependent. Most of the effects were small.

In this study (Venerosi *et al.*, 2008) CD-1 mice (bred in-house, conception = E0) were treated S.C. with CFP at 0 or 3 mg/kg dissolved in peanut oil on P11-14. Litters were culled 4M/4F. Mice were evaluated in a social interaction test at P40-45. Female offspring bred and after delivery tested for nest building on P1-7 and other maternal behavior on P1 and later tested in the light/dark test of anxiety. They also did put retrieval test and a test for maternal aggression. These authors did a power calculation and sample size determination; they controlled for litter effects and used mixed model ANOVAs for most data analyses, but for some data they used non-parametric methods. They had 15 litters using a split-litter design. Results: they found no effects on ultrasonic vocalizations or social investigation. They found females exposed to chlorpyrifos did not build nests as well or defend their territory as much against a stranger male mouse, and took less time to emerge from the dark side of the light/dark box, but no other measure on this test was affected. Strengths: This was one of the most rigorous experiments in terms of sample size, control for litter effects and statistical methods for analyzing the data. Weakness: The relevance of the tests as indices of neurotoxicity are entirely speculative as none have been validated in this context, or in any context as strong evidence of developmental abnormality no matter what the independent variable.

The study by (Johnson *et al.*, 2009) used Sprague-Dawley CD IGS rats (Charles River) bred in-house. Twenty litters were used in a split litter design. Litter size was adjusted to 12-14 balanced for sex, with 7 groups per litter to the extent possible. Exposure was on day P1-5, 6-13, 14-20 as follows: Controls received oil from P1-20, and the low dose group received chlorpyrifos at 1.0 mg/kg from P1-2, but the mid and high doses groups received escalating doses: the mid dose received chlorpyrifos 1.0, 2.0, and 4.0 mg/kg during each of the aforementioned exposure ages, and the high dose received doses of 1.5, 3.0, and 6.0 mg/kg, respectively. The remaining three groups were exposed to methyl parathion at doses of 0.2 mg/kg throughout, or escalating doses of 0.2, 0.4, and 0.6 mg/kg/day (mid dose) or 0.3, 0.6, or 0.9 mg/kg/day (high dose) in oil by gavage. Results: The authors' report no effects of physical landmarks of development (pinna unfolding, fur appearance, day of eye opening, or day that incisors erupted) and no effects on early reflexes (surface righting, air righting, startle emergence, cliff avoidance, or inclined plane). Ad adults, they tested rats in a 12-arm RAM (8 baited, 4 unbaited

arms). They found no significant working memory effects in females but a significant increase in working errors in the high dose chlorpyrifos males across sessions and at lower doses in final week only. For reference memory, the female mid and high dose chlorpyrifos groups made fewer errors, whereas for males in the mid and high dose groups made significantly more errors. Strengths: Used a split-litter design and controlled for litter effects, had adequate sample sizes, included multiple doses of chlorpyrifos and tested two OPs (methyl parathion), had a strong statistical approach. Weaknesses: Did not include an ITI delay in the RAM test. Overall, this was one of the stronger studies.

In this study (Carr *et al.*, 2001) Sprague-Dawley CD rats (Charles River) were used and bred in-house. Rats were assigned to four groups with whole litters assigned to each group with a total of 5 litters per group with two offspring tested per sex per litter, *i.e.*, final numbers were 10 per sex per treatment group. Rats were gavaged on P1-21 every other day with corn oil, or corn oil containing a lower dose CFP 3 mg/kg, a mid-dose P1-5 of 3 mg/kg, P7-21 of 6 mg/kg, or a higher dose of P1-5 of 3 mg/kg, P7-13 of 6 mg/kg, and P15-21 of 12 mg/kg. The offspring were tested in an open-field on P10 and P12 for 3 min. each time and on P14, 16, 18, 20, 25, and 30 for 6 min each time). Statistically, they used a general linear model ANOVA and they set a significance level at $p<0.01$. The pairwise method used was not mentioned. They found reduced locomotion at P25 and P30 at the mid and high dose levels in both males and females. Strengths: They accounted for litter effects in the design and used appropriate statistical methods. They included three doses levels of chlorpyrifos plus control. Weaknesses: The sample size of five litters per group made the study under-powered and they slightly over-sampled per litter by using two per sex per litter.

This study (Jett *et al.*, 2001) used Long-Evans (Charles River) rats and obtained timed-pregnant animals. They culled litters to 10 and randomized pups among dams (no mention of sex balancing). Offspring were treated with chlorpyrifos on P7, 11, 15 at doses of 0.3 or 7 mg/kg given S.C. in oil to entire litters. A major concern is this sentence: "2 or more litters were used for random selection of pups used in behavioral studies". This suggests that severe over-sampling from a few as two litters were used. Another group was treated with chlorpyrifos (same doses) on P22 & 26). They tested offspring in the MWM on P24-28 hence in postweaning treatment group one dose was given in the middle of the testing regimen. The MWM was 90 cm in diameter and the goal platform was 25 cm^2; hence search ratio was 245:1 (which is within the range typically used for mice). They tested for 5 days, 2 trials on day-1, then 1 trial per day on days 2-5 with a probe trial 30 min after last training trial. They gave cued trials on day-5 the method used was not described. Statistically, they used ANOVAs but the details are not provided. They report a main effect on MWM latency for both CFP exposed groups but the method of pairwise comparisons is not given, and they report a high dose effect on the probe trial for time spent in the target quadrant. There is no mention of path length; no differences were reported on cued performance or on swim speed. Their final Ns were (M/F): Control 10/10, low dose 10/9, high dose 9/8. For the postweaning chlorpyrifos treatment, they report a treatment main effect which they report sorting by day by an unspecified statistical method. Looking at the figure, most of the effect

appears to be on days 3-5; also both CFP groups spent less time in target quadrant on the probe trial. They report no speed differences and state that no cued differences were found but the data are not shown. For the postnatal experiment the Ns are (M/F): Control 4/3; low dose 4/3; high dose 4/4. Strengths: They did most of the procedures in the MWM that should be included, such as assessing swim speed and cued performance. Weaknesses: There appears to be no control for litter effects and the number of litter used was as low as two, indicating a severely under-powered and potentially fatally flawed design. This is unfortunate because this is one of the few studies to test a lower dose of chlorpyrifos (0.3 mg/kg). Also, giving the probe trial for the MWM shortly after the last training trial provides somewhat ambiguous information. Changes may be attributed to either working or reference memory since the interval was too short to rule out working memory as a principal contribution.

Pre- and Postnatal Studies

This study (Maurissen *et al.*, 2000) used Sprague-Dawley CD rats (Charles River) bred in-house. They used 20 litters per treatment group (conception = E0). Treatment was by gavage on E6-P10. Doses given were 0, 0.3, 1.0, 5.0 mg/kg/day, but note that the postnatal exposure was to the dams, not the pups. On P4 litters were culled to 5/5 M/F. They used different subsets of pups per litter for different tests as follows: Set-1: Brain morphometry on P11; Set-2: Delayed spatial alternation on P22-24 and again on P61-90 (but using only 8/sex/group from 16 litters rather than 10/sex/group from all 20 litters); Set-3: Locomotor activity on P13, 17, 21, 60, and ASR on P22 and 61 (used 1 M/1 F from all 20 litters/group for these tests); Set-4: Developmental landmarks; body weight, and on P65-70 brained dissected and fixed for neuropathology. For the delayed spatial alternation test they used 3 delay intervals at each test age. For locomotion they used a 40 x 25 photocell system. For ASR they gave 50 trials with acoustic signals of 120 dB with ITI = 10 s. Data were analyzed by ANOVA with litter taken into account. P-values were considered significant at $P<0.02$. Where treatment main effect occurred, follow up was by a stepwise approach by first removing of high dose and re-analyzing the data, if still significant, then removal of the mid dose and reanalyze, *etc.* Interactions were followed up using simple-effect ANOVAs. Results: They report delayed vaginal patency at the high dose; delayed pinna detachment and prenuptial separation at $p<0.03$ and $P<.05$ neither of which reached their $P<.02$ cut-off. However these would be more commonly regarded as significant. They found no significant effects on the delayed spatial alternation test. They found no significant effects on locomotor activity. They found a trend on ASR latency at $P<.03$ but no effect on startle amplitude (the principal measure on this test). Strengths: This study has the most robust sample size in this entire group of 21 articles. They controlled for litter effects and used appropriate statistical methods. They delayed spatial alternation test was a particular strength and in their Fig. 7 they show the working memory decay as a function of the length of the delay interval, proving that they are measuring working memory. They included control plus 3 dose levels of chlorpyrifos and this is one of the only studies to test a low dose (0.3 mg/kg chlorpyrifos). They also avoided the use of DMSO as the vehicle. Weaknesses: The delayed spatial alternation test used much reduced sample sizes compared to the study as

a whole (8 per sex per group rather than 20). While this is of some concern, there were no trends in the results suggesting that a latent effect might have been missed. For the ASR, they performed a simple startle habituation test rather than a PPI procedure which is more informative. Note: It may be significant given the discussion at the meeting that they used an oral rather than subcutaneous route of exposure. Given the first pass metabolism of chlorpyrifos, this may have implications for the total amount of exposure the rats received.

This study (Braquenier *et al*., 2010) used CD-1 mice (Charles River) but the number of dams was not given nor was how conception was dated (a relevant factor in studies with prenatal exposure). Doses of chlorpyrifos were 0, 0.2, 1.0, 5 mg/kg/d given on E14-P14. For the postnatal CFP exposure the compound was given to dams (not pups). Litters were culled to 4M/4F with one female per litter used for testing. They tested offspring for locomotor activity (5 min/session) for 8 days (scored manually). They also did a 5 min light/dark test (mice started on the light side). EPM was assessed (standard 5-min procedure). Data were analyzed by ANOVA with follow-up by Dunnett's test. Results: They found no significant effects on locomotor activity. For the light/dark test, they found no effect on the percent of time spent in the dark compartment but a significant decrease in the percent of time in the light compartment in the chlorpyrifos1 group, but not in the chlorpyrifos0.2 or 5 mg/kg groups. Side transitions showed trend (P<.08) which they followed-up anyway and report a significant reduction in the CFP1 group by Dunnett. In the EPM they report a trend at p<.10, did Dunnett follow up tests anyway and found reduced time in open arms in the CFP1 group but not the other dose groups. They also found that the percentage of arm entries into open arms was significantly decreased in the CFP1 group. Total transitions were not significantly affected. Strengths: Tested three dose levels of chlorpyrifos, including a low dose (0.3 mg/kg), and gave the compound in oil rather than DMSO, and used standard methods. Weaknesses: No indication that litter effects were accounted for in the design or statistical analyses. Effects were found only at the 1 mg/kg dose and were not dose-dependent, they did follow-up tests on many trends that were not statistically significant and declared these follow-up effects to be significant findings.

Summary

Among these 21 reviewed articles (which include more than 21 experiments), many effects are reported at chlorpyrifos doses ranging from at 1.0 to 7.0 mg/kg (and in one study 10 mg/kg). However, these are dose levels known to significantly inhibit cholinesterase in RBC; therefore, based on these 21 studies, cholinesterase inhibition is an adequate threshold as no credible evidence of neurobehavioral effects below 1.0 mg/kg were found.

Three studies tested doses < 1.0 mg/kg chlorpyrifos. Two studies used 0.3 mg/kg and one used 0.2 mg/kg (Jett *et al.*, 2001; Braquenier *et al.*, 2010; Maurissen *et al.*, 2000, respectively). Of these, two found no effects at these lower doses *(Maurissen et al., 2000;Braquenier et al.*, 2010). Only one study found effects at 0.3 mg/kg (Jett *et al.*, 2001); however, this study contains serious methodological flaws which are of sufficient

magnitude to cast serious doubt on the credibility of the findings. While the data from Jett *et al.* (2001) raise the possibility of neurobehavioral effects at 0.3 mg/kg/d, their data require replication in a study that is properly designed, adequately powered, and appropriately analyzed. Until such time as new data at such lower doses become available, it is concluded that no dose <1.0 mg/kg in any neurodevelopmental behavioral study shows evidence of adverse effects (or of any effects, even including those outcome measures whose effect is indeterminate or unknown).

In addition, effects of chlorpyrifos at 1.0 mg/kg are difficult to interpret because of methodological limitations, inconsistencies, and variation in study design, sometimes lack of control for litter effects, oversampling issues, behavioral methods used, and lack of dose-response findings.

Above 1.0 mg/kg, the data show somewhat more consistency, but even here, dose-response experiments are the exception. At 5.0 mg/kg of chlorpyrifos, reduced body weight is sometimes seen, and at doses above 5.0 mg/kg, increased mortality may occur along with other evidence of toxicity. Given this, a significant gap in the literature of dose-response studies exists in the range downward toward 0.2 mg/kg and extending up to and including doses previously tested of 1.0-2.0 mg/kg that is needed in order to determine what, if any, dose-effect curve for neurobehavioral effects occurs in this range.

It appears that neurobehavioral outcomes are more sensitive to prenatal and prenatal-neonatal exposures than to neonatal exposure alone . This implies that prenatal exposure may be the exposure period contributing to this observation, but unfortunately, most of the pre- and neonatal studies are not entirely informative because the neonatal exposure was to the dam rather than directly to the progeny. This makes it unclear what the dose to the offspring actually was . More studies, especially dose-response studies, in the lower dose ranges with exposure from implantation to the end of major neurogenesis (approximately P20) are needed, again with doses below 1.0 mg/kg and with concomitant measurement of maternal, fetal, and neonatal cholinesterase activity.

Exposures in many of the existing studies are for only a narrow interval during gestation or the neonatal period. Prenatal exposures should be from E6-20 to 21 for rats, and E6-18 or 19 in mice in order to span most of early brain development (equivalent to human first and part of the second trimester). And for neonatal treatment, exposures should be from shortly after birth to approximately P20 (equivalent to latter half of the second and all of the third trimester equivalent brain development for humans). If the critical period or most sensitive period is within this range, then such comprehensive exposure should cover the entire span of CNS development that represents the species being modeled, *i.e*, human beings.

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all of the third trimester equivalent brain development for humans). If the critical period or most sensitive period is within this range, then such comprehensive exposure should cover the entire span of CNS development that represents the species being modeled, *i.e*, human beings.

In the prenatal studies, the use of timed-pregnant females shipped from breeders is problematic for behavioral studies because maternal stress, even if regarded as equivalent across dams assigned to the treated and control groups, introduces a variable that has the potential to interact with the independent variable. Were maternal stress to interact with chlorpyrifos, it would confound the outcome and make a result difficult to interpret (which is exactly what is found in the 21 reviewed studies). Since no one has tested for this, it is currently impossible to rule it out.

Many studies use diurnal and some nocturnal testing. If additional dose-response studies are undertaken, this factor should be held constant so that results can be better compared.

ATTACHMENT J

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

JUL 16 2012

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Re: Chlorpyrifos petition dated September 12, 2007

Dear Mr. Colangelo and Dr. Reeves:

This letter constitutes the U.S. Environmental Protection Agency's (EPA or Agency) partial response to the petition dated September 12, 2007 (Petition), submitted jointly by the Natural Resources Defense Council (NRDC) and Pesticide Action Network North America (PANNA). The petition specifically requested that EPA revoke all tolerances and cancel all registrations for the insecticide chlorpyrifos.[1] The petition provided that it was filed pursuant to 21 U.S.C. section 346a(d) (section 408(d)) of the Federal Food Drug and Cosmetic Act (FFDCA)).

In the petition, NRDC and PANNA claimed that in 2006, when the Agency finalized its Organophosphorus (OP) Cumulative Risk Assessment – 2006 Update[2] (CRA) for all organophosphates, which included chlorpyrifos, and reaffirmed the 2001 chlorpyrifos Interim Reregistration Eligibility Decision (IRED), it failed to properly consider data that demonstrated adverse effects from chlorpyrifos exposure. More specifically, petitioners made the following ten claims regarding the Agency's cumulative assessment for all organophosphates and the chlorpyrifos IRED:

[1] Petition of Natural Resources Defense Council and Pesticide Action Network North America to Revoke All Tolerances and Cancel All Registrations for the Pesticide Chlorpyrifos (September 12, 2007) at 1. (hereinafter Petition) Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0005.
[2] U.S. Environmental Protection Agency (EPA) (2006). Organophosphorus Cumulative Risk Assessment – 2006 Update. Available at http://www.epa.gov/opp00001/cumulative/2006-op/op_cra_main.pdf.

1. EPA ignored genetic evidence of vulnerable populations
2. EPA needlessly delayed a decision regarding endocrine disrupting effects
3. EPA ignored data regarding cancer risks
4. EPA's CRA misrepresented risks, failed to apply FQPA 10X safety factor[3]
5. EPA over-relied on registrant data
6. EPA failed to properly address the exporting hazard from chlorpyrifos
7. EPA failed to quantitatively incorporate data demonstrating long-lasting effects from early life exposure to chlorpyrifos in children
8. EPA disregarded data demonstrating that there is no evidence of a safe level of exposure during pre-birth and early life stages
9. EPA failed to cite or quantitatively incorporate studies and clinical reports suggesting potential adverse effects below 10% cholinesterase inhibition
10. EPA failed to incorporate inhalation routes of exposure

This partial Agency response to the petition addresses the first six claims listed above: genetic evidence of vulnerable populations; endocrine disrupting effects; cancer risks; CRA misrepresents risks, fails to apply FQPA 10X safety factor; over-reliance on registrant data; and exporting hazard. EPA's response to three of petitioners' remaining four claims -- that EPA failed to quantitatively incorporate data exhibiting long-lasting effects from early life exposure to chlorpyrifos in children; that EPA disregarded data demonstrating that there is no evidence of a safe level of exposure during pre-birth and early life stages; and that EPA failed to cite or quantitatively incorporate studies and clinical reports suggesting potential adverse effects below 10% cholinesterase inhibition -- involve highly complex assessments, using precedent setting risk assessment methodologies. For this reason, consistent with EPA's external scientific peer review policy, the Agency sought advice on these issues from the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel (SAP) at a meeting that occurred April 10-13, 2012. As to the petitioners' fourth remaining claim -- that EPA failed to incorporate inhalation routes of exposure -- EPA is today releasing its "Evaluation of the Potential Risks From Spray Drift and the Impacts of Potential Risk Reduction Measures"[4] that further refines its analysis of spray drift from chlorpyrifos that was presented in the preliminary human health risk assessment (HHRA) released in July 2011. In connection with this spray drift assessment, the chlorpyrifos registrants have agreed to implement label mitigation (in the form of rate reductions and spray drift buffers) that will reduce risks to bystanders from spray drift. EPA's spray drift assessment and the associated mitigation action are not intended to provide a complete response to petitioners' fourth claim since this mitigation action does not take into account potential exposures from volatilization following chlorpyrifos applications. That work is ongoing. Further, the spray drift risk assessment may be impacted by those issues reviewed by the recent SAP. Accordingly, the Agency will address this claim fully when it provides its complete response in December 2012.

[3] For convenience's sake, the legal requirements regarding the additional safety margin for infants and children in section 408(b)(2)(C) are referred to throughout this response as the "FQPA 10X safety factor" or simply the "FQPA safety factor. Due to Congress' focus on both pre- and post-natal toxicity, EPA has interpreted this additional safety factor as pertaining to risks to infants and children that arise due to pre-natal exposure as well as to exposure during childhood years.

[4] Available at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2008-0850.

In connection with this peer review process, the Agency has recently received the final SAP report[5], dated July 11, 2012. EPA will carefully review and consider the SAP's recommendations in developing the Agency's response to the remaining petition issues while also informing the final human health risk assessment for the statutorily mandated registration review program. Following EPA's complete written response to the petition, which it plans to provide petitioners by the end of this year, EPA intends to work to complete its human health risk assessment in connection with the registration review of chlorpyrifos under section 3(g) of FIFRA. That assessment will include consideration of issues not raised in the petition, including human dietary exposures from drinking water. It is important to note, however, that EPA may take regulatory action at any time if and when it determines that existing tolerances are unsafe or that chlorpyrifos presents unreasonable adverse effects on the environment.

The first section of this response provides the applicable statutory and regulatory background. The second section discusses this petition in the context of the legal framework of FIFRA and FFDCA. The third section discusses the regulatory background for chlorpyrifos. The fourth section contains EPA's response to each of the six issues identified above. The final section is the conclusion.

I. Statutory and Regulatory Background/Framework

A. FFDCA/FIFRA and Applicable Regulations

EPA establishes maximum residue limits, or ''tolerances,'' for pesticide residues in food and feed commodities under section 408 of the FFDCA.[6] Without such a tolerance or an exemption from the requirement of a tolerance, a food containing a pesticide residue is ''adulterated'' under section 402 of the FFDCA and may not be legally moved in interstate commerce.[7] Monitoring and enforcement of pesticide tolerances are carried out by the U.S. Food and Drug Administration and the U.S. Department of Agriculture. Section 408 was substantially rewritten by the Food Quality Protection Act of 1996 (FQPA), which added the provisions discussed below establishing a detailed safety standard for pesticides, additional protections for infants and children, and the estrogenic substances screening program.[8]

EPA also regulates pesticides under the FIFRA.[9] While the FFDCA authorizes the establishment of legal limits for pesticide residues in food, FIFRA requires the approval of pesticides prior to their sale and distribution,[10] and establishes a registration regime for regulating the use of pesticides. FIFRA regulates pesticide use in conjunction with its registration scheme by requiring EPA review and approval of pesticide labels and specifying that use of a pesticide inconsistent with its label is a violation of federal law.[11] In the FQPA, Congress integrated action under the two statutes by requiring that the safety standard under the FFDCA be used as a criterion in FIFRA registration actions as to pesticide uses which result in dietary

[5] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2012-0040-0029
[6] 21 U.S.C. 346a
[7] 21 U.S.C. 331, 342
[8] Public Law 104– 170, 110 Stat. 1489 (1996).
[9] 7 U.S.C. 136 et seq.
[10] 7 U.S.C. 136a(a).
[11] 7 U.S.C. 136j(a)(2)(G).

risk from residues in or on food,[12] and directing that EPA coordinate, to the extent practicable, revocations of tolerances with pesticide cancellations under FIFRA.[13]

In addressing the review of petitions, however, Congress has expressly provided that any issue that can be raised through the FFDCA review process can only be reviewed through that process.[14] Accordingly, to the extent a petition to revoke tolerances and cancel registrations raises issues relevant to the establishment or revocation of tolerances, EPA's response to those issues may be challenged only through the administrative and judicial review procedures provided in section 408 of the FFDCA and are not reviewable under FIFRA or any other provision of law.

II. Legal Framework and the NRDC and PANNA Petition

A. FFDCA

All but one of the issues raised in the petition relate to EPA's establishment of tolerances under the FFDCA. For this reason, as explained in section I, the FFDCA directs that consideration of these petition issues be undertaken under FFDCA section 408. Under the FFDCA, EPA takes final action on a petition to revoke tolerances by either issuing an order in the Federal Register denying the petition or by publishing a final rule revoking the tolerances. EPA does not intend to issue a formal denial in the Federal Register for the five issues addressed in this response that are subject to FFDCA review until after it completes its review of the four remaining petition issues. That way, petitioners will not be compelled to assess whether to file objections to EPA's responses on separate occasions and EPA will not be compelled to produce separate responses. However, if petitioners wish to begin the objections process on today's partial response, EPA will publish a formal denial order for those claims. As explained previously, EPA intends to address, in writing, the remaining issues raised in the petition by December 2012. Following this written response, to the extent EPA denies the petition, EPA would expect to publish any denial order by February 2013. While the December response will address all the remaining issues raised in the petition, it is possible that EPA will not be taking final agency action on all the petition issues immediately following the December 2012 response.

While petitioners have raised a number of issues related to the assessment of the toxicity of chlorpyrifos, the petition did not address in any detail, dietary exposure to chlorpyrifos that must be taken into account in determining whether tolerances are safe. Reassessing the exposure to chlorpyrifos is one of the issues EPA intends to address in the registration review assessment of the chemical, which is currently underway. It is possible that if EPA concludes that the toxicity profile of chlorpyrifos needs to be modified based upon this reassessment, the final decision on the petition would need to wait for the conclusion of the chlorpyrifos exposure reassessment under the registration review program. The registration review assessment will include consideration of issues not raised in the petition, including human dietary exposures from drinking water. It is important to note, however, that EPA may take regulatory action at any time

[12] 7 U.S.C. 136(bb).
[13] 21 U.S.C. 346a(l)(1).
[14] 21 U.S.C. 346a(h)(5); NRDC v. Johnson, 461 F.3d 164, 176 (2d Cir. 2006).

if and when it determines that existing tolerances are unsafe or that chlorpyrifos presents unreasonable adverse effects on the environment.

Tolerances are established, amended, or revoked by rulemaking under the unique procedural framework set forth in the FFDCA. Generally, a tolerance rulemaking is initiated by the party seeking to establish, amend, or revoke a tolerance by means of filing a petition with EPA.[15] EPA publishes in the Federal Register a notice of the petition filing and requests public comment.[16] After reviewing the petition, and any comments received on it, EPA may issue a final rule establishing, amending, or revoking the tolerance, issue a proposed rule to do the same, or deny the petition.[17]

Once EPA takes final action on the petition by establishing, amending, or revoking the tolerance or denying the petition, any party may file objections with EPA and seek an evidentiary hearing on those objections.[18] Objections and hearing requests must be filed within 60 days.[19] The statute provides that EPA shall ''hold a public evidentiary hearing if and to the extent the Administrator determines that such a public hearing is necessary to receive factual evidence relevant to material issues of fact raised by the objections.''[20] EPA regulations make clear that hearings will only be granted where it is shown that there is ''a genuine and substantial issue of fact,'' the requestor has identified evidence ''which, if established, resolve one or more of such issues in favor of the requestor,'' and the issue is ''determinative'' with regard to the relief requested.[21] Further, a party may not raise issues in objections unless they were part of the petition and an objecting party must state objections to the EPA decision and not just repeat the allegations in its petition.[22] EPA's final order on the objections is subject to judicial review in the U.S. Court of Appeals.[23]

B. FIFRA and Exporting Hazard

The exporting hazard issue raised in the petition is the sole claim that raises issues that are subject to FIFRA review. Petitioners claim that "unless chlorpyrifos is banned, and all tolerances cancelled [sic], chlorpyrifos will continue to be used, often unsafely, in other countries thus creating a health and environmental hazard in those countries and on contaminated food re-entering the US."[24] While the claim includes a request to both cancel registrations and revoke tolerances, the policy arguments raised in the petition regarding the consideration of the international impacts of the U.S. registration of chlorpyrifos are not relevant to the establishment of tolerances under the FFDCA. FIFRA Section 6 authorizes EPA to cancel pesticide registrations that do not comply with FIFRA and, in certain circumstances, to suspend those registrations pending the completion of cancellation proceedings. EPA takes final Agency action

[15] See 21 U.S.C. 346a(d)(1).
[16] 21 U.S.C. 346a(d)(3).
[17] 21 U.S.C. 346a(d)(4).
[18] 21 U.S.C. 346a(g)(2).
[19] *Id*.
[20] 21 U.S.C. 346a(g)(2)(B).
[21] 40 CFR 178.32(b)
[22] *See Nat'lCorn Growers Assoc., et al. v. EPA* 613 F.3d 266 (D.C. Cir. 2010).
[23] 21 U.S.C. 346a(h)(1).
[24] Petition at 21.

when it issues a response to petitioners either denying their petition or by initiating and completing the cancellation process under FIFRA.

EPA considers this portion of the response to NRDC's petition to be a final action, and believes the petitioner may challenge now this portion of the Agency's petition denial in federal court pursuant to section 16 of FIFRA. Because, as explained below, EPA is today denying petitioners' request to cancel on the basis of the export hazard issue, this letter will constitute a final Agency action as it relates to that specific issue. As noted, the remaining issues are subject to review as provided in section 408 of the FFDCA.

III. Background

In 2000, the chlorpyrifos technical registrants entered into an agreement with the Agency regarding the use of chlorpyrifos which eliminated virtually all homeowner residential uses, phased-out all termiticide uses, eliminated use on tomatoes, and changed use on grapes and apples from a foliar use to a dormant use.

In September 2001, the Agency completed its IRED for chlorpyrifos. At the time of the IRED the Agency was also working on the OP CRA, which addresses all those OP pesticides sharing the common mechanism endpoint, acetylcholinesterase (AChE) inhibition. Specifically, the members of this class share the ability to bind to and phosphorylate the enzyme AChE in both the central (brain) and peripheral nervous systems.

In August 2006, the Agency released its 2006 Update to the OP CRA. With EPA's 2006 release of the OP CRA, all reregistration eligibility decisions (REDs) for individual OP pesticides, including chlorpyrifos, were considered complete. OP IREDs, therefore, were considered completed REDs.

In September 2007, EPA received NRDC and PANNA's joint petition to revoke all tolerances and cancel all registrations for chlorpyrifos. The petition largely challenged the conclusions of the Agency's IRED and 2006 OP CRA.

Although EPA completed reregistration and tolerance reassessment for the OP pesticides in 2006, the Agency made the decision to move chlorpyrifos and other OP pesticides forward in the re-evaluation schedule so that they began registration review in 2008 and 2009. The chlorpyrifos registration review docket opened in 2009.

In connection with its ongoing re-evaluation of chlorpyrifos and analyses of the complex issues raised in the petition, in 2008 EPA convened an SAP meeting to review a draft science issue paper on the human health effects of chlorpyrifos to provide a preliminary review of the scientific literature on experimental toxicology and epidemiology studies available at that time. Specifically, the focus was on studies that evaluated the effects of chlorpyrifos on infants and children from *in utero* and/or post-natal exposures and on studies that evaluated population variability with respect to response to paraxonase (PON1). In summary, the SAP expressed confidence that the studies conducted by Columbia University are epidemiologically sound. The SAP agreed with the Agency that human epidemiological studies have utility for risk

characterization, but not as the principal basis for establishing the point of departure (PoD), in part due to uncertainty in attributing observed adverse neurodevelopmental effects in children solely to chlorpyrifos, when exposure was to multiple anticholinesterase insecticides.[25]

In December 2009, EPA convened a SAP to review scientific issues associated with interpreting risks related to the field volatilization of conventional pesticides.[26] The objectives of the meeting were to review both the exposure and hazard aspects of the risk assessment process. The primary focus of the discussion on exposure assessment was on methods for predicting emissions from treated fields in lieu of having actual field volatilization studies as well as how such information should be considered in exposure assessment. With regard to hazard evaluations, the impact on inhalation risk estimates based on differences in how doses are experimentally administered to rodents (oral or inhalation) was considered. The Agency's goal for the SAP review was to receive feedback on procedures, methodologies, and data inputs to inform the assessment of bystander exposure resulting from field volatilization of conventional pesticides. The procedures, refined in part from the SAP's feedback, will inform the Agency's analysis as it considers chlorpyrifos emissions data identified in the literature, as well as data from a field study Dow Agrosciences undertook and submitted to the Agency on July 6, 2012.

In February 2010, EPA convened an SAP to review the Agency's draft "Framework for Incorporating Human Epidemiologic & Incident Data in Health Risk Assessment,"[27] which provides the conceptual foundation for evaluating multiple lines of scientific evidence in a human health risk assessment. This draft framework draws from the mode of action framework[28] and its use of the modified Bradford Hill Criteria[29] and, thus, explicitly considers such concepts as strength, consistency, dose response, temporal concordance and biological plausibility.

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[25] U.S. EPA (2008). Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held September 16-18, 2008 on the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos. Available at http://www.epa.gov/scipoly/sap/meetings/2008/september/sap0908report.pdf

[26] U.S. EPA (2009). Transmittal of Meeting Minutes of the FIFRA SAP Meeting Held December 1-3, 2009 on the Scientific Issues Associated with "Field Volatilization of Conventional Pesticides." Available at http://www.epa.gov/scipoly/sap/meetings/2009/december/120309meetingminutes.pdf

[27] U.S. EPA (2010). Transmittal of Meeting Minuets of the FIFRA Scientific Advisory Panel Meeting on the Draft Framework and Case Studies on Atrazine, Human Incidents, and the Agricultural Health Study: Incorporation of Epidemiology and Human Incident Data into Human Health Risk Assessment. Available at http://www.epa.gov/scipoly/sap/meetings/2010/020210minutes.pdf

[28] U.S. EPA (2007). Framework for Determining a Mutagenic Mode of Action for Carcinogenicity: Using EPA's 2005 Cancer Guidelines and Supplemental Guidance for Assessing Susceptibility from Early-Life Exposure to Carcinogens. Available at http://www.epa.gov/osa/mmoaframework/pdfs/MMOA-ERD-FINAL-83007.pdf

[29] U.S. EPA (1999). Guidelines for Carcinogen Risk Assessment, Risk Assessment Forum, SAB Review Draft. Available at http://www.epa.gov/cancerguidelines/draft-guidelines-carcinogen-ra-1999.htm; Sonich-Mullin, C., R. Fielder, J. Wiltse et al, 2001. IPCS conceptual framework for evaluating a mode of action for chemical carcinogenesis. Regul Toxicol Pharmacol. 34:146-152 Meek, M.E., J.R. Bucher, S.M. Cohen et al, 2003. A framework for human relevance analysis of information on carcinogenic modes of action. Crit. Rev. Toxicol. 33:591-653; Seed, J., E.W. Carney, R.A. Corley et al., 2005. Overview: Using mode of action and lifestage information to evaluate the human relevance of animal toxicity data. Crit. Rev. Toxicol. 35(8-9):664-672; U.S. EPA (2005). Guidelines for Carcinogen Risk Assessment. Available at http://www.epa.gov/raf/publications/pdfs/CANCER_GUIDELINES_FINAL_3-25-05.PDF

In July 2011, the Agency released the preliminary HHRA for chlorpyrifos for public comment. The comment period officially closed on October 6, 2011. The Agency received 48 unique comments totaling over 1000 pages, which included a number of significant study citations. The Agency continues to work on reviewing the public comments and studies cited to further inform the final HHRA for chlorpyrifos.

Since the 2008 SAP on chlorpyrifos, and in part due to the SAP's feedback, the Agency has performed further analyses on the existing and new epidemiology studies with mothers and children, available biomonitoring data, and experimental toxicology studies evaluating proposed adverse outcome pathways in the context of human health risk assessment. Specifically, the Agency is evaluating available literature on the potential for chlorpyrifos to cause long term adverse effects from early life exposure, *in vivo* and *in vitro* studies evaluating mechanistic aspects of chlorpyrifos, and the potential for adverse effects below PoDs established from cholinesterase inhibition used for regulatory purposes. This analysis is complicated and multifaceted as it involves many lines of scientific evidence (i.e., *in vivo & in vitro* experimental toxicology studies, explicit consideration of adverse outcome pathway framework analyses, exposure, human epidemiology, and biomonitoring data). As the Agency works to finalize the HHRA and respond to the remaining petition issues, the Agency is working towards a weight of evidence evaluation integrating the epidemiology studies with the experimental toxicology studies for the neurodevelopmental outcomes. As noted previously the Agency convened a FIFRA SAP in April 2012 to review the Agency's analyses to ensure it is utilizing sound science in making its regulatory determinations and has recently received the final SAP report, dated July 11, 2012, of that meeting.

IV. Petition Response

1. Genetic Evidence of Vulnerable Populations

a. Petitioners' claim

Petitioners claim that the Agency failed to calculate an appropriate intra-species uncertainty factor (i.e., within human variability) for chlorpyrifos in both its aggregate and cumulative assessments.[30] They assert that certain relevant, robust data, specifically the Furlong et al. (2006) study[31] that addresses intra-species variability in the behavior of the detoxifying enzyme PON1, indicates that the Agency should have applied an intra-species safety factor "of at least 150X in the aggregate and cumulative assessments" rather than the 10X factor EPA applied.[32] Petitioners conclude by noting that applying an intra-species factor of 100X or higher would require setting tolerances below the level of detection, which therefore should compel EPA to revoke all chlorpyrifos tolerances.

[30] Petition at 6.
[31] Furlong CE, Holland N, Richter RJ, Bradman A, Ho A, Eskenazi B (2006). PON1status of farmworker mothers and children as a predictor of organophosphate sensitivity. Pharmacogenet Genomics. 2006 Mar; 16(3):183-90.
[32] Petition at 6.

b. *Agency Response*

Petitioners are correct that the Agency, as part of the 2006 OP CRA, evaluated, but did not rely on Furlong et al. in setting the intra-species uncertainty factor for that assessment. The Agency did not rely on the results of the PON1 data in the OP CRA because these data do not take into consideration the complexity of OP pesticide metabolism, which involves multiple metabolic enzymes, not just PON1. In addition, EPA believes the methodology utilized in the Furlong et al. study to measure intra-species variability – i.e., combining values from multiple species to determine the range of sensitivity within a single species – is not consistent with well-established international risk assessment practices. Further, EPA believes that petitioners' assertion that the Furlong et al. study supports an intra-species uncertainty factor of at least 150X is based on an analysis of the data that is inconsistent with EPA policy and widely- accepted international guidance on the development of intra-species uncertainty factors. For these reasons, as further explained below, EPA believes it is not appropriate to rely on the results of the Furlong et al. study, or petitioners' interpretation of those results, for purposes of determining the intra-species uncertainty factor. At this time, there is not a reasonable scientific basis for departing from the standard 10-fold intra-species uncertainty factor for extrapolating variability based on PON1.

Addressing human variability and sensitive populations is an important aspect of the Agency's risk assessment process. The Agency is well aware of the issue of PON1 and has examined the scientific evidence on this source of genetic variability. PON1 is one of the key detoxification enzymes of chlorpyrifos. Specifically, PON1 is an A-esterase which can metabolize chlorpyrifos oxon without inactivating the enzyme.[33] Indeed, as part of the 2008 SAP, EPA performed a literature review of PON1 and its possible use in informing the intra-species (i.e., within human variability) uncertainty factor. This literature review can be found in the draft Appendix E: Data Derived Extrapolation Factor Analysis to the draft Science Issue Paper: Chlorpyrifos Hazard and Dose Response Characterization.[34] In sum, the Agency considered available PON1 data from more than 25 studies from diverse human populations worldwide.

The Agency focused on the PON1-192 polymorphism since it has been linked to chlorpyrifos oxon sensitivity in experimental toxicology studies and, has been evaluated in epidemiology studies attempting to associate PON1 status with health outcomes following OP pesticide exposure in adults and children (Holland et al., 2006[35]; Chen et al., 2003).[36] However,

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[33] Sultatos LG; Murphy SD, (1983). Kinetic Analysis Of The Microsomal Biotransformation Of The Phosphorothioate Insecticides Chlorpyrifos And Parathion. Fundemental and Applied Toxicology. 3:16-21.

[34] U.S. EPA (2008). Draft Appendix E available at http://www.epa.gov/scipoly/sap/meetings/2008/september/appendixe.pdf . Draft Science Issue Paper: Chlorpyrifos Hazard and Dose Response Characterization. August 21, 2008. Available at http://www.epa.gov/scipoly/sap/meetings/2008/september/chlorpyrifoscharacter.pdf

[35] Note, Holland et al (2006) and Furlong et al (2006) report findings from the same cohort. The Holland reference provides enzymes activities for specific polymorphisms in Table 4; the Furlong paper does not report such values and provides information primarily in graphical form.

[36] Holland, N., Furlong, C., Bastaki, M., Richter, R., Bradman, A., Huen, K., Beckman, K., and Eskenazi, B. (2006). Paraoxonase polymorphisms, haplotypes, and enzyme activity in Latino mothers and newborns. Environ. Health Perspect. 114(7), 985-991; Chen, J., Kumar, M., Chan, W., Berkowitz, G., and Wetmur, J. (2003). Increased Influence of Genetic Variation on PON1 Activity in Neonates. Environmental Health Perspective 111, 11:1403-9

EPA believes that focusing on PON1 variability in isolation from other metabolic action is not an appropriate approach for developing a data-driven uncertainty factor. The Agency solicited feedback from the SAP on the utility of the PON1 data, by itself, for use in risk assessment; the SAP was similarly not supportive of using such data in isolation. Specifically the SAP report states:

> *"…the information on PON1 polymorphisms should not be used as the sole factor in a data-derived uncertainty factor for two main reasons: 1) it is only one enzyme in a complex pathway, and is subsequent to the bioactivation reaction; therefore it can only function on the amount of bioactivation product (i.e., chlorpyrifos-oxon) that is delivered to it by CYP450); and 2) the genotype of PON1 alone is insufficient to predict vulnerability because the overall level of enzyme activity is ultimately what determines detoxification potential from that pathway; thus, it is better to use PON1 status because it provides information regarding PON1 genotype and activity. Some of the data from laboratory animal studies in PON knockout animals are using an unrealistic animal model and frequently very high dose levels, and do not reflect what might happen in humans."* [37]

Based on a detailed review of the literature and the comments from the SAP, the Agency has determined that such data are not appropriate for use alone in deriving an intra-species uncertainty factor for use in human health risk assessment. As indicated by the SAP report, multiple factors (e.g., other enzymes such as P450s, carboxylesterases, butyrlcholinesterase) are likely to impact potential population sensitivity, rendering the results of the PON1 data, by themselves, insufficiently reliable to support a regulatory conclusion about the potential variation of human sensitivity to chlorpyrifos. It is noteworthy that a recent report by the CDC-ATSDR,[38] is in agreement with EPA's conclusion. It states that the "correlation between PON-1 genotype, cholinesterase activity, and clinical toxicity needs to be further evaluated." Population variability data on PON1 would be more useful incorporated into a physiologically-based pharmacokinetic model which would account for all the metabolic processes relevant for chlorpyrifos at a range of dose levels.

Since the 2008 SAP, several epidemiological studies have been published that considered the association between PON1 status/genotype and health outcome. Hofmann et al. (2009) recently reported associations between PON1 status and inhibition of butyrylcholinesterase in a group of pesticide handlers in Washington. The authors note that this study requires replication with larger sample size(s) and more blood samples.[39] Given the limitations of Hofmann et al.,

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[37] U.S. EPA (2008). Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held September 16-18, 2008 on the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos. Available at http://www.epa.gov/scipoly/sap/meetings/2008/september/sap0908report.pdf at 61.

[38] Matthews, A.R., Sutter, M.E., Rentz, D.E. (2011) Serum Paraoxonase-1 (PON-1) Genotype and Exposure to Organophosphorous Insectides—Is There a High-Risk Population? J. Med. Toxicol. (2011) 7:243–247 DOI 10.1007/s13181-011-0166-2.

[39] Hofmann, J.N., Keifer, M.C., Furlong, C.E., De Roos, A.J., Farin., F.M., Fenske, R.A., van Belle, G., Checkoway, H. (2009) Serum Cholinesterase Inhibition in Relation to Paraoxonase-1 (PON1) Status among Organophosphate-Exposed Agricultural Pesticide Handlers./ Environ Health Perspect 117:1402–1408 (2009). doi:10.1289/ehp.0900682. Available at http://dx.doi.org/ [Online 9 June 2009].

the Agency has not drawn any conclusions from this study. The Q/R-192 and/or C/T -108 polymorphism at the promoter site have been evaluated recently as a factor affecting birth or neurobehavioral outcomes following gestational exposure to OP pesticides.[40] These studies (Eskanazi., et al., 2010; Harley et al., 2011; Engel et al., 2011) were evaluated by EPA in preparation for the April 2012 SAP review.

Petitioners further emphasize that the Furlong et al. study supports an intra-species uncertainty factor of over 164X given the range of variability seen in that study. The 164X value is derived from sensitivity observed in transgenic mice expressing human PON1Q-192 compared with mice expressing human PON1R-192 combined with the range of plasma arylesterase from the newborn with the lowest PON1 level compared with the mother with the highest PON1 level from a group of 130 maternal-newborn pairs from the CHAMACOS (Center for the Health Assessment of Mothers and Children of Salinas) cohort.

EPA believes it is fundamentally at odds with international risk assessment practices to combine values from both mouse and human data to determine the potential range of variability within a single species – regardless of whether the test animals express a human PON1 enzyme. As the 2008 FIFRA SAP explained, PON1 is but a single enzyme that should not be considered in isolation to predict the overall level of enzyme activity that may affect human sensitivity to a substance. Using a 164X intra-species uncertainty factor derived from the Furlong et al. study would take this practice one step further by relying upon combined PON1 values from different species with differing overall metabolic activity to derive the intra-species factor. EPA does not believe this approach is an appropriate means of determining the potential range of intra-species variability.

Finally, petitioners' assertion that the Furlong study supports an intra-species uncertainty factor of at least 150X is based on an analysis of that study that is inconsistent with EPA policy and widely- accepted international guidance on the development of intra-species uncertainty factors. In deriving the intra-species uncertainty factor in its risk assessments, EPA is guided by the principles of the 2005 IPCS guidance on chemical specific adjustment factors.[41] The guidance recommends that intra-species factors should be extrapolated from a measure of central tendency in the population to a measure in the sensitive population (i.e., to extrapolate from a typical human to a sensitive human). This is conceptually consistent with the way EPA applies the intra-species uncertainty factor. To base the factor on the difference between the single lowest and highest measurements in a given study, as petitioners suggest in this instance, would

[40] Eskenazi,B; Huen, K., Marks, A., Harley, K.G., Bradman, A., Boyd Barr, D., Holland, N. (2010) PON1 and Neurodevelopment in Children from the CHAMACOS Study Exposed to Organophosphate Pesticides in Utero. Environmental Health Perspectives. Vol 118 (12): 1775-1781); Engel,S.M., Wetmur, J., Chen, J., Zhu, C., Boyd Barr, D., Canfield, R.L., Wolff, M.S., (2011) Prenatal Exposure to Organophosphates, Paraoxonase 1, and Cognitive Development in Childhood Environ Health Perspect 119:1182–1188 (2011). doi:10.1289/ehp.1003183 [Online 21 April 2011]; Harley KG, Huen K, Schall RA, Holland NT, Bradman A, et al. (2011) Association of Organophosphate Pesticide Exposure and Paraoxonase with Birth Outcome in Mexican-American Women. PLoS ONE 6(8): e23923. doi:10.1371/journal.pone.0023923.

[41] IPCS (International Programme on Chemical Safety) 2005. Chemical-Specific Adjustment Factors for Interspecies Differences and Human Variability: Guidance Document for Use of Data in Dose/Concentration-Response Assessment. Harmonization Project Document No. 2. World Health Organization, International Programme on Chemical Safety, Geneva, Switzerland.

likely greatly exaggerate potential intra-species variability. That approach effectively assumes that the PoD in an EPA risk assessment will be derived from the least sensitive test subject, thereby necessitating the application of an intra-species factor that accounts for the full range of sensitivity across a species. Since EPA does not develop its PoDs in this fashion; the approach suggested by petitioners is not appropriate.

In summary, the Agency has carefully considered the issue of PON1 variability and determined that data addressing PON1 in isolation are not appropriate for use alone in deriving an intra-species uncertainty factor. Further, the derivation of the 164X value advocated by the petitioners is based on combining values from humanized mice with human measured values with a range from highest to lowest; the Furlong et al. derivation is inappropriate and inconsistent with international risk assessment practice.[42] Finally, petitioners' statement that the Furlong et al. study supports an intra-species uncertainty factor of at least 150X likely overstates potential variability. While EPA does believe that further research in this area may be required, in part, because multiple factors most likely impact potential population sensitivity, at this time, there is no scientific basis for departing from the standard 10-fold intra-species uncertainty factor for extrapolating variability based on PON1. The Agency will continue to monitor the scientific literature regarding PON1. At this point in time, however, petitioners' claims regarding PON1 would not be a factor in any risk determination the Agency might make to revoke chlorpyrifos tolerances. EPA therefore intends to deny this aspect of the petition when it publishes its response to the petition in the Federal Register.

2. *Endocrine Disrupting Effects*

a. Petitioners' claim

Petitioners summarize a number of studies evaluating the effects of chlorpyrifos on the endocrine system, asserting that, taken together, the studies "suggest that chlorpyrifos may be an endocrine disrupting chemical, capable of interfering with multiple hormones controlling reproduction and neurodevelopment." The petitioners then assert that EPA should not have delayed consideration of endocrine effects absent finalization of the Endocrine Disruptor Screening Program[43] (EDSP) and should have quantitatively incorporated the studies into the chlorpyrifos IRED.

b. Agency Response

This portion of the petition appears largely to be a complaint about the completeness of EPA's reregistration decision and a request that EPA undertake quantitative incorporation of endocrine endpoints into its assessment of chlorpyrifos. The petition does not explain whether and how endocrine effects should form the basis of a decision to revoke tolerances. The basis for seeking revocation of a tolerance is a showing that the pesticide is not "safe." Petitioners have neither asserted that EPA should revoke tolerances because effects on the endocrine system render the tolerances unsafe, nor have petitioners submitted a factual analysis demonstrating that aggregate exposure to chlorpyrifos presents an unsafe risk to humans based on effects on the

[42] *Id*.
[43] See http://www.epa.gov/endo/

endocrine system. Rather, the petition appears to collect a number of studies suggesting that chlorpyrifos may have effects on the endocrine system and that EPA should have considered those health impacts at reregistration in a quantitative assessment.

To the extent that petitioners are seeking tolerance revocation on these grounds, the petition fails to provide a sufficient basis for revocation because, in addition to the preceding defects, the cited data do not provide quantitative data (i.e. endpoints/PoDs) that indicate endocrine effects at doses that are more sensitive than the PoDs currently used in the chlorpyrifos risk assessment. While the cited studies provide qualitative information that exposure to chlorpyrifos may be associated with effects on the androgen and thyroid hormonal pathways, these data alone do not demonstrate that current human exposures from existing tolerances are unsafe. The Agency noted similar effects during its evaluation of information submitted by People for the Ethical Treatment of Animals (PETA) and the Physicians Committee for Responsible Medicine (PCRM) during its review of existing information as part of EPA's EDSP, as discussed below. Based on the review of that data, EPA concluded that the effects seen in those studies do not call into question EPA's prior safety determinations supporting the existing tolerances; the data do not indicate a risk warranting regulatory action, and the petitioners have provided no specific information to alter this determination.

Consequently, the petition does not support a conclusion that existing tolerances are unsafe due to potential endocrine effects. EPA, therefore, intends to deny this portion of the petition when it publishes its response to the petition in the Federal Register. However, because the cited literature studies provide qualitative information to screen chlorpyrifos for the potential to interact with the estrogen, androgen, and thyroid hormonal pathways, EPA will include them in its upcoming weight of evidence evaluation of chlorpyrifos under EPA's EDSP, as required by section 408(p) of the FFDCA.

As petitioners may be aware, since the filing of the petition, EPA has initiated the evaluation of chlorpyrifos under EPA's EDSP, as required under FFDCA section 408(p).

On April 15, 2009, a Federal Register notice was published in which chlorpyrifos was included in the initial list of chemicals to receive EDSP Tier 1 test orders. The EDSP program is a two-tiered screening and testing program; Tier 1 assays and Tier 2 tests. Tier 1 includes 11 assays in the battery; these data are intended to allow EPA to determine whether certain substances (including pesticide active and other ingredients) have the potential to interact with the endocrine system and cause an effect in humans or wildlife similar to an effect produced by a "naturally occurring estrogen, or other such endocrine effects as the Administrator may designate." The purpose of Tier 2 tests is to identify and establish a quantitative, dose-response relationship for any adverse effects that might result from the interactions with the endocrine system.

On November 5, 2009, EPA issued Tier 1 test orders to the registrants of chlorpyrifos, requiring a battery of 11 screening assays to identify the potential to interact with the estrogen,

androgen, or thyroid hormonal systems.[44] On February 13, 2010, EPA received the 90-day responses to the test orders. In the initial response, the test order recipients agreed to conduct 9 of the 11 assays that comprise the EDSP Tier 1 screening battery. The test order recipients also sought to rely on existing data for two of the 11 assays, the male and female pubertal assays. The Agency also received a submission from PETA and PCRM that cited existing studies that they claimed adequately fulfill the requirements of the EDSP Tier1 battery for chlorpyrifos.

On October 20, 2010, following the review of the Other Scientifically Relevant Information (OSRI) that had been submitted, EPA determined that the male pubertal assay test order was satisfied based on the OSRI submitted by PETA/PCRM; this conclusion was based on an interpretation that the data demonstrated potential interaction with the androgen and thyroid hormonal pathways.[45] For the female pubertal assay, however, EPA determined that the assay was still necessary because of deficiencies and unanswered questions in the studies cited in the OSRI submissions. The study deficiencies included the lack of thyroid weights and thyroid hormone measurements. In addition, organ weights and histopathology data were obtained in adult animals, whereas in the Tier 1 female pubertal assay, this data is obtained in pubertal animals.

The test order recipients disagreed with the Agency's interpretation that the OSRI submitted by PETA/PCRM demonstrated potential interaction with the androgen and thyroid hormonal pathways. Because the test order recipients disagreed with the Agency's interpretation of the data cited for the Male Pubertal Assay, they elected to conduct the full Tier 1 battery of assays. The Agency has received all 11 Tier 1 screening assays and is in the data review process. EPA intends to review these chlorpyrifos data as part of its larger process for reviewing all of the Tier 1 data submitted on List 1 EDSP chemicals.

Consistent with the recommendation of the joint Scientific Advisory Board and FIFRA SAP in 1999 (EPA-SAB-EC-00-013, July 1999), the Agency plans to conduct a mid-course review of the functionality of each assay and the battery as a whole. These performance evaluations of the Tier 1 battery will be conducted on an adequate sample of chemicals and it is further anticipated that these Tier 1 performance review results will be submitted for external scientific peer review by the FIFRA SAP in fiscal year 2013. Specifically, EPA intends to first review the data from each individual assay for several chemicals to ensure that EPA consistently interprets the measurements for particular endpoints in that assay; e.g., EPA will review the thyroid weight measurements reported in the male pubertal assays from a number of chemicals to ensure that the Agency reaches consistent conclusions as to the significance of the reported results. After that process has been completed, EPA will evaluate each chemical's response across the battery of 11 assays to determine whether there is evidence of interaction with the estrogen, androgen, and/or thyroid hormone systems.

EPA believes that the results from the entire battery of Tier 1 screening and Tier 2 testing under the EDSP program are necessary to make the statutory determination of whether a

[44] See http://www.epa.gov/endo/pubs/toresources/index.htm for information related to the status of EDSP test orders/DCIs, status of EDSP OSRI: order recipient submissions and EPA responses, and other EDSP assay information.
[45] See http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0634-0159.

substance may have an effect similar to an effect produced by a naturally occurring hormone (FR Vol, 63, No. 248/Monday December 28, 1998). In other words, a positive result in the Tier 1 screening assays would not be sufficient to make the determination of whether chlorpyrifos interacted with the endocrine system. The citations included in the petition referred to as evidence that chlorpyrifos may affect estrogen, androgen, and thyroid hormonal pathways do not establish quantitative, dose-response relationships for potential endocrine effects, which is the purpose of Tier 2 testing.

The information cited in the petition will be considered along with all other information submitted to the Agency by either the test order recipient or the public, including the information already submitted by the petitioners, using a weight-of-evidence (WoE) approach consistent with the Agency's September 14, 2011 EDSP WoE guidance.[46] Based on this WoE assessment, EPA will determine whether chlorpyrifos has the potential to interact with hormone pathways and, if so, whether any Tier 2 or other, more targeted testing, is required to confirm interaction with specific hormone systems and to characterize any potential effects identified through Tier 1 screening, and to establish the dose response relationships for adverse effects necessary to conduct a quantitative risk assessment.

In summary, EPA believes that evaluating all of the evidence with respect to chlorpyrifos's potential for endocrine disruption through the Agency's standard EDSP process, as explained above, is the appropriate approach to address petitioners' request that EPA incorporate endocrine effects into its risk assessment for chlorpyrifos. This is a transparent and scientifically sound process that has been the subject of external peer review[47] and is designed to ensure that all data are appropriately considered. By relying on this robust scientific process, EPA believes that it will ultimately reach a final, scientifically credible determination more efficiently than if EPA were to conduct repeated reviews of the data in piecemeal and without context.

3. Cancer Risks

a. Petitioners' claim

Petitioners claim that the Agency "ignored" a December 2004 National Institutes of Health Agricultural Health Study (AHS) by Lee et al. (2004) [48] that evaluated the association between chlorpyrifos and lung cancer incidence.[49] The petition summarizes the results of the AHS study, stating that the incidence of lung cancer has a statistically significant association with chlorpyrifos exposure. The petition then asserts that these data are highly relevant and

[46] U.S. EPA (2011), Endocrine Disruptor Screening Program: Weight-of-Evidence: Evaluating Results of EDSP Tier 1 Screening to identify the Need for Tier 2 Testing. Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPPT-2010-0877-0021

[47] See Scientific Advisory Panel, 2008 SAP Meetings, March 25-28, 2008: Review the Endocrine Disruptor Screening Program (EDSP) Proposed Tier-1 Screening Battery. Available at http://www.epa.gov/scipoly/sap/meetings/2008/032508_mtg.htm

[48] Lee WJ, Blair A, Hoppin JA, Lubin JH, Rusiecki JA, Sandler DP, Dosemeci M, Alavanja MC. (2004) Cancer incidence among pesticide applicators exposed to chlorpyrifos in the Agricultural Health Study. *J Natl Cancer Inst, 96*(23), 1781-1789. (hereinafter Lee et al. 2004).

[49] Petition at 10

therefore should have been referenced in the final aggregate assessment for chlorpyrifos or the OP CRA. Petitioners do not otherwise explain whether and how these data support revocation of tolerances or cancellation of pesticide registrations.

b. Agency Response

As explained in the previous section, the basis for seeking revocation of a tolerance is a showing that the pesticide is not "safe." Claiming that EPA failed to reference certain data in its risk assessment regarding carcinogenicity does not amount to illustrating that the tolerances are unsafe. To show a lack of safety, petitioners would have to present some fact-based argument demonstrating that aggregate exposure to chlorpyrifos poses an unsafe carcinogenic risk. Petitioners have not presented such an analysis. Accordingly, when EPA publishes its response to the petition in the Federal Register, EPA intends to deny the petitioners' request to revoke chlorpyrifos tolerances or cancel chlorpyrifos registrations to the extent that request relies on claims pertaining to carcinogenicity.

Despite the inadequacy of petitioners' cancer claims, in the course of the Agency's review of chlorpyrifos, EPA has examined the Lee et al. study cited by petitioners,[50] among other lines of evidence. EPA has concluded that the Lee et al. investigation does not alter the Agency's weight of evidence determination concerning chlorpyrifos' carcinogenic potential, and therefore does not alter the Agency's current cancer classification for chlorpyrifos. Specifically, the Agency does not believe this evidence raises sufficient grounds for concern regarding chlorpyrifos that EPA should consider initiating action based upon this information that might lead to revocation of the chlorpyrifos tolerances or cancellation of the chlorpyrifos registrations.

The Agency was aware of the December 2004 study cited by petitioners. While Lee et al. observed a possible association between chlorpyrifos use and the incidence of lung cancer, the authors also stressed that further evaluation was necessary before concluding the association was causal in nature.[51] Additional evaluation is necessary because of possible alternative explanations for the Lee et al. study, which include unmeasured confounding factors or confounding factors not fully accounted for in the analysis, and possible false positive results due to the performance of multiple statistical tests.

EPA has been a collaborating agency with the AHS since 1993, and continues to closely monitor the AHS literature. The Agency is working closely with the AHS researchers to clearly understand the results of their research efforts to ensure the Agency appropriately interprets these data as future studies are published. Between 2003 and 2009 there have been six nested case-control analyses within the AHS which evaluated the use of a number of agricultural pesticides, including chlorpyrifos, in association with specific anatomical cancer sites, in addition to the previously published cohort study[52] cited by the petitioners. As noted below, both the Agency and Health Canada have comprehensively reviewed these data. Further, the Agency has

[50] U.S. EPA (2011). Christenson Memorandum "Chlorpyrifos Carcinogenicity: Review of Evidence from the U.S. Agricultural Health Study (AHS) Epidemiologic Evaluations 2003-2009. (hereinafter Christenson 2011).
[51] Lee et al. 2004 at 1788.
[52] Lee et al 2004.

proposed a draft framework[53] to consider epidemiologic information in the risk assessment process, and additionally utilized AHS data in a case study illustrating the similarities and differences in exposure assessment methodology between epidemiologic research and regulatory risk assessment.

In accordance with the Agency's 2005 Guideline for Cancer Risk Assessment,[54] chlorpyrifos is classified as "Not Likely to be Carcinogenic to Humans" based on the lack of evidence of carcinogenicity in male or female mice and male or female rats. In chronic toxicity/ carcinogenicity studies, animals received chlorpyrifos in their feed every day of their lives (78 weeks for mice and 104 weeks for rats) at doses thousands of times greater than any anticipated exposure to humans from authorized uses. There was no evidence of cancer in the experimental animal studies. Additionally, available evidence from *in vivo* and *in vitro* assays did not support a mutagenic or genotoxic potential of chlorpyrifos.

Recently, the Agency conducted its own review of the six nested case-control analyses and one cohort study within the AHS concerning the carcinogenic potential of chlorpyrifos.[55] EPA concluded with respect to the AHS lung cancer results that the findings are useful for generating hypotheses, but require confirmation in future studies. This conclusion is consistent with that of researchers from Health Canada. Specifically, Weichenthal et al. (2010)[56] recently published a review article in Environmental Health Perspectives on pesticide exposure and cancer incidence in the AHS cohort. Their review of these same studies concluded that the weight of experimental toxicological evidence does not suggest that chlorpyrifos is carcinogenic, and that epidemiologic results currently available from the AHS are inconsistent, lack replication, and lack a coherent biologically plausible carcinogenic mode of action. The authors did note positive exposure-response associations for chlorpyrifos and lung cancer in two separate evaluations. The Agency will continue to review additional AHS data as well as other epidemiologic evaluations during the development of the HHRA.

In summary, while there is initial suggestive epidemiological evidence of an association between chlorpyrifos and lung cancer to only form a hypothesis as to a carcinogenic mode of action, additional research (including follow-up AHS research) is needed to test the hypothesis. Consequently, at this time it is reasonable to conclude chlorpyrifos is not a carcinogen in view of the lack of carcinogenicity in the rodent bioassays and the lack of a genotoxic or mutagenic potential. The Agency concludes that existing epidemiological data (including Lee et al.) do not change the current weight of the evidence conclusions. The Agency continues to believe there is not a sufficient basis to alter its assessment of chlorpyrifos as not likely to be carcinogenic to humans when multiple lines of evidence are considered (e.g., epidemiology findings, rodent bioassay, genotoxicity); therefore, chlorpyrifos cancer risk would not be a factor in any potential Agency risk determination to revoke tolerances for chlorpyrifos.

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[53] U.S. EPA Presentation to the FIFRA Science Advisory Panel, February 2010. Avaliable at http://www.epa.gov/scipoly/sap/meetings/2010/020210meeting.html#materials.
[54] U.S. EPA (2005). Guidelines for Carcinogen Risk Assessment. Available at http://www.epa.gov/raf/publications/pdfs/CANCER_GUIDELINES_FINAL_3-25-05.PDF.
[55] Christenson 2011.
[56] Weichenthal S, Moase C, Chan P (2010). A review of pesticide exposure and cancer incidence in the agricultural health study cohort. Cien Saude Colet. 2012 Jan;17(1):255-70. PubMed PMID: 22218559.

4. CRA misrepresents risks, failed to apply FQPA10X Safety Factor

a. Petitioners' claim

Petitioners assert that EPA relied on limited data and inaccurate interpretations of data to support its decision to remove the FQPA safety factor in the CRA.[57] Specifically, the petitioners challenge the Agency's use of data from a paper by Zheng et al. (2000)[58] claiming that, in contrast to the Agency's analysis of the study data, the data does show an obvious difference between juvenile and adult responses to chlorpyrifos.[59] Petitioners conclude by asserting that the Zheng et al. study supports using a 10X safety factor for chlorpyrifos in the CRA.

b. Agency Response

Petitioners' assertions do not provide a sufficient basis for revoking chlorpyrifos tolerances. As explained previously, the ground for seeking revocation of a tolerance is a showing that the pesticide is not "safe." The petitioners' claim that the data EPA relied upon support a different FQPA safety factor for chlorpyrifos in the CRA does not amount to a showing that chlorpyrifos tolerances are unsafe. To show a lack of safety, petitioners would have to present a factual analysis demonstrating that the lack of a 10X safety factor in the CRA for chlorpyrifos poses unsafe cumulative exposures to the OP pesticides. Petitioners have not made such a showing. For this reason, when EPA publishes its response to the petition in the Federal Register, EPA intends to deny the petitioners' request to revoke chlorpyrifos tolerances or cancel chlorpyrifos registrations to the extent that request relies on claims pertaining to EPA's failure to provide a 10X safety factor in the OP CRA based on the results of the Zheng et al. study.

Despite the inadequacy of petitioners' FQPA safety factor claims, EPA has examined the evidence cited by petitioners for the purpose of evaluating whether the evidence raises sufficient grounds for concern regarding chlorpyrifos that EPA should consider initiating action that might lead to revocation of the chlorpyrifos tolerances.

In general, when the Agency conducts a cumulative assessment, the scope of cumulative risk is limited to the common mechanism endpoint -- which in this case is cholinesterase inhibition, the primary toxicity mode of action and the most sensitive, quantifiable endpoint for the OP pesticides. As such, for the OP CRA, experimental toxicology data on AChE inhibition are used for developing relative potency estimates, PoDs, and informing the FQPA safety factor. EPA has relied on brain AChE data from adult female rats dosed for 21 days or longer for estimating relative potency and PoDs. At approximately three weeks of oral exposure to OP pesticides, AChE inhibition reaches steady state in the adult rat such that continued dosing does not result in increased inhibition. This timeframe of toxicity (21-days and longer) was selected as there was high confidence in the potency estimates derived from the steady state toxicology studies due to the stability of the AChE inhibition.

[57] Petition at 16.
[58] Zheng Q, Olivier K, Won YK, Pope CN. 2000. Comparative cholinergic neurotoxicity of oral chlorpyrifos exposures in preweaning and adult rats. Toxicological Sciences, 55(1): 124-132.
[59] Petition at 14.

The Agency's 2006 OP CRA contains EPA's complete FQPA safety factor analysis,[60] which involved consideration of pre-natal and post-natal experimental toxicology studies, in addition to exposure information. In the OP CRA, pre-natal exposure AChE studies in rats show that the fetus is no more sensitive than the dam to AChE inhibition and the fetus is often less sensitive than the dam. Thus, evaluating the potential for increased toxicity of juveniles from post-natal exposure was a key component in determining the magnitude of the FQPA safety factors in the OP CRA. Furthermore, because characteristics of children are directly accounted for in the cumulative exposure assessment, the Agency's methods are not expected to underestimate exposure to OP pesticides.

In the CRA, each OP pesticide was assigned a 10X FQPA safety factor unless chemical-specific AChE data on young animals were available to generate a data derived safety factor. To best match the relative potency factor and PODs based on repeated dosing, the Agency used repeated dosing data in juveniles for developing the FQPA safety factors. For chlorpyrifos, at the time of the 2006 OP CRA, the only such data available were from the Zheng et al. literature study.

The petitioners are correct that Dr. Carey Pope of Oklahoma State University, who is also a member of the FIFRA SAP, provided the Agency with the raw data from the Zheng et al. study. These raw data were used to develop the plot in the 2006 OP CRA which was reproduced in the petition. Petitioners accurately note that for other OP pesticides a benchmark dose (BMD) modeling approach was used and that no BMD values were reported for chlorpyrifos. In determining the FQPA safety factor, petitioners claim that the Agency misinterpreted the brain AChE data from Zheng et al.

As shown in the plot reproduced on page 15 of the petition, the dose-response data in the Zheng et al. study are variable and lack a monotonic shape at the low dose end of the dose response curve. The Agency acknowledges that at the high dose, the pups appear to be more sensitive. However, at the low dose end of the response curve, relevant for human exposures and, thus, the cumulative risk assessment (i.e., at or near the 10% inhibition level), little to no difference is observed. Therefore, despite the lack of BMD estimates for the Zheng et al. study, in 2006 the Agency was confident in the value used. Since that time, the Agency attempted BMD modeling of the Zheng et al. data as part of the 2011 preliminary chlorpyrifos HHRA[61] which yielded low confidence results due to the variability in the data

Dow AgroSciences recently submitted a new comparative cholinesterase study (CCA) for chlorpyrifos. CCA studies are specially designed studies to compare the dose-response relationship in juvenile and adult rats. This CCA study includes two components: 1) acute, single dosing in post-natal day (PND) 11 and young adult rats and 2) 11-days of repeating dosing in rat pups from PND11-21 and 11-days of repeated dosing in adult rats. The CCA study for chlorpyrifos is considered by EPA to be high quality and well-designed. The preliminary risk assessment for chlorpyrifos' reports BMD estimates from this CCA study. Specifically for the

[60] Available at http://epa.gov/pesticides/cumulative/2006-op/op_cra_main.pdf.
[61] U.S. EPA (2011). Chlorpyrifos: Preliminary Human Health Risk Assessment for Registration. Available in docket number EPA-HQ-OPP-2008-0850, http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0025.

repeated dosing portion of the study, the BMD_{10s} of 0.80 (0.69 $BMDL_{10}$) and 1.0 (0.95 $BMDL_{10}$) mg/kg/day respectively for female pups and adults results support the FQPA safety factor of 1X used in the 2006 OP CRA. Therefore, the Agency remains confident in the FQPA safety factor of 1X used in the cumulative risk assessment for chlorpyrifos. As such, petitioners' claims regarding the OP CRA and FQPA safety factor, at this time would not be a factor in a determination by the Agency to revoke tolerances for chlorpyrifos.

5. Over-reliance on registrant data

a. Petitioners' Claim

Petitioners assert that EPA "cherry picked" data, "ignoring robust, peer-reviewed data in favor of weak, industry-sponsored data to determine that chlorpyrifos could be re-registered and food tolerances be retained."[62] As such, the Agency's reassessment decision is not scientifically defensible.[63]

b. Agency Response

This portion of the petition does not purport to be an independent basis for revoking chlorpyrifos tolerances or cancelling chlorpyrifos registrations. Rather, this claim appears to underlie petitioners' arguments in other sections of the petition. While petitioners claim that EPA ignored robust, peer-reviewed data in favor of weak, industry-sponsored data for the reregistration of chlorpyrifos, petitioners do not cite to any studies other than those used to support their other claims. In general, petitioners did not provide any studies in their petition that EPA failed to evaluate. Since the specific studies cited by petitioners are not associated with this claim, but rather their other claims, EPA's response to the specific studies are, therefore, addressed in its responses to petitioners' other claims. However, EPA explains below why, as a general matter, the Agency does not believe it has "over-relied" on registrant data in evaluating the risks of chlorpyrifos or other pesticides.

In spite of petitioners' claim, the Agency does not ignore robust, peer-reviewed data in favor of industry-sponsored data. Further, EPA has a very public and well-documented set of procedures that it applies to the use and significance accorded all data utilized to inform risk management decisions. Registrant generated data, in response to FIFRA and FFDCA requirements, are conducted and evaluated in accordance with a series of internationally harmonized and scientifically peer-reviewed study protocols designed to maintain a high standard of scientific quality and reproducibility.[64]

Additionally, to further inform the Agency's risk assessment, EPA is committed to the consideration of other sources of information such as data identified in the open, peer-reviewed literature and information submitted by the public as part of the regulatory evaluation of a

[62] Petition at 16.
[63] *Id.*
[64] See http://www.epa.gov/opp00001/science/guidelines.htm for information on EPA's Harmonized Test Guidelines and international efforts at harmonization.

pesticide. An important issue, when evaluating any study, is its scientific soundness and quality, and thus, the level of confidence in the study findings to contribute to the risk assessment.

The literature was searched, fully considered, and provided additional information on, chlorpyrifos mode of action, pharmacokinetics, epidemiology, neurobehavioral effects in laboratory animals, and age dependent sensitivity to cholinesterase inhibition. This information is discussed in the 2008 chlorpyrifos SAP paper and the chlorpyrifos 2011 preliminary HHRA.

Therefore, by evaluating registrant data in accordance with internationally harmonized and scientifically peer-reviewed study protocols, undertaking thorough open literature searches, and considering information provided by the public, the Agency is confident that its assessment for chlorpyrifos was reasonably based upon the best available science at the time of the assessment. Previous sections of this response to petitioners' claims regarding the Agency's inadequate use of various data only further highlights and supports the scientifically defensible results of the Agency's assessment. Petitioners' claim that the Agency overly relies on registrant data is unfounded and not supported by the record and as such, it would form no basis of the Agency's decision to revoke chlorpyrifos tolerances or cancel chlorpyrifos registrations.

6. *Export Hazard*

a. Petitioners' claim

Petitioners assert that EPA must ban chlorpyrifos and cancel all tolerances because, otherwise, chlorpyrifos will continue to be used unsafely by workers, including children, in other countries who may not utilize worker protection equipment required for use in the U.S.[65] In addition, petitioners assert that continued chlorpyrifos use internationally presents a health hazard from contaminated food re-entering the United States.[66]

b. Agency Response.

The Agency takes very seriously its leadership role and commitment to international efforts to promote the safe use of pesticides. EPA's principal goal in international pesticide activities is to improve the protection of public health and the environment throughout the world.[67] Under FIFRA and FFDCA, however, EPA's primary focus in regulating pesticides is to address risk from domestic use of pesticides and from pesticide residues on imported food. It is far from clear that EPA has any authority under FIFRA to address the risks to foreign workers

[65] Petition at 21.
[66] *Id.*
[67] EPA actively participates in Codex, which is a joint food standards program of the United Nations Food and Agriculture Organization (FAO) and the World Health Organization (WHO). Codex develops international food safety and quality standards, including Maximum Residue Limits (MRLs) for pesticides. EPA contributes technical expertise to the development of these standards and related policies. Additional information can be found at http://www.codexalimentarius.net/web/index_en.jsp. EPA is also active in the Organization for Economic Co-operation and Development (OECD) Working Group on Pesticides. The objective of the OECD Pesticide Program is to help governments co-operate in assessing and reducing the risks of agricultural pesticides. Additional information on the OECD pesticide program can be found at http://www.oecd.org/document/34/0,3746,en_2649_37465_48447010_1_1_1_37465,00.html.

who may not be utilizing the protective equipment that EPA requires when a pesticide product is used within EPA's jurisdiction. Further, to the extent that EPA could in some limited fashion take international considerations into account under its authorities, it is extremely difficult to predict what the effects would be internationally from a U.S. ban on chlorpyrifos use because a U.S. ban does not legally limit either the export of cancelled pesticides from the U.S. or the manner of their use overseas. Section 17 of FIFRA (7 U.S.C. 136o), and EPA regulations at 40 C.F.R. Part 168 Subpart D have specific requirements that apply to the export of unregistered pesticides, but these provisions do not provide EPA with authority to ban the export of cancelled pesticides.

In addition to concerns about international workers, petitioners claim that if the Agency does not revoke all tolerances for chlorpyrifos, contaminated food will enter the U.S. The FFDCA specifically requires EPA to establish maximum permissible residue levels (tolerances) for pesticides only when such residues have been deemed "safe' within the meaning of section 408(b)(2)(A)(ii) (21 U.S.C. 346a(b)(2)(A)(ii). These domestic tolerances also serve as tolerances for food imported into the U.S. The risk from the domestic use of chlorpyrifos and from residues of chlorpyrifos on imported food will be addressed by the Agency's final response to the petition and EPA's final HHRA. If EPA determines in that process that there is an unsafe risk from chlorpyrifos in food, the Agency will revoke tolerances as necessary. This action would obviously limit chlorpyrifos' use overseas on crops bound for the U.S. and, thus, would likely limit chlorpyrifos use internationally to some extent. But again, EPA has no authority to ban the export of cancelled pesticides.

In conclusion, there is no substantive information in petitioners' export hazard assertion that provides a basis for the cancellation of all chlorpyrifos registrations or the revocation of all chlorpyrifos tolerances.

V. Conclusion

The Agency has carefully considered the six petition claims addressed in this response and has determined that none of these claims warrants revoking tolerances or canceling registrations for chlorpyrifos at this time. This response does not constitute a final Agency action to petitioners' request to revoke all tolerances for chlorpyrifos.

This response does, however, constitute EPA's final action on the petition's sole FIFRA issue, exporting hazard. As such, I hereby deny petitioners' claim to cancel all chlorpyrifos registrations based upon the exporting hazard claim.

Steven P. Bradbury, Ph.D.
Director, Office of Pesticide Programs

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ATTACHMENT K

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

JUL 16 2012

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

Spray Drift Mitigation Decision for Chlorpyrifos (059101)
July 2012

Summary

This document discusses mitigation measures that the chlorpyrifos technical registrants Dow AgroSciences, AAKO B.V., Cheminova Inc., Drexel Chemical Company, Gharda Chemicals Ltd, and Makhteshim Chemical Works Ltd. (registrants) have voluntarily agreed to implement in order to mitigate risks to children and bystanders from spray drift that occurs during agricultural applications of chlorpyrifos. To ensure timely implementation of the spray drift mitigation, EPA is taking steps to make sure that the new use restrictions appear on all chlorpyrifos agricultural product labels starting in late 2012.

The new mitigation measures require buffer zones for groundboom, airblast, and aerial application methods around sensitive sites such as residential lawns, homes, pedestrian sidewalks, outdoor recreational areas, and all property associated with buildings typically occupied by people. In addition, rates for aerial application are being reduced from 6 lbs ai/A to 2.0 lbs ai/A.[1]

This effort to mitigate a chemical's risks early in the registration review process is consistent with the Agency's approach for registration review. Where risks are identified early in the registration review process and opportunities for early mitigation exist, the Agency will pursue those opportunities as they arise, rather then waiting for completion of a chemical's registration review in order to mitigate risks. Potential risks to children and bystanders due to spray drift from chlorpyrifos applications are an area where the opportunity for early mitigation existed.

Background

Chlorpyrifos is used widely for controlling insects on food crops including fruits, nuts, vegetables, and grains, and on non-food sites such as golf course turf, industrial sites, greenhouses, nurseries, sod farms, and wood products. Public health uses include aerial and ground-based fogger treatments to control adult mosquitoes. An organophosphate, chlorpyrifos can cause cholinesterase inhibition in humans; i.e., it can over-stimulate the nervous system if there is sufficient exposure.

Chlorpyrifos is currently undergoing registration review, EPA's periodic reevaluation of all registered pesticides to ensure that they continue to meet the statutory standard of no

[1] The lone exception is up to a 2.3 lbs ai/A rate to control Asian Citrus Psylla (ACP). Chlorpyrifos is one of the few options available for protecting mature bearing trees from ACP – currently FL is the only state with a 2.3 lb ai/A for ACP.

unreasonable adverse effects. As part of registration review, the chlorpyrifos preliminary Human Health Risk Assessment (HHRA) was released for public comment in July 2011.[2] In the preliminary HHRA, risks to bystanders from spray drift and exposure from volatilization were identified as concerns. The public comment period closed in October 2011. As the Agency works to finalize its HHRA, it has further refined its analysis regarding spray drift from various chlorpyrifos application scenarios in order to have a broader understanding of the potential risks. That assessment, Evaluation of the Potential Risks from Spray Drift and the Impact of Potential Risk Reduction Measures[3], is being released in conjunction with this decision document.

In addition, in 2007, the Natural Resources Defense Council (NRDC) and Pesticide Action Network North America (PANNA) jointly petitioned the Agency to revoke all tolerances and cancel all registrations for chlorpyrifos.[4] One of the issues identified in the Petition deals with inhalation routes of exposure from spray drift and volatilization. This more refined spray drift assessment and subsequent mitigation measures will inform the Agency's response to the spray drift portion of petitioners' inhalation claim. The Agency's volatilization assessment continues to be refined in the context of the final HHRA.

This more refined spray drift analysis resulted in a better estimate of potential exposures and risks to bystanders, particularly children, around treated fields. While the analysis showed there were health risks due to spray drift, the analysis also indicated that the risks could be mitigated by requiring buffer distances and specific application methods. Specifically, by linking droplet size with application rates and application methods in order to dictate appropriate buffer distances. Table 1 indicates the various buffer distances that will be required when using certain application rates, nozzle droplet type, and application method.

Table 1: Buffer Distances from Sensitive Sites

Application rate (lb ai/A)	Nozzle Droplet Type	Required Setback (Buffer Zones) (feet)		
		Aerial	Airblast	Ground
>0.5 - 1	coarse or very coarse	10	10	10
>0.5 - 1	medium	25	10	10
>1 - 2	coarse or very coarse	50	10	10
>1 - 2	medium	80	10	10
>2 - 3	coarse or very coarse	80[1]	10	10
>2 - 3	medium	100[1]	10	10
>3 - 4	medium or coarse	NA[2]	25	10
>4	medium or coarse	NA	50	10

[1]Aerial application of greater than 2 lb ai/A is only permitted for Asian Citrus Psylla control, up to 2.3 lb ai/A.
[2]NA is not allowed.

[2] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0025.
[3] Dawson J, Britton W, Bohaty R, Mallampalli N, Grube A. U.S. EPA (2012). Chlorpyrifos, PC Code 059101, DP Barcode 399483 and 399485; Evaluation of the Potential Risks From Spray Drift and the Impact of Potential Risk Reduction Measures. Available at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2008-0850.
[4] Petition of Natural Resources Defense Council and Pesticide Action Network North America to Revoke All Tolerances and Cancel All Registrations for the Pesticide Chlorpyrifos (September 12, 2007) at 1. (hereinafter Petition). Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0005.

As Table 2 demonstrates, theses new buffer distance requirements presented in Table 1 will address risk concerns regarding bystanders' margin of exposure (MOE).[5]

Table 2: Target MOEs > 100, Dermal

Application method	Lowest MOE Pre-Mitigation	Lowest MOE Post-Mitigation
Aerial	10	104
Airblast	57	132
Groundboom	10	143

In addition, the mitigation measures will provide greater specificity on chlorpyrifos labels regarding what constitutes a sensitive site. The new sensitive site language is the following

> *Sensitive sites are areas frequented by non-occupational bystanders (especially children). These include residential lawns, pedestrian sidewalks, outdoor recreational areas such as school grounds, athletic fields, parks and all property associated with buildings occupied by humans for residential or commercial purposes. Sensitive sites include homes, farmworker housing, or other residential buildings, schools, daycare centers, nursing homes, and hospitals. Non-residential agricultural buildings, including barns, livestock facilities, sheds, and outhouses are not included in this prohibition.*

For the complete spray drift analysis, refer to the Evaluation of the Potential Risks from Spray Drift and the Impact of Potential Risk Reduction Measures, itself. This and all other documents related to chlorpyrifos registration review are located at http://www.regulations.gov under docket number EPA-OPP-2008-0850. Documents related to the Petition are located in docket number EPA-OPP-2007-1005.

Conclusion

This document presents the mitigation measures being taken voluntarily by registrants to address the current risks to bystanders, particularly children from spray drift that occurs during agricultural applications of chlorpyrifos.

Richard P. Keigwin, Jr.
Director
Pesticide Re-evaluation Division

[5] The ratio of the estimated NOAEL to the exposure is referred to as the Margin of Exposure (MOE). Generally, MOEs that are less than 100 exceed the Agency's level of concern for worker risk.

EXHIBIT 2

PESTICIDES *in the* DIETS OF INFANTS AND CHILDREN

NATIONAL RESEARCH COUNCIL

PESTICIDES *in the* DIETS OF INFANTS AND CHILDREN

Committee on Pesticides in the Diets of
Infants and Children

Board on Agriculture
and
Board on Environmental Studies and Toxicology

Commission on Life Sciences

National Research Council

NATIONAL ACADEMY PRESS
Washington, D.C. 1993

Contents

Executive Summary

PESTICIDES ARE USED WIDELY in agriculture in the United States. Their application has improved crop yields and has increased the quantity of fresh fruits and vegetables in the diet, thereby contributing to improvements in public health.

But pesticides may also cause harm. Some can damage the environment and accumulate in ecosystems. And depending on dose, some pesticides can cause a range of adverse effects on human health, including cancer, acute and chronic injury to the nervous system, lung damage, reproductive dysfunction, and possibly dysfunction of the endocrine and immune systems.

Diet is an important source of exposure to pesticides. The trace quantities of pesticides that are present on or in foodstuffs are termed residues. To minimize exposure of the general population to pesticide residues in food, the U.S. Government has instituted regulatory controls on pesticide use. These are intended to limit exposures to residues while ensuring an abundant and nutritious food supply. The legislative framework for these controls was established by the Congress through the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and the Federal Food, Drug, and Cosmetic Act (FFDCA). Pesticides are defined broadly in this context to include insecticides, herbicides, and fungicides.

Tolerances constitute the single, most important mechanism by which EPA limits levels of pesticide residues in foods. A tolerance is defined as the legal limit of a pesticide residue allowed in or on a raw agricultural commodity and, in appropriate cases, on processed foods. A tolerance must be established for any pesticide used on any food crop.

Tolerance concentrations are based primarily on the results of field trials conducted by pesticide manufacturers and are designed to reflect the highest residue concentrations likely under normal conditions of agricultural use. Their principal purpose is to ensure compliance with good agricultural practice. Tolerances are not based primarily on health considerations.

This report addresses the question of whether current regulatory approaches for controlling pesticide residues in foods adequately protect infants and children. The exposure of infants and children and their susceptibility to harm from ingesting pesticide residues may differ from that of adults. The current regulatory system does not, however, specifically consider infants and children. It does not examine the wide range of pesticide exposure patterns that appear to exist within the U.S. population. It looks only at the average exposure of the entire population. As a consequence, variations in dietary exposure to pesticides and health risks related to age and to such other factors as geographic region and ethnicity are not addressed in current regulatory practice.

Concern about the potential vulnerability of infants and children to dietary pesticides led to U.S. Congress in 1988 to request that the National Academy of Sciences (NAS) appoint a committee to study this issue through its National Research Council (NRC). In response, the NRC appointed a Committee on Pesticide Residues in the Diets of Infants and Children under the joint aegis of the Board on Agriculture and the Board on Environmental Studies and Toxicology.

The committee was charged with responsibility for examining scientific and policy issues faced by government agencies, particularly EPA, in regulating pesticide residues in foods consumed by infants and children. Specifically, the committee was asked to examine the adequacy of current risk assessment policies and methods; to assess information on the dietary intakes of infants and children; to evaluate data on pesticide residues in the food supply; to identify toxicological issues of greatest concern; and to develop relevant research priorities. Expertise represented on the committee included toxicology, epidemiology, biostatistics, food science and nutrition, analytical chemistry, child growth and development, and pediatrics.

The committee was not asked to consider toxicities resulting from exposures to microorganisms (bacteria and viruses) or from other naturally occurring potential toxins. It was not asked to weigh the benefits and risks to be derived from a plentiful and varied food supply against the potential risks resulting from pesticide exposure. It was not asked to assess the overall safety of the food supply.

In this report, the committee considered the development of children from the beginning of the last trimester of pregnancy (26 weeks) through

18 years of age, the point when all biological systems have essentially matured.

CONCLUSIONS

Age-Related Variation in Susceptibility and Toxicity

A fundamental maxim of pediatric medicine is that children are not "little adults." Profound differences exist between children and adults. Infants and children are growing and developing. Their metabolic rates are more rapid than those of adults. There are differences in their ability to activate, detoxify, and excrete xenobiotic compounds. All these differences can affect the toxicity of pesticides in infants and children, and for these reasons the toxicity of pesticides is frequently different in children and adults. Children may be more sensitive or less sensitive than adults, depending on the pesticide to which they are exposed. Moreover, because these processes can change rapidly and can counteract one another, there is no simple way to predict the kinetics and sensitivity to chemical compounds in infants and children from data derived entirely from adult humans or from toxicity testing in adult or adolescent animals.

The committee found both quantitative and occasionally qualitative differences in toxicity of pesticides between children and adults . Qualitative differences in toxicity are the consequence of exposures during special windows of vulnerability—brief periods early in development when exposure to a toxicant can permanently alter the structure or function of an organ system. Classic examples include chloramphenicol exposure of newborns and vascular collapse (gray baby syndrome), tetracycline and dysplasia of the dental enamel, and lead and altered neurologic development.

Quantitative differences in pesticide toxicity between children and adults are due in part to age-related differences in absorption, metabolism, detoxification, and excretion of xenobiotic compounds, that is, to differences in both pharmacokinetic and pharmacodynamic processes. Differences in size, immaturity of biochemical and physiological functions in major body systems, and variation in body composition (water, fat, protein, and mineral content) all can influence the extent of toxicity. Because newborns are the group most different anatomically and physiologically from adults, they may exhibit the most pronounced quantitative differences in sensitivity to pesticides. **The committee found that quantitative differences in toxicity between children and adults are usually less than a factor of approximately 10-fold**.

The committee concluded that the mechanism of action of a toxicant—how it causes harm—is generally similar in most species and across age

and developmental stages within species. For example, if a substance is cytotoxic in adults, it is usually also cytotoxic in immature individuals.

Lack of data on pesticide toxicity in developing organisms was a recurrent problem encountered by the committee. In particular, little work has been done to identify effects that develop after a long latent period or to investigate the effects of pesticide exposure on neurotoxic, immunotoxic, or endocrine responses in infants and children. The committee therefore had to rely mostly on incomplete information derived from studies in mature animals and on chemicals other than pesticides.

The committee reviewed current EPA requirements for toxicity testing by pesticide manufacturers, as well as testing modifications proposed by the agency. In general, the committee found that current and past studies conducted by pesticide manufacturers are designed primarily to assess pesticide toxicity in sexually mature animals. Only a minority of testing protocols have supported extrapolation to infant and adolescent animals. Current testing protocols do not, for the most part, adequately address the toxicity and metabolism of pesticides in neonates and adolescent animals or the effects of exposure during early developmental stages and their sequelae in later life.

Age-Related Differences in Exposure

Estimation of the exposures of infants and children to pesticide residues requires information on (1) dietary composition and (2) residue concentrations in and on the food and water consumed. **The committee found that infants and children differ both qualitatively and quantitatively from adults in their exposure to pesticide residues in foods**. Children consume more calories of food per unit of body weight than do adults. But at the same time, infants and children consume far fewer types of foods than do adults. Thus, infants and young children may consume much more of certain foods, especially processed foods, than do adults. And water consumption, both as drinking water and as a food component, is very different between children and adults.

The committee concluded that differences in diet and thus in dietary exposure to pesticide residues account for most of the differences in pesticide-related health risks that were found to exist between children and adults. Differences in exposure were generally a more important source of differences in risk than were age-related differences in toxicologic vulnerability.

Data from various food consumption surveys were made available to the committee. In analyzing these data, the committee found it necessary to create its own computer programs to convert foods as consumed into their component raw agricultural commodities (RACs). This analytic approach

facilitated the use of data from different sources and permitted evaluation of total exposure to pesticides in different food commodities. For processed foods, the committee noted that effects of processing on residue concentrations should be considered, but that information on these effects is quite limited. Processing may decrease or increase pesticide residue concentrations. The limited data available suggest that pesticide residues are generally reduced by processing; however, more research is needed to define the direction and magnitude of the changes for specific pesticide-food combinations. The effect of processing is an important consideration in assessing the dietary exposures of infants and young children, who consume large quantities of processed foods, such as fruit juices, baby food, milk, and infant formula.

Although there are several sources of data on pesticide residues in the United States, the data are of variable quality, and there are wide variations in sample selection, reflecting criteria developed for different sampling purposes, and in analytical procedures, reflecting different laboratory capabilities and different levels of quantification between and within laboratories. These differences reflect variations in precision and in the accuracy of methods used and the different approaches to analytical issues, such as variations in limit of quantification. There also are substantial differences in data reporting. These differences are due in part to different record-keeping requirements, such as whether to identify samples with multiple residues, and differences in statistical treatment of laboratory results below the limit of quantification.

Both government and industry data on residue concentrations in foods reflect the current regulatory emphasis on average adult consumption patterns. The committee found that foods eaten by infants and children are underrepresented in surveys of commodity residues. Many of the available residue data were generated for targeted compliance purposes by the Food and Drug Administration (FDA) to find residue concentrations exceeding the legal tolerances established by the EPA under FFDCA.

Survey data on consumption of particular foods are conventionally grouped by broad age categories. The average consumption of a hypothetical "normal" person is then used to represent the age group. However, in relying solely on the average as a measure of consumption, important information on the distribution of consumption patterns is lost. For example, the high levels of consumption within a particular age group are especially relevant when considering foods that might contain residues capable of causing acute toxic effects. Also, geographic, ethnic, and other differences may be overlooked.

To overcome the problems inherent in the current reliance on "average" exposures, the committee used the technique of statistical convolution (i.e., combining various data bases) to merge distributions of food consumption

with distributions of residue concentrations. This approach permits examination of the full range of pesticide exposures in the U.S. pediatric population. As is described in the next section, this approach provides an improved basis over the approach now used for assessing risks for infants and children.

A New Approach to Risk Assessment for Infants and Children

To properly characterize risk to infants and children from pesticide residues in the diet, information is required on (1) food consumption patterns of infants and children, (2) concentrations of pesticide residues in foods consumed by infants and children, and (3) toxic effects of pesticides, especially effects that may be unique to infants and children. If suitable data on these three items are available, risk assessment methods based on the technique of statistical convolution can be used to estimate the likelihood that infants and children who experience specific exposure patterns may be at risk. To characterize potential risks to infants and children in this fashion, the committee utilized data on distributions of pesticide exposure that, in turn, were based on distributions of food consumption merged with data on the distribution of pesticide residue concentrations. The committee found that age-related differences in exposure patterns for 1– to 5-year-old children were most accurately illuminated by using 1-year age groupings of data on children's food consumption.

Exposure estimates should be constructed differently depending on whether acute or chronic effects are of concern. Average daily ingestion of pesticide residues is an appropriate measure of exposure for assessing the risk of chronic toxicity. However, actual individual daily ingestion is more appropriate for assessing acute toxicity. Because chronic toxicity is often related to long-term average exposure, the average daily dietary exposure to pesticide residues may be used as the basis for risk assessment when the potential for delayed, irreversible chronic toxic effects exists. Because acute toxicity is more often mediated by peak exposures occurring within a short period (e.g., over the course of a day or even during a single eating occasion), individual daily intakes are of interest. Examining the distribution of individual daily intakes within the population of interest reflects day-to-day variation in pesticide ingestion both for specific individuals and among individuals.

Children may be exposed to multiple pesticides with a common toxic effect, and estimates of exposure and of risk could therefore be improved by accounting for these simultaneous exposures. This can be accomplished by assigning toxicity equivalence factors to each of the compounds having a common mechanism of action. Total residue exposure is then estimated

by multiplying the actual level of each pesticide residue by its toxicity equivalence factor and summing the results. This information may be combined with data on consumption to construct a distribution of total exposure to all pesticides having a common mechanism of action. To test this multiple-residue methodology, the committee estimated children's acute health risks resulting from combined exposure to five members of the organophosphate insecticide family. This was accomplished by combining actual food consumption data with data on actual pesticide residue levels.

Through this new analytical procedure, the committee estimated that for some children, total organophosphate exposures may exceed the reference dose. Furthermore, although the data were weak, the committee estimated that for some children exposures could be sufficiently high to produce symptoms of acute organophosphate pesticide poisoning.

Compared to late-in-life exposures, exposures to pesticides early in life can lead to a greater risk of chronic effects that are expressed only after long latency periods have elapsed. Such effects include cancer, neurodevelopmental impairment, and immune dysfunction. The committee developed new risk assessment methods to examine this issue.

Although some risk assessment methods take into account changes in exposure with age, these models are not universally applied in practice. The committee explored the use of newer risk assessment methods that allow for changes in exposure and susceptibility with age. However, the committee found that sufficient data are not currently available to permit wide application of these methods.

RECOMMENDATIONS

On the basis of its findings, the committee recommends that certain changes be made in current regulatory practice. Most importantly, estimates of expected total exposure to pesticide residues should reflect the unique characteristics of the diets of infants and children and should account also for all nondietary intake of pesticides. Estimates of exposure should take into account the fact that not all crops are treated with pesticides that can be legally applied to those crops, and they should consider the effects of food processing and storage. Exposure estimates should recognize that pesticide residues may be present on more than one food commodity consumed by infants and children and that more than one pesticide may be present on one food sample. Lastly, determinations of safe levels of exposure should take into consideration the physiological factors that can place infants and children at greater risk of harm than adults.

- *Tolerances*. Tolerances for pesticide residues on commodities are currently established by the EPA under FIFRA and FFDCA. A tolerance concentration is defined under FFDCA as the maximum quantity of a pesticide residue allowable on a raw agricultural commodity (RAC) (FFDCA, Section 408) and in processed food when the pesticide concentrates during processing (FFDCA Section 409). Tolerance concentrations on RACs are based on the results of field trials conducted by pesticide manufacturers and are designed to reflect the highest residue concentrations likely under normal agricultural practice. More than 8,500 food tolerances for pesticides are currently listed in the Code of Federal Regulations (CFR). Approximately 8,350 of these tolerances are for residues on raw commodities (promulgated under section 408) and about 150 are for residues known to concentrate in processed foods (promulgated under section 409).

The determination of what might be a safe level of residue exposure is made by considering the results of toxicological studies of the pesticide's effects on animals and, when data are available, on humans. Both acute and chronic effects, including cancer, are considered, although acute effects are treated separately. These data are used to establish human exposure guidelines (i.e., a reference dose, RfD) against which one can compare the expected exposure. Exposure is a function of the amount and kind of foods consumed and the amount and identity of the residues in the foods (i.e., Theoretical Maximum Residue Contributions, TMRCs). If the TMRCs exceed the RfD, then anticipated residues are calculated for comparison with the proposed tolerance. The percent of crop acreage treated is also considered. If the anticipated residues exceed the RfD, then the proposed tolerance is rejected, and the manufacturer may recommend a new tolerance level.

Although tolerances establish enforceable legal limits for pesticide residues in food, they are not based primarily on health considerations, and they do not provide a good basis for inference about actual exposures of infants and children to pesticide residues in or on foods.

Tolerances constitute the only tool that EPA has under the law for controlling pesticide residues in food. **To ensure that infants and children are not exposed to unsafe levels of pesticide residues, the committee recommends that EPA modify its decision-making process for setting tolerances so that it is based more on health considerations than on agricultural practices. These changes should incorporate the use of improved estimates of exposure and more relevant toxicology, along with continued consideration of the requirements of agricultural production. As a result, human health considerations would be more fully reflected in tolerance levels. Children should be able to eat a healthful diet**

containing legal residues without encroaching on safety margins. This goal should be kept clear.

- *Toxicity testing*. The committee believes it is essential to develop toxicity testing procedures that specifically evaluate the vulnerability of infants and children. Testing must be performed during the developmental period in appropriate animal models, and the adverse effects that may become evident must be monitored over a lifetime. Of particular importance are tests for neurotoxicity and toxicity to the developing immune and reproductive systems. Extrapolation of toxicity data from adult and adolescent laboratory animals to young humans may be inaccurate. Careful attention to interspecies differences in pharmacokinetics and metabolism of pesticides and the relative ages at which organ systems mature is essential. It is also important to enhance understanding of developmental toxicity, especially in humans, during critical periods of postnatal development, including infancy and puberty.
- *Uncertainty factors*. For toxic effects other than cancer or heritable mutation, uncertainty factors are widely used to establish guidelines for human exposure on the basis of animal testing results. This is often done by dividing the no-observed-effect level (NOEL) found in animal tests by an uncertainty factor of 100-fold. This factor comprises two separate factors of 10-fold each: one allows for uncertainty in extrapolating data from animals to humans; the other accommodates variation within the human population. Although the committee believes that the latter uncertainty factor generally provides adequate protection for infants and children, this population subgroup may be uniquely susceptible to chemical exposures at particularly sensitive stages of development.

At present, to provide added protection during early development, a third uncertainty factor of 10 is applied to the NOEL to develop the RfD. This third 10-fold factor has been applied by the EPA and FDA whenever toxicity studies and metabolic/disposition studies have shown fetal developmental effects.

Because there exist specific periods of vulnerability during postnatal development, the committee recommends that an uncertainty factor up to the 10-fold factor traditionally used by EPA and FDA for fetal developmental toxicity should also be considered when there is evidence of postnatal developmental toxicity and when data from toxicity testing relative to children are incomplete. The committee wishes to emphasize that this is not a new, additional uncertainty factor but, rather, an extended application of a uncertainty factor now routinely used by the agencies for a narrower purpose.

In the absence of data to the contrary, there should be a presumption of greater toxicity to infants and children. To validate this presumption,

the sensitivity of mature and immature individuals should be studied systematically to expand the current limited data base on relative sensitivity.

- *Food consumption data*. The committee recommends that additional data on the food consumption patterns of infants and children be collected within narrow age groups. The available data indicate that infants and children consume much more of certain foods on a body weight basis than do adults. Because higher exposures can lead to higher risks, it is important to have accurate data on food consumption patterns for infants and children. At present, data are derived from relatively small samples and broad age groupings, making it difficult to draw conclusions about the food consumption patterns of infants and children. Because the composition of a child's diet changes dramatically from birth through childhood and adolescence to maturity, "market basket" food consumption surveys should include adequate samples of food consumption by children at 1-year intervals up to age 5, by children between the ages of 5 and 10 years, and by children between 11 and 18 years. Food consumption surveys should be conducted periodically to ascertain changes in consumption patterns over time.
- *Pesticide residue data*. To maximize the utility of pesticide residue data collected by various laboratories, the committee recommends the use of comparable analytical methods and standardized reporting procedures and the establishment of a computerized data base to collate data on pesticide residues generated by different laboratories. Reports on pesticide residue testing should describe the food commodity analyzed (whether processed or raw), the analytical methods used, the compounds for which tests were conducted, quality assurance and control procedures, and the limit of quantification of the tests. All findings should be reported, whether or not the residue sought is found.

  – In its surveillance of pesticide residues, FDA should increase the frequency of sampling of the commodities most likely to be consumed by infants and children. The residue testing program should include all toxic forms of the pesticide, for example, its metabolites and degradation products.
  – Food residue monitoring should target a special "market basket" survey focused toward the diets of infants and children.
  – Pesticide field trials currently conducted by pesticide manufacturers in support of registration provide data on variation in residue concentrations associated with different rates and methods of application. Such data should be consulted to provide a basis for estimating potential maximum residue levels.

- More complete information is needed on the effects of food processing on levels of pesticides—both the parent compound and its metabolites—in specific food-chemical combinations potentially present in the diets of infants and children.

- *Risk assessment*. All exposures to pesticides—dietary and nondietary—need to be considered when evaluating the potential risks to infants and children. Nondietary environmental sources of exposure include air, dirt, indoor surfaces, lawns, and pets.

- Estimates of total dietary exposure should be refined to consider intake of multiple pesticides with a common toxic effect. Converting residues for each pesticide with a common mechanism of action to toxicity equivalence factors for one of the compounds would provide one approach to estimating total residue levels in toxicologically equivalent units.
- Consumption of pesticide residues in water is an important potential route of exposure. Risk assessment should include estimates of exposure to pesticides in drinking water and in water as a component of processed foods.

Given adequate data on food consumption and residues, the committee recommends the use of exposure distributions rather than single point data to characterize the likelihood of exposure to different concentrations of pesticide residues. The distribution of average daily exposure of individuals in the population of interest is most relevant for use in chronic toxicity risk assessment, and the distribution of individual daily intakes is recommended for evaluating acute toxicity. Ultimately, the collection of suitable data on the distribution of exposures to pesticides will permit an assessment of the proportion of the population that may be at risk.

Although the committee considers the use of exposure distributions to be more informative than point estimates of typical exposures, the data available to the committee did not always permit the distribution of exposures to be well characterized. Existing food consumption surveys generally involve relatively small numbers of infants and children, and food consumption data are collected for only a few days for each individual surveyed. Depending on the purpose for which they were originally collected, residue data may not reflect the actual distribution of pesticide residues in the food supply. Since residue data are not developed and reported in a consistent fashion, it is generally not possible to pool data sets derived from different surveys. Consequently, the committee recommends that guidelines be developed for consumption and residue data permitting characterization of distributions of dietary exposure to pesticides.

The committee identified important differences in susceptibility to the toxic effects of pesticides and exposure to pesticides in the diet with age.

For carcinogenic effects, the committee proposed new methods of cancer risk assessment designed to take such differences into account. Preliminary analyses conducted by the committee suggest that consideration of such differences can lead to lifetime estimates of cancer risk that can be higher or lower than estimates derived with methods based on constant exposure. However, underestimation of risk assuming constant exposure was limited to a factor of about 3- to 5-fold in all cases considered by the committee. Because these results are based on limited data and specific assumptions about the mechanisms by which carcinogenic effects are induced, the applicability of these conclusions under other conditions should be established.

Currently, most long-term laboratory studies of carcinogenesis and other chronic end points are based on protocols in which the level of exposure is held constant during the course of the study. To facilitate the application of risk assessment methods that allow for changes in exposure and susceptibility with age, it would be desirable to develop bioassay protocols that provide direct information on the relative contribution of exposures at different ages to lifetime risks. Although the committee does consider it necessary to develop special bioassay protocols for mandatory application in the regulation of pesticides, it would be useful to design special studies to provide information on the relative effects of exposures at different ages on lifetime cancer and other risks with selected chemical carcinogens.

In addition to pharmacodynamic models for cancer risk assessment, the committee recommends the development and application of physiologically based pharmacokinetic models that describe the unique features of infants and children. For example, differences in relative organ weights with age can be easily described in physiologic pharmacokinetic models; special compartments for the developing fetus may also be incorporated. Physiologically based pharmacokinetic models can be used to predict the dose of the proximate toxicant reaching target tissues, and may lead to more accurate estimates of risk.

In summary, better data on dietary exposure to pesticide residues should be combined with improved information on the potentially harmful effects of pesticides on infants and children. Risk assessment methods that enhance the ability to estimate the magnitude of these effects should be developed, along with appropriate toxicological tests for perinatal and childhood toxicity. The committee's recommendations support the need to improve methods for estimating exposure and for setting tolerances to safeguard the health of infants and children.

7

Estimating Exposures

THE TWO PRECEDING CHAPTERS have reviewed data on the diets of infants and children (Chapter 5) and on pesticide residues in food (Chapter 6). This chapter addresses methods for estimating ingestion of pesticides by infants and children using the data from the preceding two chapters. Although nondietary sources of pesticide exposures such as air, soil, and consumer products are also considered, emphasis is placed on the ingestion of pesticide residues present on foods consumed by infants and children.

Dietary exposure to pesticides depends both on food consumption patterns (Chapter 5) and on residue levels on food (Chapter 6). Multiplying the average consumption of a particular food by the average residue of a particular pesticide on that food yields the average level of ingestion of that pesticide from that one food commodity:

$$\text{Consumption} \times \text{Residue} = \text{Dietary Exposure.}$$

In reality, however, estimation of dietary exposure to pesticides is more complex than this simplified equation. Since many pesticides are used on a number of food crops, determination of the total exposure to a pesticide must be based on consumption data for all such foods. Also, it may be of interest to consider the total ingestion of different pesticides such as organophosphates and carbamates that fall within related classes and may pose similar risks to health.

The data presented in Chapter 5 indicate that food consumption levels vary both among and within individuals. This variation can be represented in terms of a distribution of food consumption, reflecting both high

and low consumption levels, as well as the average level of consumption. Pesticide residue levels present in food will also vary, depending on several variables including application practices in different regions, time that has elapsed since application, degradation during transportation and storage of food, and the manner in which food is prepared by the consumer. Thus, both food consumption and pesticide residue data are characterized not by a single value but, rather, by a broad distribution reflecting high, low, and average values.

The variation in food consumption and residue data produces considerable variation in dietary exposure of pesticides by infants and children. This can be represented by a distribution of exposures across individuals within a particular age group. The distribution of dietary exposures is determined by the distribution of food consumption levels and the distribution of pesticide residues in food. If both the distribution of food consumption and the distribution of residue levels are known, statistical methods can be used to infer the distribution of dietary exposures. The process for combining different distributions into one distribution is termed *convolution*. The statistical convolution methods that can be used for this purpose are discussed later in this chapter.

Since ingestion of pesticides is dependent upon both food consumption and pesticide residue levels in food, it follows that the quality of dietary exposure data is determined by the quality of consumption and residue data. Although food consumption surveys such as the Nationwide Food Consumption Survey (NFCS) provide data on consumption patterns in the population at large, these surveys have generally not targeted infants and children. Hence, they included relatively small sample sizes within the age groups of primary interest for this report. One exception is the 1985-1986 Continuing Surveys of Food Intakes of Individuals (CSFII), which did focus on food consumption patterns of children.

Determination of the distribution of pesticide residues in foods consumed by infants and children is also difficult: only a fraction of all food consumed can be tested for the presence of pesticide residues. Many of the available residue data are based on surveillance studies that because of their focus on potential problem areas may overstate residue levels in the general food supply. The detection limit of residue monitoring methods can also impart uncertainty as to the residue levels actually present on food, especially when many residues are below the limit of detection and the detection limit is relatively high.

Recognizing these data limitations, the committee has included in this chapter several examples to illustrate possible approaches to estimating the distribution of dietary exposure to pesticides for infants and children. Each of these examples is designed to illustrate different aspects of exposure estimation, including the estimation of average daily exposures for use in chronic toxicity risk assessment and the estimation of peak expo-

sures for evaluating acute toxic effects. Examples are included to illustrate how total exposure to pesticides used on more than one food crop can be estimated, and how exposures from different pesticides falling within the same toxicological class can be combined based on their relative toxicity.

Because of the limitations in the available consumption and residue data, it must be stressed that the purpose of the examples is to identify methods for estimating exposure and not to produce representative estimates of actual exposure. The particular compounds chosen for study were selected because sufficient data were available to illustrate the approaches to exposure estimation considered by the committee. All results should be taken in the context of the limitations of the data as described in this and the previous two chapters. Application of these methods in a regulatory context will be possible only if adequate data on the distribution of both food consumption and pesticide residues in food can be obtained.

The first example deals with benomyl, a systemic fungicide that has not been permitted for postharvest use in the United States since 1989. Because of the chronic toxic effects of this compound (benomyl has been shown to cause malignant liver tumors in mice), the average daily ingestion of benomyl was considered to be most relevant for estimating long-term exposure. Note that although the focus is on the average daily ingestion by individuals over an extended period, the daily ingestion will vary from person to person, depending on their food consumption habits and the residues of benomyl in the foods consumed by each person. Since residue data were available for apples, grapes, oranges, peaches, and tomatoes, this example was used to illustrate the estimation of total exposure to a single pesticide from multiple food commodities.

Data on benomyl from different residue monitoring programs were available to the committee, permitting a comparison of exposure estimates based on different residue data. For example, field trial data derived from pesticide analysis in the manufacturer's laboratory (using a special method not adapted to multiresidue screening) usually show higher detection rates than those found by government agencies in random sampling of food shipments. Field trial data are useful only as estimates of maximum residue concentrations from field test plot trials at treatment levels proposed for registration purposes. Because field tests are generally conducted at the maximum pesticide use allowed in its registration, the residue concentrations are often higher than those found in random sampling. The results of field trials are generally used to establish farm tolerances and analytical methodology for purposes of registration. Further evaluation of field trial data is required in order to evaluate pesticide degradation following application.

The impact of residue data below the limit of quantification (LOQ), a

concentration below which residues cannot be accurately measured, was also investigated in this example. For nondetectable residues, it is possible that the actual (unknown) residue could be as low as zero or as high as the LOQ itself. The limitation of data on actual residue concentrations below the LOQ imparts additional uncertainty about the level of exposure to infants and children.

Aldicarb is the subject of the second example. This acutely toxic pesticide exerts its effects by inhibiting cholinesterase enzymes in the nervous system. The example focuses on dietary exposure to aldicarb first from potatoes and bananas separately and then from potatoes and bananas combined. It serves to illustrate how estimates of exposure to a single pesticide found on more than one food can be derived. In contrast to benomyl, where average daily exposures are of interest, individual daily intakes are examined in this example because of the acute toxicity of aldicarb.

Part of the aldicarb residue data is derived from composite sampling, which may underestimate peak residues found in individual potatoes or bananas as a consequence of compositing prior to residue analysis. Composite samples are not very satisfactory in acute risk assessment for raw food commodities like potatoes and bananas. However, residue levels in processed foods can be estimated by using composite samples.

The third example addresses methods for estimating exposure to a class of pesticides inducing a common toxic effect. Specifically, the committee considered five organophosphate compounds used on different fruits and vegetables. All these compounds can inhibit plasma cholinesterase. A measure of total exposure to all five organophosphates is proposed based on their relative potencies.

Before these short examples are presented, there is a discussion of statistical methods for combining the distribution of food consumption with the distribution of residue levels in food to arrive at a distribution of dietary exposures based on the method of convolution. The chapter concludes with a brief summary of nondietary sources of exposure to pesticides.

THE USE OF FOOD CONSUMPTION AND RESIDUE DATA FOR EXPOSURE ASSESSMENT

Food Consumption Data

The most appropriate dietary exposure data for risk assessment depends on the nature of the adverse health effects of concern. In the absence of specific dose-response effects, the average level of exposure of an individual over a certain period provides a reasonable measure on which to

base estimates of chronic toxic effects such as cancer. For acute toxic effects, peak exposures over shorter periods are more appropriate for risk assessment.

Average Levels of Consumption

The development of food consumption data for evaluating chronic toxicity requires careful consideration. In general, food consumption surveys yield data on the consumption of that food over all days for which data are available. The average daily consumption for children within a given age class is then obtained by averaging across all the individuals in the age class.

Estimating the average daily consumption of a particular food within a given class warrants some discussion. Since some foods will not be consumed at all by some individuals, estimates of average daily consumption based on all individuals in the sample will underestimate average consumption for the subpopulation of individuals who consume the food in question. For this reason, separate estimates of average daily consumption for "all children" and for "eaters only" are considered when estimating exposure. Average consumption levels for "eaters only" are typically 2 to 3 times higher than those for "all children."

Because food consumption data are available for only a few days each year, the proportion of children falling into the eaters-only group is underestimated. This problem is accentuated if only a 24-hour recall or 24-hour food record is used. If food consumption data were available for every day of the year, more children who consume the food of interest on an infrequent basis would be included in the eaters-only group. Thus, since the eaters-only group omits some individuals whose consumption levels are low, the average food consumption for "eaters only" calculated in this way actually overestimates the average consumption for this group. This bias does not occur when information on food consumption is obtained through food frequency questionnaires rather than 24-hour recalls or 24-hour food diaries, since food frequency tables in principle accurately identify those individuals who consume the food at any time during a given year.

Scientists working with food consumption data have long recognized that consumption by a "typical" individual will not be representative of consumption by people who eat large amounts of a particular food. This has stimulated interest in examining the distribution of average daily consumption levels across individuals in order to estimate consumption by individuals who consistently consume greater quantities of the food of interest than the average. This distribution of average daily consumption across individuals can be used to estimate upper quantiles of consump-

tion, such as the 90th, 95th, or 99th percentile. Reliable estimates of extreme percentiles can, however, be obtained only with relatively large sample sizes. Because the distribution of average daily intakes based on a sample of food consumption records for several days includes variability both between children and among days within children, this distribution will be subject to greater dispersion than would be the case if day-to-day variability were eliminated. (In the ideal case, this could be achieved by monitoring food consumption data over a full year or by using food frequency questionnaires.) The implication of such overdispersion is that upper percentiles of consumption will be overestimated.

Peak Levels of Consumption

Although the average level of individual exposure to pesticide residues in food is an important determinant of chronic toxicity, peak levels of exposure are more relevant for evaluating acute toxicity. Episodes of relatively high exposure occurring in a single day or even during a single meal may be more pertinent for acute risk assessment, depending on the toxic effect of interest.

The 1977-1978 NFCS provides information about food consumption during individual eating occasions for 3 different days. These data permit estimation of the total ingestion of a particular pesticide for each individual in the survey on each day. Using data for different individuals in the survey, one can estimate the distribution of person-days of consumption of specific foods. By combining this information with data on the distribution of pesticide residues in the food product or products of interest, it is then possible to estimate the number of person-days each year during which exposure to pesticides in the diet will exceed a critical level such as the reference dose (RfD), as defined in Chapter 8.

Although average levels of consumption and exposure will be reasonably well estimated with this approach, upper percentiles will be underestimated since food consumption data are available for only 3 of the 365 days in a year that are of interest. This is in contrast to the case for chronic risk assessment, where upper percentiles of exposure are likely to be overestimated.

Residue Monitoring

The point at which food samples are taken will influence the residue levels found. The highest residue levels generally occur immediately following application, and are reflected in field trial data. In samples taken for surveillance or compliance purposes, the residues will generally be higher than those in samples randomly drawn from the entire stock of a

particular food commodity available for sale in a particular region of the country. Market basket surveys are based on a composite sample of a limited number of commonly consumed foods after they have been cooked or prepared for consumption in the usual manner. Although market basket surveys provide residue data under conditions designed to emulate foods as consumed, they are limited because they provide only composite sampling results on a few foods included in a typical meal.

Most analytical methods for measuring pesticide residues in food are subject to an LOQ below which residue levels cannot be accurately determined. Although improved analytical methods for testing for pesticide residues in food have made it possible to detect lower and lower residue levels, even the most sensitive techniques are subject to an LOQ. When residue levels below the LOQ are reported, it is not possible to determine whether the food contains no residue of the pesticide of interest or whether there is a residue present but at a lower level than can be detected with the analytical methods used.

This uncertainty about the actual residue level with residues below the LOQ confers uncertainty on the distribution of pesticide residues in food products and, subsequently, on the distribution of dietary exposure to pesticide residues by individuals consuming those foods. For example, consider a hypothetical distribution of residue levels based on the analysis of a number of food samples that may have been treated with a particular pesticide, as shown in Figure 7-1. The residues above the LOQ will generally follow a log-normal distribution. However, an appreciable proportion of the samples will produce results below the LOQ.

What can be inferred about residue levels in samples below the LOQ? The only certain inference is that the actual residue level lies between a lower limit of zero and an upper limit equal to the LOQ. (Even this upper bound may not be entirely correct, since analytical results near the LOQ will be subject to some degree of measurement error.) Because not all crops grown in the United States are treated with pesticides approved for use on those crops, it is possible that results below the LOQ may be entirely pesticide free.

Consider, for example, the data on the use of different pesticides approved for use on apples shown in Figure 7-2. The percentages of the U.S. apple crop treated with specific pesticides varies widely, ranging from a low of 1% for malathion to a high of 90% for azinphos-methyl. Thus, most apples will not contain residues of malathion and would produce residue levels below the LOQ when tested. It is also possible tests for azinophos could yield results generally below the LOQ if residues of this widely used pesticide were present at low but nondetectable levels.

The data on pesticide use in Figure 7-2 also reveal marked regional differences in pesticide usage patterns in different regions of the country.

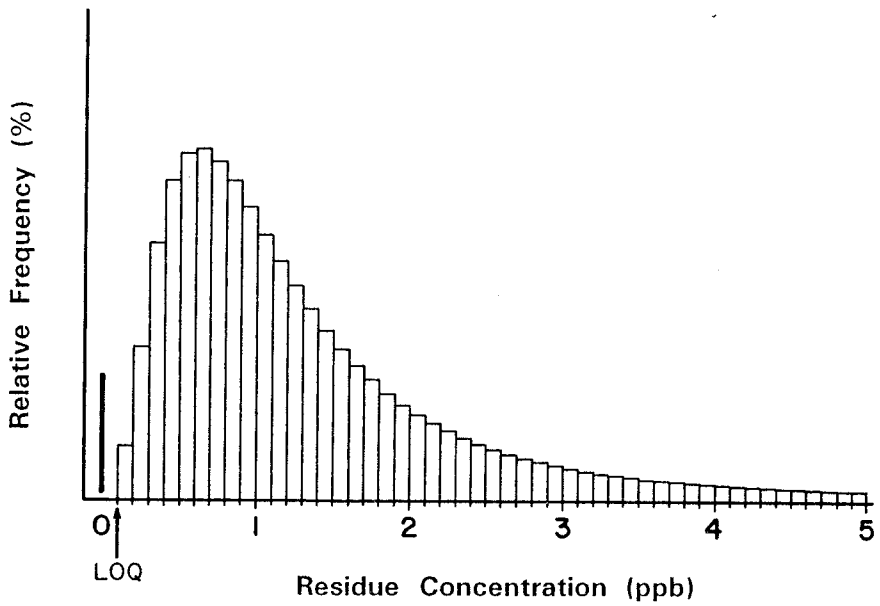


FIGURE 7-1 Hypothetical distribution of residue levels with a log-normal distribution for residues greater than zero.

Captan, for example, is widely used on apples grown in the central, northeast, and northwest regions of the United States but is virtually unused in the western regions of the country. Variation between pesticide usage patterns in different countries also warrants consideration with regard to imported food products.

In the past, results below the LOQ have been handled in different ways. A simple resolution of this uncertainty is to assume that all the results below the LOQ contain no residue and to assign them a residue level of zero. This is an optimistic approach, since the possibility of small but undetectable residues in some or all such samples cannot be excluded. A conservative approach is to assume that all residue levels are present at the LOQ. Although this may provide an upper bound on undetectable residues, it is unlikely that all the samples for which no residue was detected actually contain residues equal to the LOQ. An intermediate approach is to assume all nondetectable residues are present at one-half the LOQ. Clearly, the lower the LOQ, the less difference there will be between these different approaches, and the less uncertainty the LOQ will confer on estimates of potential human exposures.

Combining Residue and Exposure Data

Variation in food consumption patterns and in levels of pesticide residues in food leads to variation in dietary exposure to pesticides among infants and children. This variation in the ingestion of pesticide residues is characterized by a distribution of exposures, reflecting high, low, and average exposure concentrations. Statistically, the distribution of exposures can be obtained by convoluting (i.e., combining) the distribution of food consumption with the distribution of pesticide residues in food (Feldman and Fox, 1991). Thus, once the food consumption and residue distributions have been determined, the distribution of dietary exposures can be calculated (Figure 7-3).

The technical basis of convoluting two distributions can be described briefly as follows. Let C denote the consumption of a particular food by an individual, R the residue level in that food, and e the corresponding dietary intake or exposure level. The level of consumption will vary from person to person in accordance with the cumulative distribution $F_C(c)$ with corresponding density $f_C = F^1$. Note that $F_C(c)$ denotes the proportion

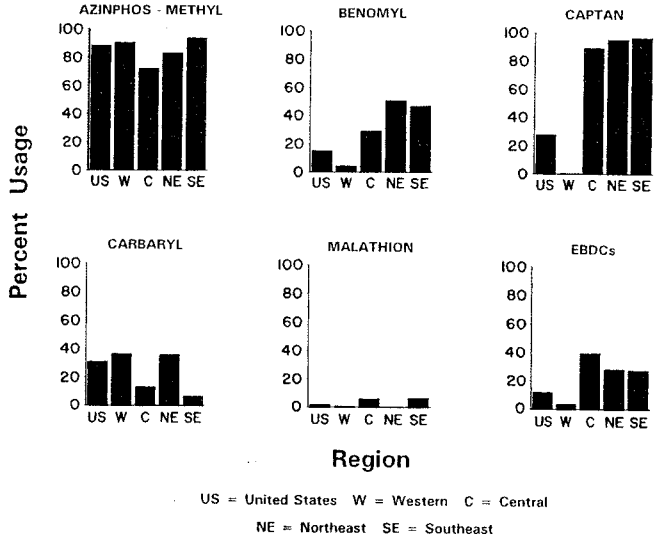


FIGURE 7-2 In the 1990 apple crop, percent of apple production treated with the following chemicals: azinphos-methyl, benomyl, captan, cabaryl, malathion, and EBDCs.

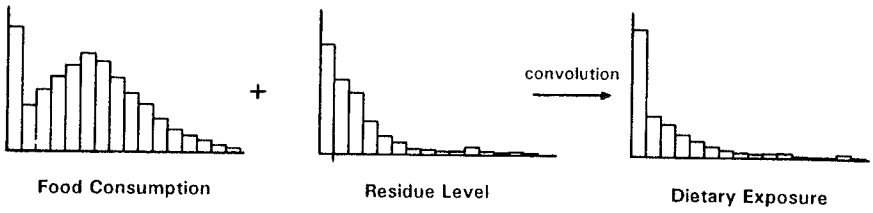


FIGURE 7-3 Convolution of food consumption distributions and residue distributions to produce dietary exposure distributions.

of the people in a given age group whose consumption C is less than a particular value c; the densities f_C and f_R reflect the relative frequency of different levels of consumption and residue, respectively, within the group. Letting F_R denote the residue distribution with density $f_R = F_R^1$, the distribution $F_E(e)$ of dietary intakes is defined by

$$F_E(e) = \int_o^e F_C\left(\frac{e}{r}\right) f_R(r)\,dr,$$

assuming that consumption C and residues R are statistically independent (Feldman and Fox, 1991, p. 349). This relationship provides the technical basis for combining the consumption distribution F_C with the residue distribution F_R to obtain the exposure distribution F_E.

In practice, estimates of consumption and residue distributions are based on survey data and are represented as histograms based on the observed sample. (If different weights are attached to the survey observations, a weighted distribution should be used.) Computationally, these two distributions can then be convoluted simply by taking the result of each point from the consumption distribution and multiplying it by each point in the residue distribution; the distribution of dietary intakes is then defined by the distribution of these products.

This empirical approach to convolution will work well, provided that the number of observations used to obtain the consumption and residue distributions is not large. With large distributions, the computation burden can be reduced by working with a random sample of both the consumption and residue data. The Monte Carlo approach (i.e., random sampling) to convolution was used by the committee in those examples where the computational effort required to convolute the two distributions was found to be excessive. The form of Monte Carlo sampling used by the committee was simply a means of reducing the amount of computational time required for convolution by using the original consumption and exposure distributions; no artificial distributional assumptions were re-

quired to implement this technique. The Monte Carlo distribution of dietary exposures will converge to that based on the entire exposure distribution as the number of Monte Carlo samples increases. As the number of samples converge, the distributions become identical.

The convolution method can be extended to more complex situations such as the estimation of total exposure to a pesticide that may be present on more than one food commodity. In this case, a single point on the exposure distribution is estimated by randomly combining points from the consumption distributions for all foods of interest with points from the corresponding residue distributions (one for each food), and then summing the total exposure across all foods. This process is repeated to generate a distribution of total exposures from all foods combined.

Total exposure to pesticides within the same class can be estimated in a similar fashion using the relative potency values for those pesticides to express the intake in toxicity equivalence factors. This is illustrated in the example of organophosphate pesticides later in this chapter.

LONG-TERM EXPOSURE TO BENOMYL

The Compound

Benomyl, or Benlate, is a systemic fungicide that was used in the United States from the time of its registration in 1972 until registration was voluntarily withdrawn for postharvest use by the manufacturer in 1989. Before then, it was the most widely used of the fungicides in the family of benzimidazole pesticides.

Benomyl is effective in preventing more than 190 fungal diseases, and it acts as a protective surface barrier while also penetrating the plant tissue to arrest infections. It was applied as a seed treatment, a transplant dip, and a foliage spray and was registered for use on more than 70 crops in 50 countries, including imported foods such as bananas and pineapples. In the United States, more than 100 EPA tolerances were established for benomyl in a variety of foods and feeds.

Benomyl has been shown to induce hepatocellular carcinomas, and combined hepatocellular neoplasms occurred in male and female mice treated with benomyl at all doses. In tests that included methyl-2-benzimidazole carbamate (MBC)—a metabolite of benomyl—investigators observed combined hepatocellular neoplasms in male mice and hepatocellular adenomas, carcinomas, and the combined hepatocellular neoplasms in female mice (NRC, 1987). Because of its carcinogenic potential, exposure assessment for benomyl is based on the distribution of average ingestion levels for different individuals.

The Consumption Data

Data from USDA's 1985-1986 Continuing Survey of Food Intake of Individuals (CSFII) were found to be more suitable for use in this example than the data from the 1977-1978 NFCS: the CSFII is more current than the NFCS, and includes consumption data collected over 6 days at 2-month intervals over an entire year, as compared to the 3-consecutive-day sample in the 1977-1978 NFCS. The CSFII included 170 1-year-olds, 195 2-year-olds, 225 3-year-olds, 191 4-year-olds, and 209 5-year-olds.

Intake data were divided into person-day food intake, and consumption was then averaged for each individual in each age class across the days of the reporting period. For example, 170 average daily intake values were recorded for each food for each of the 1-year-old children surveyed. A probability distribution of exposures could then be constructed for each yearly age class.

The Residue Data

Five different sets of residue data on benomyl residues were reviewed by the committee for this example. They were

- results of field trials conducted by the manufacturer,
- results of a market basket survey conducted by the manufacturer,
- 1988-1989 compliance and surveillance data collected by the Food and Drug Administration (FDA),
- data provided by the food industry, and
- data from tests of raw food by a certification business operating in California.

These data were collected using different sample designs, sample sizes, and analytical methods. Table 7-1 compares the number of benomyl samples and the number of detections for the FDA surveillance data, the manufacturer's field trials and market basket surveys, the food industry's data, and data supplied by the certification business. Data on apples, grapes, oranges, peaches, and tomatoes are shown in Figure 7-4 for all but the certification business.

Estimation of Exposure

Exposure was estimated using each of the five sets of residue data reviewed by the committee separately. An individual child's exposure to benomyl from a particular food was estimated by multiplying the mean

TABLE 7-1 Number of Benomyl Samples and Detections for Selected Foods Based on Data from the FDA, a Pesticide Manufacturer, the Food Industry, and a Certification Business

	FDA		Manufacturer's Field Trials		Manufacturer's Market Basket		Food Industry		Certification Business	
Food	No. of Samples	No. of Detections	No. of Samples	No. of Detections	No. of Samples	No. of Detections	No. of Samples	No. of Detections	No. of Samples	No. of Detections
Apple	134	35	138	122	26	5	68	30	127	65
Apple juice							30	16		
Apricot							6	0	19	5
Banana	72	8					4	0		
Bean	5	0	35	29	30	3	19	0	38	10
Blueberry							3	0	14	3
Carrot									12	1
Celery									24	4
Cherry	21	5					7	0	4	1
Cucumber					21	1			4	1
Grape	27	12	71	65	11	4	6	0	11	5
Watermelon	5	0					3	0		
Nectarine	14	8	18	5					39	14
Orange	6	0	18	13	12	12	1	0	6	1
Orange juice	1	0					2	0		
Peach	26	13	82	72	37	1	15	0	81	44
Pear	23	1	15	14	24	6				
Pineapple	25	18							7	4
Plum	21	10							28	18
Raisin							13	0		
Raspberry	14	0					2	0	17	6
Rice							6	0		
Squash	4	0					4	1		
Strawberry	30	2					6	0	16	11
Tomato	20	0	35	23	25	1				
Wheat							12	0		

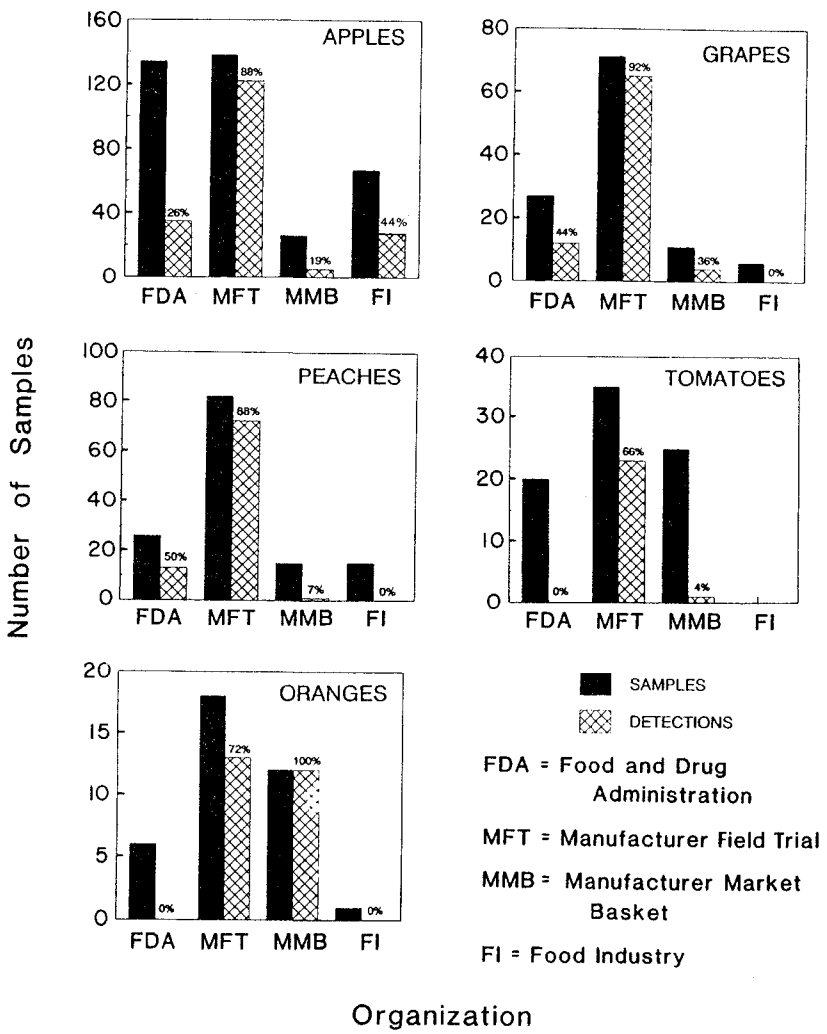


FIGURE 7-4 Number of benomyl samples and detections in apples, grapes, oranges, peaches, and tomatoes.

residue for that food by the average daily intake of that food. The exposures were then summed across up to 26 foods that 1-year-old children consume most to produce an average daily exposure estimate for each child. Note that different foods would be included with different residue data sets, depending on the availability of residue data for those foods. Finally, the distribution of average daily exposure from all foods combined across individuals was calculated.

The committee did not adjust these estimates for the percentage of the crop acreage treated. That adjustment is customarily applied by the EPA to residue data from field trials, thereby substantially reducing estimated exposures. EPA also multiplies the number of samples with no detected residues by the percentage of crop treated and assumes that residues in those samples are at the LOQ while the remainder of the undetected residues are at zero, i.e., *anticipated residues*.

In the procedure described in this chapter it is assumed that all crops are treated with benomyl. The committee notes that this is an unlikely scenario; however, the purpose of this analysis is to illustrate the probability distribution approach to estimating exposure.

The Manufacturer's Field Trials

In 1989 the manufacturer submitted to the EPA a substantial amount of residue data obtained from field trials in support of the continued registration of benomyl. These data are useful because they included information on the application rate (i.e., frequency of application [%] and amount used) and on the residue levels detected in each sample of raw agricultural commodity. Furthermore, sample sizes for single raw commodities were often large enough to permit statistical analysis. Unfortunately, many processed foods were not sampled for residues, thus forcing the EPA to rely on assumptions about the fate of residues during processing.

Data generated by the manufacturer for benomyl residues on fruit products following processing are shown in Table 7-2. Detected levels shown in those data are likely to be far higher than those in actual market basket data, due to the uneven use of the compound throughout the United States. In a nationwide market basket survey, fewer samples treated with lower amounts of benomyl would actually be found than in the manufacturer's field trials, which focused on crops known to be treated with benomyl.

As shown in Table 7-1, a total of 412 samples of eight unprocessed foods (apples, beans, grapes, nectarines, oranges, peaches, pears, and tomatoes) were tested. Of these, 343 (83%) contained residues above the detection limit. Since neither apple juice nor orange juice were sampled,

TABLE 7-2 Changes in Benomyl Concentrations During Washing and Processing

Food	Food Form	Reduction, %
Apples	Washed	13
	Juice	69
	Applesauce	82
Peaches	Washed	73
	Canned	99
Bananas	Pulp	No detectable residue

SOURCE: Based on data from the pesticide manufacturer.

EPA must rely on the results of processing studies, such as those shown in Table 7-2, to determine the fate of residues in juices most consumed by young children.

The committee conducted two separate analyses of these data based on two different assumptions: that all reported nondetections were actually zero (Figure 7-5) and that nondetections were really residues at the LOQs, which were provided for each sample (Figure 7-6). (The actual exposure is somewhere in between those shown in the two figures.) The exposure estimates were not greatly affected by either assumption, principally due to the relatively high number of samples containing detectable residues. Estimates of young children's benomyl exposure based on the manufacturer's field trial residue data are almost identical, regardless of whether a value of zero or the LOQ is used in exposure calculations when no residue is detected. Estimates based on the manufacturer's market basket data are also comparable, regardless of the value assigned to nondetectable residues.

That portion of Figures 7-5 and 7-6 displaying the manufacturer's field trial data was constructed by combining the individual consumption reports with the mean of field trial residues for each of 10 foods: apples, apple juice, oranges, orange juice, grapes, grape juice, peaches, pears, green beans, and tomatoes. The committee could therefore produce separate exposure estimates for each food for each child from 1 to 5 years of age. Each analysis was conducted under the assumptions that only those 10 foods were consumed in a child's diet and that the juices lost no benomyl during processing.

The Manufacturer's Market Basket Survey

The sample design of a market basket survey is important, since the results can be dramatically affected by regional patterns of pesticide use and food distribution. A good design can obviate the need to make com-

plex assumptions regarding processing, percentage of crop treated, and food distribution effects.

A limited number of foods was surveyed in the manufacturer's market basket survey. A total of 143 samples of 7 foods, and no juices, were analyzed for a benomyl. Thirty-two (22%) of the samples contained residues at levels above the LOQ. This percentage is similar to that found by the food industry but approximately 50% lower than that detected by the more focused sampling design used by the certification business.

FDA Surveillance Data

The committee used only FDA surveillance data in assessing chronic exposures, and it did not estimate exposures for any food for which there were fewer than 20 samples. Although benomyl was registered for use on many foods, sample size exceeded 20 for only 10 of the 26 foods listed in Table 7-1. Of the total of 448 samples tested, 112 (25%) had residues that exceeded the LOQ.

FDA monitoring is focused on fresh rather than processed foods. Therefore, many of the processed foods often consumed by young children are never or seldom sampled, and the utility of small samples is limited in estimating exposures. As shown in Table 7-1, a number of foods sampled by other groups were not sampled at all by FDA in 1988 and 1989. Other weaknesses of FDA surveillance data are noted in Chapter 6 on pesticide residues.

The Food Industry

A food industry association provided a large amount of data collected from its member organizations. These data identified the food, the pesticide used, the residue level, and the LOQ of the analytical method used. Since the food industry used a variety of sampling and analytical methods, the representativeness of the data for the nation's food supply is uncertain.

Despite this uncertainty, these data are useful in illustrating the method proposed here for exposure estimation. The majority of the positive findings in this data set relate to apples (with 30 residues above the detection limit observed in 68 samples) and apple juice (with 16 of 30 samples showing positive).

A Certification Business

The committee obtained residue data from a certification business operating in California, a commercial organization that guarantees to grocery store owners and consumers that any residues in produce will be below

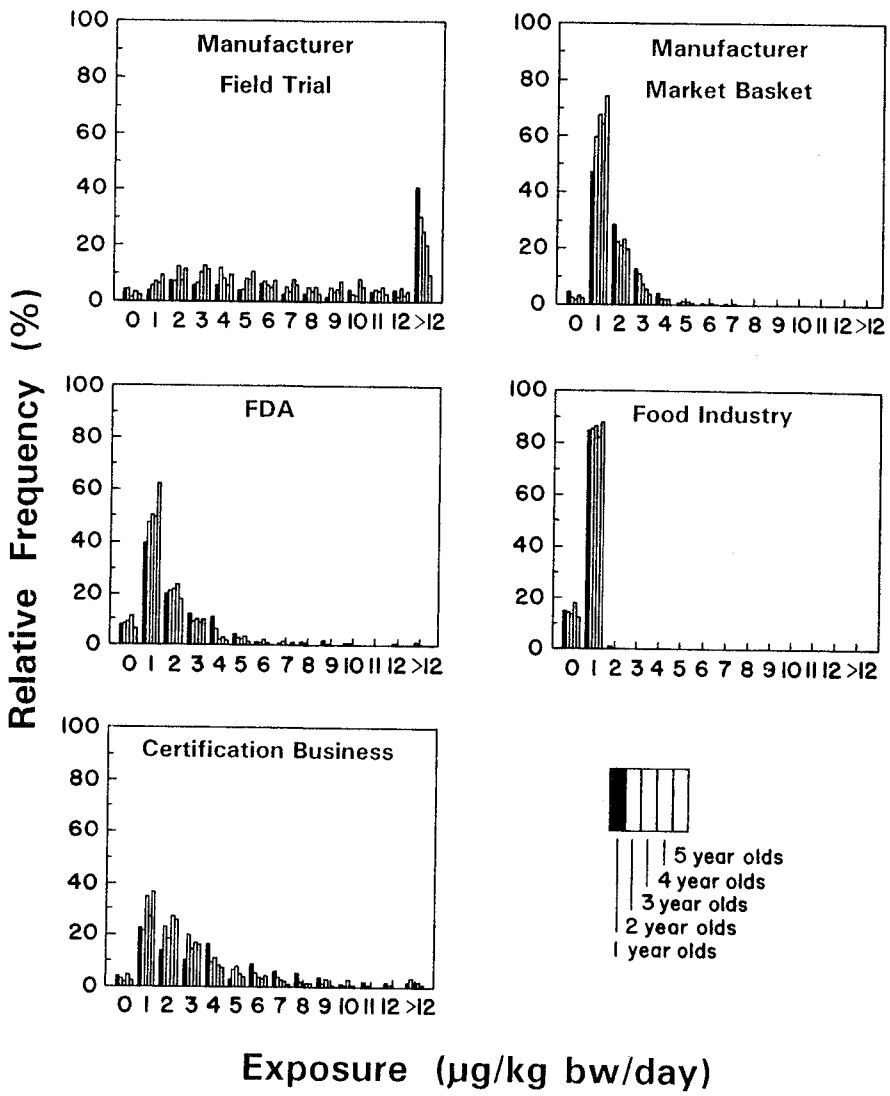


FIGURE 7-5 Daily exposure of 1- to 5-year-old children to benomyl in different combinations of foods, as shown by residue data from a certification business, FDA, the food industry, and the manufacturer (field trials and market baskets). Based on the assumption that the nondetects were equal to zero.

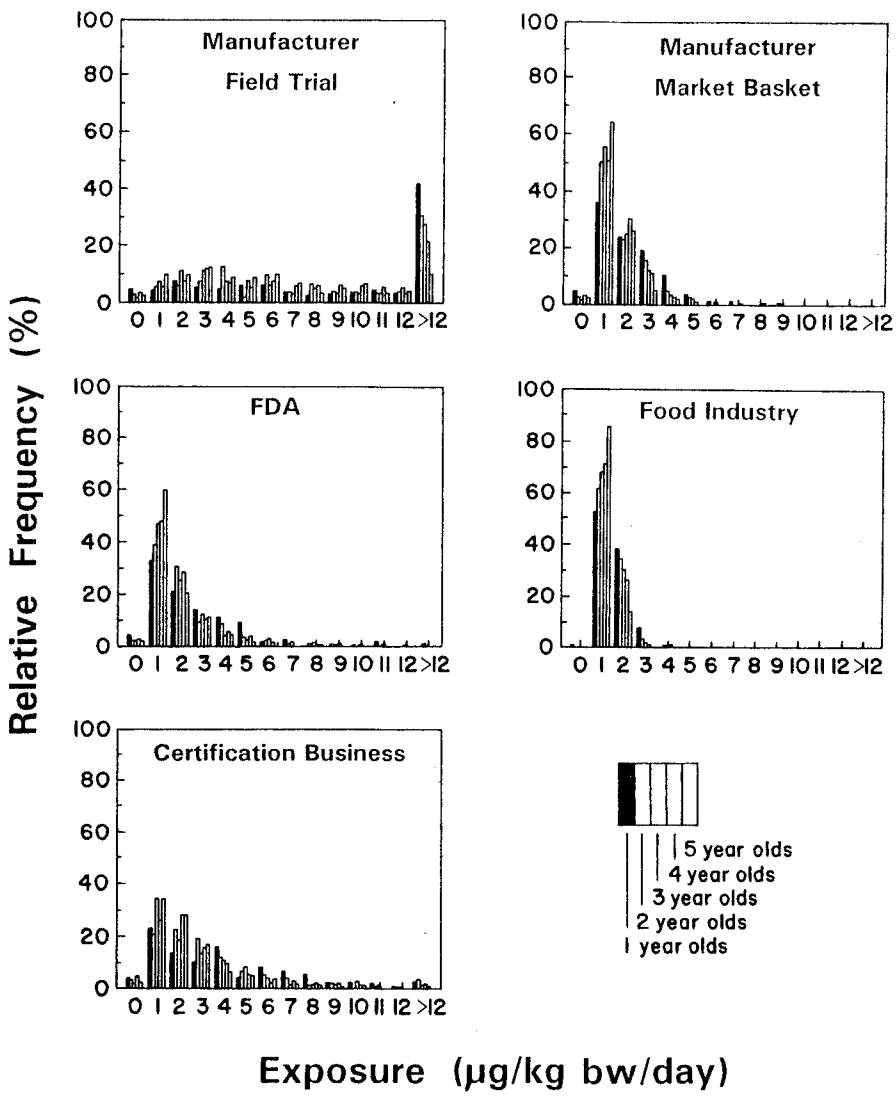


FIGURE 7-6 Daily exposure of 1- to 5-year-old children to benomyl in different combinations of foods, as shown by residue data from a certification business, FDA, the food industry, and the manufacturer (fields trials and market baskets). Based on the assumption that the nondetects were equal to the LOQ.

a detectable level. These data are subject to certain limitations such as nonrepresentative localized sampling, unidentified analytical methods, and analysis of selected produce. Nonetheless, they are used in this example to provide a range of data for comparison purposes. As shown in Table 7-1, this organization tested 447 samples of which 193 (43%) had benomyl residues above the detection limit. This compares to 448 FDA samples with 112 (25%) positive detections and to 203 food industry samples with 42 (20.6%) detections over the LOQ. The difference could be explained by the certification business's focus on produce with a history of residues of concern. Moreover, although the certification business, FDA, and the food industry tested a similar number of foods (16, 17, and 18, respectively), the selection of foods varied. For example, the food industry detected residues in 16 (53.3%) of 30 samples of apple juice, whereas the certification business tested no apple juice samples. Similarly, the certification business detected residues in 18 (64.3%) of 28 plum samples, but plums were not sampled by the food industry.

Summary

In this example, the committee examined multiple data sets reflecting concentrations of benomyl in several different foods. These data sets varied in different ways (Table 7-1). The manufacturer's field trial survey showed a much higher frequency of benomyl detections than found in the market basket survey, mainly because the study focused on crops known to be treated with benomyl. In the market place, fewer foods would actually have been treated with benomyl than in the field survey.

The effect of processing on benomyl concentrations in foods is shown in Table 7-2. These data indicate that substantial reductions in residues may occur because of processing. Figures 7-5 and 7-6 show the estimated exposure distributions for children 1 to 5 years of age to total benomyl residues for several common foods where residues below the LOQ were set at zero and at the LOQ, respectively.

The most important result of this analysis is that most children are exposed to relatively small concentrations of benomyl in their diets (less than 0.012 mg/kg bw/day), if they are exposed to any at all. Current exposures could be even less since registration of benomyl for postharvest use has been suspended. In any case, their exposures would be much below the current reference dose (RfD). In the committee's analysis, it is only the manufacturer's field trial data that suggest some children have larger exposures, and as noted above, these data would be inappropriate for this type of analysis. Both the manufacturer's market basket and the certification business's residue data show relatively small total residue exposures. Finally, Figures 7-5 and 7-6 show that assigning a value of

zero or the LOQ to nondetectable residues had little effect on the overall outcome of the committee's calculations for benomyl. A principal reason for this finding is that a relatively high number of benomyl samples contained detectable residues. Certain impacts on exposure estimates would be seen for pesticides where a relatively low number of samples contained detectable residues.

SHORT-TERM EXPOSURE TO ALDICARB

The Compound

The evaluation of short-term peak exposures is illustrated using data on aldicarb residues in potatoes and bananas. Aldicarb is an acutely toxic pesticide whose use on potatoes and bananas was voluntarily suspended by the manufacturer in 1990 and 1992, respectively. It is an *N*-methyl carbamate that exerts its toxic effects by inhibiting the enzyme cholinesterase in the central and peripheral nervous system and at the neuromuscular junctions. Single oral doses of 25 μg/kg bw in humans produces approximately 50% inhibition of blood cholinesterase (NRC, 1986). Inhibition above 30% is usually of concern in humans.

Aldicarb is a systemic toxicant that is used primarily as an insecticide and nematocide. It is absorbed by the roots, stems, leaves, and fruits of plants. Aldicarb sulfoxide is a toxic metabolite that is distributed throughout the plant and degrades relatively slowly. Aldicarb-treated crops commonly eaten by children include potatoes, bananas, and citrus fruits. As demonstrated in Chapter 5, infants and children consume proportionately more of these foods than do adults with the exception that infants do not eat potatoes.

Acute Effects of Dietary Aldicarb Exposure

In 1970 Union Carbide gave three groups of four healthy male adult volunteers doses of aldicarb at concentrations of 25, 50, or 100 μg/kg bw. Subjects given the highest dose became acutely ill; one of those who received the lowest dose developed severe mood symptoms (i.e., anxiety reaction). Whole blood cholinesterase depression was observed in all the subjects. After reviewing the results of this study, the Safe Drinking Water Committee of the National Research Council estimated a no-observed-adverse-effect level (NOAEL) of 10 μg/kg bw/day (NRC, 1986). Applying a 10-fold uncertainty factor, the EPA established an RfD, formerly an acceptable daily intake (ADI), for aldicarb at 1.0 μg/kg bw/day.

Studies in the dog demonstrate depression of plasma cholinesterase at doses as low as 1 ppm (20 μg/kg bw/day). At doses of 50 μg/kg bw/

day, there were statistically significant increases in diarrheal stools in both sexes, along with statistically significant increases in both plasma and RBC cholinesterase inhibition in males. Applying an uncertainty factor of 100 on the lowest dose level (20 μg/kg bw/day) to account for interspecies and intraspecies variation, EPA identified an RfD of 0.2 μg/kg bw/day. EPA's Scientific Advisory Panel recommended on November 6, 1992, that the RfD for aldicarb be reestablished at 1.0 μg/kg bw/day, consistent with the 1986 NRC recommendation (EPA, unpublished data, 1993).

In July 1985, severe acute illness was observed in more than 1,000 people in the western United States a few hours after they had eaten watermelons treated with aldicarb—a nonregistered (illegal) use. The symptoms included nausea, vomiting, diarrhea, muscle fasciculations, mood changes, and other symptoms of cholinergic poisoning. The most seriously ill person was a 62-year-old woman who had eaten approximately one-fourth of a watermelon later found to contain a 2.7-ppm concentration of aldicarb sulfoxide, which presented an estimated dose of 57 μg/kg bw. She required emergency room treatment and atropine to reverse the symptoms.

The Consumption Data

Data on consumption of bananas and potatoes by children between 12 and 24 months of age were obtained from the 1977-1978 NFCS. The mean, median, and 90th and 95th percentiles of the average daily consumption of bananas and potatoes are shown in Table 7-3. Of the 529 children surveyed, 157 did not eat either bananas or potatoes on any of the days during which the survey was conducted; of those that did, fewer children ate potatoes than bananas. Of the 1,831 person-days included in the survey, there were 1,077 days on which neither bananas nor potatoes were consumed. The distribution of daily consumption of bananas and potatoes by children between 12 and 24 months of age is shown in Table 7-4.

TABLE 7-3 Average Daily Consumption by Children Between 12 and 24 Months of Age

Food	Sample Size	Number of Noneaters	Consumption, g/kg bw/day			
			Mean	Median	P90	P95
Bananas	529	208	0.90	0.16	3.01	4.17
Potatoes	529	349	0.72	0	2.25	3.41

SOURCE: Based on data from the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978.

TABLE 7-4 Daily Consumption by Children Between 12 and 24 Months of Age

Food	Sample Size	Number of Days No Food Eaten	Consumption, g/kg bw/day			
			Mean	Median	P90	P95
Bananas	1,831	1,224	0.91	0	2.52	7.47
Potatoes	1,831	1,602	0.69	0	2.38	5.20

SOURCE: Based on data from the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978.

The Residue Data

Until the early 1980s, residue sampling focused on aldicarb rather than on aldicarb sulfoxide—its persistent and more toxic metabolite. Furthermore, only composite samples were used. That is, many individual samples of a single commodity were blended and the resulting mixture was analyzed. Because aldicarb is an acute toxicant, and the foods it contaminates are often eaten individually, EPA required a survey in which individual foods were examined separately. In this survey, concentrations higher than the EPA tolerance level were found in individual bananas and potatoes, although they had not been detected previously in blended samples.

Sampling revealed that the distribution of residues in individual potatoes from treated fields followed a log-normal distribution pattern, that is, most individual sample results were clustered at low concentrations. The highest single concentration, 8.7 ppm, was found in one potato. In the event that a 20-kg child consumed that one 200-g potato, cooked by itself in a microwave oven, the child could receive an exposure of 87 μg/kg bw—an acutely toxic level. This illustrates the potential problems associated with use of composite samples for evaluation of exposure to acute toxicants.

Banana trees have been treated with aldicarb since 1977. In 1991 composite and individual samples of bananas were analyzed in five strictly controlled field trials. Half the bananas from one field were found to contain aldicarb residues higher than the tolerance level of 0.3 ppm. If a 20-kg child were to eat the 170-g edible portion of a single banana at the highest level found, 3.14 ppm, the resulting dose would be 26 μg/kg bw—again a potentially toxic dose. Even at the 0.3-ppm tolerance level for aldicarb in bananas, that child would be exposed to approximately 3 μg/kg bw—a level well above the RfD. This does not take into account exposure to other cholinesterase inhibitors in the diet, including possible aldicarb residues in citrus fruit or potatoes.

Since pesticides are usually approved for use on more than one food, it is important to consider the total exposure to a particular pesticide from

TABLE 7-5 Residues of Aldicarb

Food	Sample Size	Number Below LOQ	Mean Residue, ppm			
			Mean	Median	P90	P95
Bananas	2,697	2,442	0.008	0	0.02	0.05
Potatoes	294	6	0.239	0.085	0.510	0.840

SOURCE: Based on data from the 1987 National Aldicarb Food Survey and survey data from the manufacturer.

all dietary sources. The methods for assessing exposure to aldicarb in bananas can be extended to cover multiple foods. Although the committee used only two foods (bananas and potatoes) in its example, extension of the method to more than two foods is straightforward. In fact, the FDA data on aldicarb discussed in Chapter 6 failed to identify residue levels above the LOQ in the 350 samples tested. Consider the data on residues of aldicarb in bananas examined earlier, along with data on residues of aldicarb in potatoes given in Table 7-5. The former data are from the National Aldicarb Food Survey. The latter data are from a special survey conducted by the manufacturer of the compound in 1989. In the manufacturer's survey, representative samples were taken from 26 locations in the states of Washington, Oregon, California, Michigan, and Maine. Only three of the locations, Washington, Oregon, and Maine, had composite samples with residues above the LOQ. Residue data from these three states were also selected because they gave data for individual potatoes. Of the 294 reported residue values, 6 were below the LOQ. The mean aldicarb residue of 0.239 ppm in potatoes is much higher than the mean residue of 0.008 ppm in bananas. This difference is due largely to the use of field trial data for potatoes, which were obtained from crops known to have been recently treated with aldicarb.

Effects of Assumptions Regarding Residues Below the LOQ

The implications of results below the LOQ for exposure estimation can be illustrated using data on the levels of aldicarb in bananas obtained from the 1987 National Aldicarb Food Survey. The composite samples tested in this survey were obtained from 225 groups averaging 12 bananas each—a total of 2,700 bananas. These samples were initially tested for the presence of aldicarb with an analytical method that had an LOQ of 0.01 ppm. If any composite sample was found to have a residue greater than 0.01 ppm, each banana in that group was analyzed individually. In this survey, residues over 0.01 ppm were detected in 27 of the 225 composite samples. The investigators then conducted separate tests on the 299 bananas that were available for testing out of the 302 bananas in the 27

TABLE 7-6 Residues of Aldicarb in Bananas

Number Below LOQ/Sample Size	Value Used for Residues Below LOQ	Mean Residue, ppm			
		Mean	Median	P90	P95
2,442/2,697	LOQ	0.017	0.01	0.02	0.05
	0	0.008	0	0.02	0.05

SOURCE: Based on data from the 1987 National Aldicarb Food Survey.

samples. They found aldicarb concentrations above the LOQ in 255 of those bananas.

For risk assessment purposes, let us assume that the remaining 2,442 bananas in the sample had residue levels below the LOQ. This underestimates actual residue levels because compositing masks any unusually high residue levels on individual bananas in a given batch. Let us also assume that individual bananas testing negative do not contain residues above the LOQ. Despite these approximations, it is instructive to examine the impact of assumptions regarding residues lower than the LOQ on estimation of dietary exposures to aldicarb from bananas.

Table 7-6 presents the mean, median, and upper 90th and 95th percentiles of aldicarb on the 2,697 bananas in the survey sample. The mean residue level obtained by assigning a value of 0.01 ppm to all residues below the LOQ is 0.017 ppm—slightly more than twice the value of 0.008 ppm obtained by assigning a value of 0 to nondetectable residues. The median value of 0.01 ppm obtained by substituting the LOQ for nondetectable residues is close to the corresponding mean residue. Assigning a value of 0 to the bananas with no detectable residues leads to a median residue of 0. Since less than 10% of the detections were above the LOQ, both the 90th and 95th percentiles of the residue distribution are unaffected by the value chosen for observations below the LOQ.

Estimating Dietary Exposure

Of the 529 children between 12 and 24 months of age in the 1977-1978 NFCS, only 321 reported eating bananas on any of the 3 days during which food consumption data were recorded. The mean daily consumption of bananas among all the children surveyed was 0.90 g/kg bw/day (Table 7-7). The mean consumption by the 321 children who ate bananas on at least one occasion during the survey was 1.47 g/kg bw/day. Since 61% of the children consumed bananas at least once, the upper 90th and 95th percentiles for the subgroup of eaters are only slightly higher than the corresponding consumption percentiles for the entire sample. The 90th and 95th percentiles will be overestimated since the distribution of average daily consumption contains variability between children and among days.

TABLE 7-7 Daily Consumption of Bananas by Children Between 12 and 24 Months of Age

Subsample	Sample Size	Consumption, g/kg bw/day			
		Mean	Median	P90	P95
All	529	0.90	0.16	3.01	4.17
Eaters only	321	1.47	0.48	3.66	5.67

SOURCE: Based on data from the 1987 National Aldicarb Food Survey and USDA's Nationwide Food Consumption Survey, 1977-1978.

The aldicarb residue distribution shown in Table 7-6 may be combined with the distribution of mean daily intake of bananas shown in Table 7-7 to estimate the distribution of mean daily intakes of aldicarb residues on bananas. Statistically, this is accomplished by convoluting the two distributions by pointwise multiplication of the residue and consumption distributions to obtain an estimate of the distribution of intakes.

The results of this calculation are summarized in Table 7-8. Separate estimates of intake are presented for the entire sample and for the subsample of children who ate bananas during the survey period. Separate estimates of intake are given for residue levels below the LOQ using the assumptions that the residues were either 0 or at the LOQ.

These estimates of mean intake in Table 7-8 are identical to those obtained simply by multiplying the mean consumption of bananas by the mean residue concentration (Table 7-9). As indicated in Table 7-9, however, multiplication of the upper 90th percentile of the residue and consumption distribution in this fashion does not yield the 90th percentile of intake based on the method of convolution (Table 7-8). The discrepancy between these two values is particularly large for the subgroup of banana eaters only with nondetectable residues assigned a value of 0. Thus, esti-

TABLE 7-8 Daily Intake of Aldicarb from Bananas for Children Between 12 and 24 Months of Age

Subsample	Value Used for Residues Below LOQ	Intake, μg/kg bw/day			
		Mean	Median	P90	P95
All	LOQ	0.015	0.002	0.034	0.058
	0	0.007	0	0	0.008
Eaters only	LOQ	0.025	0.005	0.050	0.087
	0	0.012	0	0.003	0.032

SOURCE: Based on data from the 1987 National Aldicarb Food Survey and USDA's Nationwide Food Consumption Survey, 1977-1978.

TABLE 7-9 Methods for Estimating the Mean and 90th Percentiles of Aldicarb Intake

	Chronic	Acute			
Subsample	Values Used for Residues Below LOQ	Mean	P90	Mean	P90
All	LOQ	0.015	0.060	0.015	0.050
	0	0.007	0.060	0.007	0.050
Eaters only	LOQ	0.025	0.073	0.047	0.208
	0	0.012	0.073	0.022	0.208

SOURCE: Based on data from the 1987 National Aldicarb Food Survey and the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978.

mates of upper percentiles of ingestion should be based on the more accurate method of convolution. At the upper percentiles, estimates are higher than the true percentiles since the average consumption distribution incorporates day-to-day variability.

This method is most appropriate for estimating the average daily ingestion of pesticide residues over an extended period. Although average daily ingestion is an appropriate measure of exposure for chronic risk assessment, a different approach is required for acute toxic effects caused by short-term exposure to relatively high levels of substances.

Assume that the total intake of a particular pesticide in a single day represents a good indicator of whether an acute toxic response will occur. In this event, we may examine the distribution of individual daily intakes in Table 7-10 rather than the distribution of average daily intakes shown in Table 7-7. In the present context, this corresponds to the distribution of individual daily intake of bananas for all days for which observations were recorded for all children in the survey. Convolution of this distribution with the aldicarb residue distribution provides an estimate of the distribution of the number of person-days in the sample associated with

TABLE 7-10 Daily Consumption of Bananas by Children Between 12 and 24 Months of Age

		Consumption, g/kg bw/day			
Subsample	Sample Size	Mean	Median	P90	P95
All	1,831	0.91	0	2.52	7.47
Eaters only	607	2.74	0.72	10.4	8.73

SOURCE: Based on data from the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978.

TABLE 7-11 Individual Daily Intake of Aldicarb from Bananas for Children Between 12 and 24 Months of Age

Subsample	Value Used for Residues Below LOQ	Intake, μg/kg bw/day			
		Mean	Median	P90	P95
All	LOQ	0.016	0	0.038	0.086
	0	0.008	0	0	0
Eaters only	LOQ	0.047	0.008	0.097	0.135
	0	0.023	0	0.007	0.047

SOURCE: Based on data from the 1987 National Aldicarb Food Survey and the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978.

a given daily intake of aldicarb (Table 7-11). This distribution thus provides a basis for estimating the percentage of person-days during which exposure would exceed a health-based exposure standard, such as a reference dose based on toxicity studies. The upper percentiles will be underestimated since the food consumption data are available for only 3 to 4 days of the 365 days in the year. Although several methods for dealing with results below the detection limit of the analytical method were discussed previously, all nondetectable residues were assumed to be zero in this analysis for simplicity.

The mean, median, and 90th and 95th percentiles of average daily intake and individual daily intake of aldicarb from bananas and potatoes alone and for bananas and potatoes combined are shown in Tables 7-12 and 7-13 for children between 12 and 24 months of age. The distribution is dominated by the intake of potatoes. Figures 7-7 and 7-8 show the distribution of individual and average intakes of aldicarb from potatoes and bananas, separately and combined. Intake values greater than 0.8 g/kg bw/day represented a very small proportion and were therefore omitted from the figures.

The distribution of aldicarb intake from both bananas and potatoes

TABLE 7-12 Average Daily Intake of Aldicarb for Children Between 12 and 24 Months of Age

Food	Intake, μg/kg bw/day			
	Mean	Median	P90	P95
Bananas	0.007	0	0	0.008
Potatoes	0.172	0	0.302	0.673
Bananas and potatoes	0.179	0	0.327	0.705

SOURCE: Based on data from the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978, the 1987 National Aldicarb Food Survey, and survey data from the pesticide manufacturer.

TABLE 7-13 Individual Daily Intake of Aldicarb for Children Between 12 and 24 Months of Age

Food	Intake, μg/kg bw/day			
	Mean	Median	P90	P95
Bananas	0.008	0	0	0
Potatoes	0.164	0	0.123	0.537
Bananas and potatoes	0.172	0	0.164	0.593

SOURCE: Based on data from the U.S. Department of Agriculture's Nationwide Food Consumption Survey, 1977-1978, the 1987 National Aldicarb Food Survey, and survey data from the pesticide manufacturer.

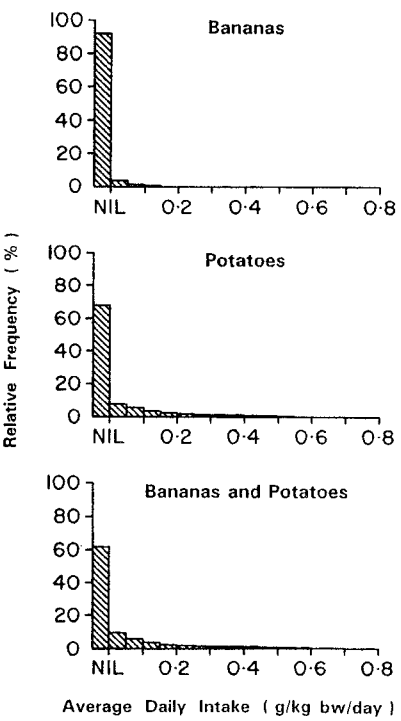


FIGURE 7-7 Distribution of the average daily intake of aldicarb from bananas and potatoes, separately and combined. SOURCE: Based on data derived from USDA, 1983, the National Aldicarb Food Survey, and survey data from the manufacturer.

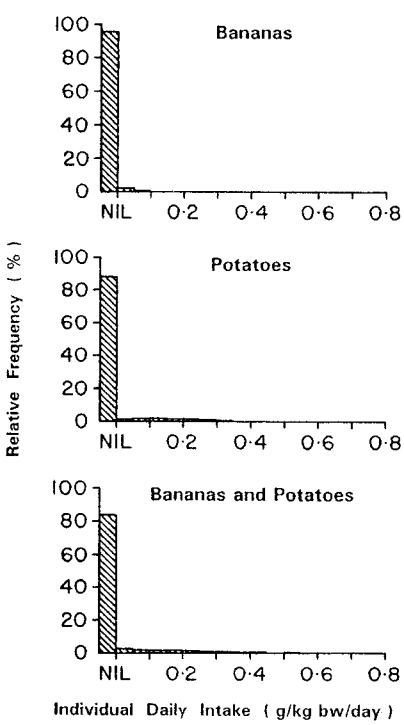


FIGURE 7-8 Distribution of individual daily intake of aldicarb from bananas and potatoes, separately and combined. SOURCE: Based on data derived from USDA, 1983, the National Aldicarb Food Survey, and survey data from the manufacturer.

resembles that of aldicarb intake from bananas or potatoes alone except for a slight shift toward nonzero values; the number of zeros has decreased from 66% of the values to 63%. This is to be expected since some children ate bananas but not potatoes or potatoes but not bananas.

Summary

Aldicarb was examined by the committee because it is an acutely toxic chemical that may potentially be found in several foods consumed by children and because there were also good sampling data for aldicarb residues in several commodities. This example could also be used to illustrate approaches to estimating residue concentrations in both individual foods and in multiple foods combined in a child's diet to provide total estimated residue.

Composite samples were used until the early 1980s for measuring residues in foods. Samples of a single commodity were blended, and the resulting mixture was analyzed. Because aldicarb is an acute toxicant, new approaches do not use blended samples. Rather, single commodities are analyzed. These new residue surveys have shown log-normal distribution patterns of aldicarb residues in commodities such as potatoes. As would be expected, most individual samples show residues clustered at lower concentrations or approaching zero.

The committee estimated exposure to aldicarb from both bananas and potatoes. The estimated distribution of aldicarb residues either from the single commodity potatoes or the combination of the two commodities shows exposure above the RfD of 1.0 μg/kg bw/day. In general, when these foods are eaten in the absence of other cholinesterase inhibitors, exposures would be much lower than those that would produce toxic effects. However, in the unlikely event that a single exposure occurred at the highest residue concentrations found in either bananas or potatoes, toxic effects could occur in a child.

Since the use of aldicarb on potatoes and bananas was voluntarily withdrawn by the manufacturer in 1990 and 1991, respectively, children are not presently at risk of cholinesterase depression from residues of aldicarb on these two foods (Debra Edwards, Chief, EPA Chemistry Branch, personal commun., 1993). Nonetheless, this case study illustrates the use of food consumption and residue distributions in estimating the number of person-days on which ingestion of an acutely toxic pesticide might exceed the RfD. As in the benomyl case, total ingestion from more than one food on which residues might be present was taken into account.

MULTIPLE EXPOSURE ASSESSMENT: ORGANOPHOSPHATE INSECTICIDES

Pesticide regulation in the United States has been focused on single chemicals rather than on combinations of compounds likely to appear as mixtures in the human diet. This practice can be attributed not only to the absence of data on the residues of multiple compounds that coexist on foods but also to the lack of methods for estimating simultaneous exposures to multiple chemicals, which cannot be accomplished merely by combining mean values (or other statistical summaries) of food intake and residue data. The regulatory process has therefore progressed on a chemical-by-chemical basis without consideration of possible additive and synergistic effects that could result from exposures to mixtures.

The committee developed a method for estimating exposure to multiple pesticides with a common toxic effect: in this case, inhibition of plasma cholinesterase (ChE). This method was used to determine how many children are likely to be exposed to unsafe levels of multiple pesticides with that common effect and to express the exposures in the most desirable form—person-day exposures—using actual individual daily consumption data and actual residue data.

More than 25 compounds that inhibit cholinesterase are permitted to exist as residues in foods. Although *N*-methyl carbamates inhibit cholinesterase, their mechanism of action is reversible and duration of action is shorter than for organophosphates. For purposes of simplicity, therefore, the committee selected five commonly used organophosphates (acephate, chlorpyrifos, dimethoate, disulfoton, and ethion) and used actual data on their presence on eight foods (apples, oranges, grapes, beans, tomatoes, lettuce, peaches, and peas) and three juices (apple, orange, and grape) to explore the development of methods for assessing exposure to multiple chemicals.

Criteria for choosing the five chemicals included the following:

- They must each exert the same adverse effect, in this case, blood plasma ChE inhibition.
- Credible estimates of the no-observed-effect level (NOEL) for ChE inhibition must exist for each chemical.
- The chemicals must be permitted as residues on several of the eight foods analyzed.
- FDA residue data must exist for the chemical-food groups selected.

The selection of foods for analysis was driven by the availability of data on residues and on the amount of each food consumed by 2-year-old children sampled in the USDA's 1977-1978 NFCS. An attempt was made to include foods that children consume most; however, it became

apparent that residues were most common on other foods, such as peaches, which were therefore included in the analysis. The frequency distributions for the foods analyzed by the committee are presented in Figure 7-9A-K.

Estimating pesticide exposure in this way was considerably constrained by the absence of residue data for certain foods and compounds, especially processed foods such as juices whose type of processing could greatly influence pesticide residue levels. There are few available residue data for processed foods; thus, little is known about the effects of processing on pesticide residues.

For this analysis, the committee assumed that exposures to ChE-inhibiting compounds should be summed across foods and compounds that induce a similar type of ChE inhibition. Although exposure to a single compound may not exceed the RfD, concurrent exposures to numerous compounds could exceed a safe level because of the increased ChE inhibition. It was also assumed that the toxic potencies of diverse compounds can be standardized by developing estimates of relative potency in the manner described below.

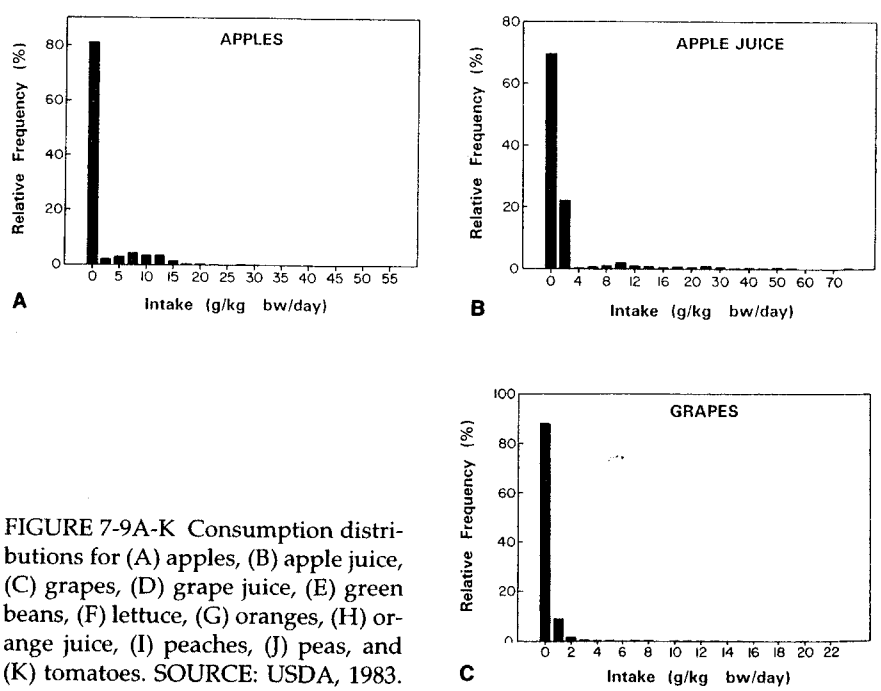
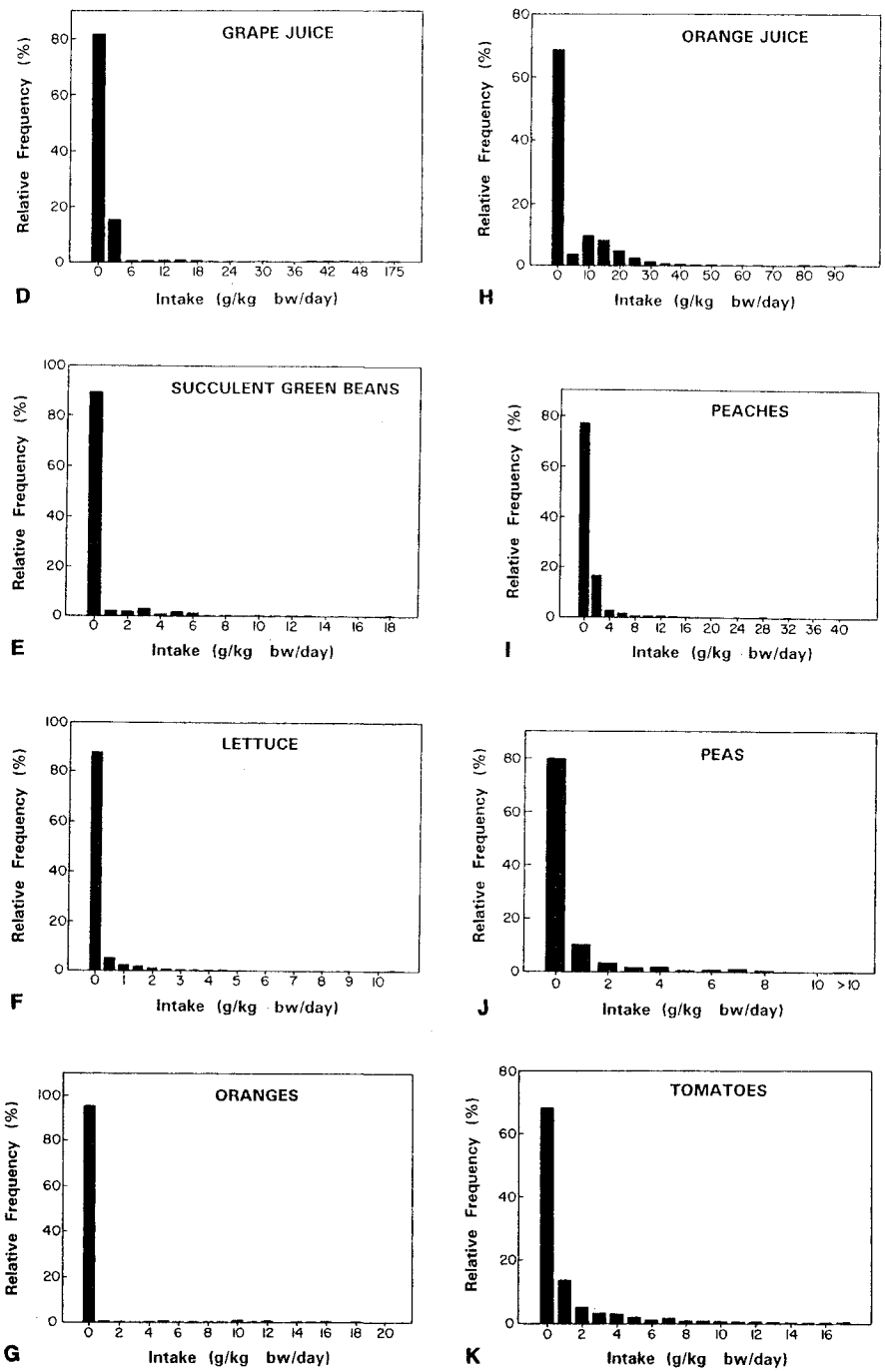


FIGURE 7-9A-K Consumption distributions for (A) apples, (B) apple juice, (C) grapes, (D) grape juice, (E) green beans, (F) lettuce, (G) oranges, (H) orange juice, (I) peaches, (J) peas, and (K) tomatoes. SOURCE: USDA, 1983.



Cholinesterase Inhibition

Among pesticides, organophosphate and carbamate insecticides are the ChE-inhibiting pesticides of primary concern. These chemicals bind with cholinesterases and block their action in the hydrolysis of the acetylcholine (ACh) neurotransmitter. ACh is the principal neurotransmitter at neuromuscular junctions in the parasympathetic nervous system and in many regions of the central nervous system. High concentrations of ACh are also found in areas of the brain linked to higher cognitive functions such as learning and memory.

Organophosphate compounds, such as acephate, chlorpyrifos, dimethoate, disulfoton, and ethion, bind and phosphorylate the active site of ChE, thereby inactivating the enzyme. Carbamates, including aldicarb, lannate, methomyl, propoxur, and carbaryl, also interact with the acetylcholinesterase (AChE) receptor by reversible carbamylation of the seryl hydroxyl moiety at the active site of the enzyme (Murphy, 1986).

Some organophosphate and carbamate insecticides are acutely toxic and are frequently implicated in poisonings of humans. Exposures to high levels of AChE-inhibiting compounds may lead to severe cholinergic toxicity, with symptoms of headache, nausea and vomiting, cramps, weakness, blurred vision, pinhole pupils, chest tightness, muscle spasms, and coma (Ecobichon, 1991). Delayed neuropathy has also been associated with exposure to some organophosphorus esters (i.e., phosphate, phosphorate, and phosphoramidate esters), some of which have been used as insecticides (Amdur et al., 1991). Symptoms of acute organophosphate toxicity are difficult to recognize in the clinical setting for two major reasons: the complaints are nonspecific, and most physicians have limited familiarity with the signs and symptoms of pesticide poisoning.

Most ChE inhibitors degrade relatively rapidly in the environment and do not appear to accumulate or concentrate in the food chain (in contrast to organochlorine pesticides). In addition, these pesticides do not accumulate in the body, since they are rapidly biotransformed and excreted. Nevertheless, ChE inhibition can occur, producing signs and symptoms of poisoning after exposure to small repeated doses. Long-term effects of acute and subchronic exposures to pesticides have been reported. Some investigators have reported chronic, subtle neurologic sequelae to acute organophosphate poisoning (Savage et al., 1988). Epidemiological literature reported by the Office of Technology Assessment provides some evidence of delayed, persistent, or latent effects in humans. The literature includes case reports and studies of agricultural workers with and without histories of acute poisoning (OTA, 1990).

Organophosphates and carbamates may be converted in the environment or in vivo to form metabolites with toxicity potentially greater than that of the parent compounds. Synergism among organophosphate com-

pounds, such as that demonstrated among malathion, *O*-ethyl *O*-*p*-nitrophenyl phenylphosphonothioate (EPN), and other organophosphates, may be an important variable to consider in assessing exposure to compound mixtures (NRC, 1977).

ChE inhibition is widely regarded as a good, general indicator of exposure to organophosphate pesticides except among people occupationally exposed over long periods who have developed persistently low active ChE levels. However, knowledge about this inhibitory effect is still incomplete. For example, the relationship between ChE inhibition and neurotoxicity has not been adequately demonstrated (EPA, 1988).

Neurotoxicity is defined by EPA as an adverse change in the structure or function of the nervous system following exposure to a chemical agent. The level at which ChE inhibition is associated with such changes is unclear (NRC, 1986). Furthermore, some investigators question the validity of measuring ChE inhibition in peripheral tissues, i.e., in plasma and blood, as a surrogate for measuring ChE inhibition of the central nervous system.

Further study is required to correlate ChE inhibition with identifiable changes in the central and peripheral nervous systems. Techniques for measuring neurotoxic effects include nerve conduction studies, sensory studies, evoked brain responses, electrocardiograms, and biochemical assays (NRC, 1992).

Relative Potency of Organophosphates

The EPA guidelines for the study of mixtures containing dioxins and dibenzofurans consider the relative potency of different components of the mixture (EPA, 1987). This method permits the estimation of toxicity equivalence factors (TEFs) by comparing the toxicity of the compounds of interest to a standard defined as the most thoroughly tested compound. In the present study, the committee selected as a standard chlorpyrifos—a commonly used organophosphate insecticide. A TEF may be derived by comparing the no-observed-effect level (NOEL), or lowest-effect level (LEL), for any other chemical shown to produce the same type of ChE-inhibiting effect, to the NOEL (or LEL) for chlorpyrifos. The ratio of the chlorpyrifos NOEL to the NOEL of a different chemical (chemical *X*) provides an estimate of relative potency for chemical *X* and was used to adjust the laboratory-detected residue levels of the five chemicals of concern (Tables 7-14 and 7-15). The new values based on relative potency may be added to estimate cumulative exposure to chemicals believed to induce similar adverse effects, in this case ChE inhibition.

The committee assumed for this example that ChE recovers to a normal level in a 24-hour period. This may not be an appropriate assumption, however, because intake data are summed over all eating occasions for

TABLE 7-14 Estimating Toxicity Equivalence Using the LOAEL for Chlorpyrifos as Reference Standard

Pesticide	LOAEL[a]	LOAEL Ratio = TEF
Acephate	0.12 mg/kg bw Rats, ChE inhibition	0.1/0.12 = 0.83
Chlorpyrifos	0.10 mg/kg bw Humans, ChE inhibition	0.1/0.10 = 1.0
Dimethoate	0.25 mg/kg bw Rats, ChE inhibition	0.1/0.25 = 0.40
Disulfoton	0.04 mg/kg bw Rats, ChE inhibition	0.1/0.04 = 2.5
Ethion	0.075 mg/kg bw Humans, ChE inhibition	0.1/0.075 = 1.33

NOTE: LOAEL, lowest-observed-adverse-effect level; TEF, toxicity equivalence factors.

[a] From Integrated Risk Information System (IRIS): EPA, July 1992.

TABLE 7-15 Estimates of Toxicity Equivalence Factors for Five Organophosphate Insecticides Using the NOAEL for Chlorpyrifos as the Reference Standard

Pesticide	NOAEL[a]	NOAEL Ratio = TEF
Acephate	NA	NA
Chlorpyrifos	0.03 mg/kg bw Humans, ChE inhibition	1
Dimethoate	0.05 mg/kg bw Rats, Che inhibition	0.03/0.05 = 0.6
Disulfoton	NA	NA
Ethion	0.05 mg/kg bw Humans, ChE inhibition	0.03/0.05 = 0.6

NOTE: NA, not applicable. NOAEL = no–observed–adverse–effect level.

[a] From Integrated Risk Information System (IRIS): EPA, July 1992.

each day. Primary exposure on day 1 may occur during dinner, whereas primary exposure for day 2 may come at breakfast. Although both meals fall within a 24-hour window, they are presumed to be different 24-hour windows for the purposes of the present example.

Food Consumption Data

Data from USDA's 1977-1978 NFCS were used in the analysis. As mentioned above, consumption rates of eight foods (apples, oranges, grapes, beans, tomatoes, lettuce, peaches, and peas) and three juices (apple, or-

ange, and grape) for 2-year-old children were selected for the analysis. There was a total of 1,831 person-days of data.

Residue Data

The committee used pooled FDA residue data from 1988 and 1989 compliance, surveillance, import, and domestic sampling. Residues for each chemical were converted to chlorpyrifos equivalents by multiplying each value by an equivalence ratio (chlorpyrifos LOAEL or NOEL / chemical X NOEL or LOAEL). For example, three chemicals allowed on a particular food item each have separate residue data sets consisting of individual sample results for each chemical detected on that food. Each of these data sets were summarized with regard to frequency and then for residue distribution.

To estimate cumulative exposure to the five organophosphate compounds, the committee adopted the assumption that the residue distributions for each compound are independent of one another. This approach may result in an overestimation of actual exposure, since there is likely to be some correlation among residue levels of the different compounds. In particular, substitution among chemicals would lead to scenarios in which all five compounds are never detected on the same sample.

The distribution of the cumulative exposure can be constructed by taking all possible combinations of chlorpyrifos equivalent values for the five chemicals and summing the values for each combination. This procedure will, however, yield large numbers of combinations and is thus impractical even on relatively large computers. A more practical alternative is to use strategic simulation in which the original shape of each component distribution is preserved. This is called a strategic simulation, since it is designed to reproduce the sample proportions for each subset of the original chemical distributions. The following procedure is used to create the distribution of cumulative exposure: a value is extracted randomly from each of the five residue distributions by using this strategic sampling technique; the resulting values are summed and the result recorded; and the procedure is repeated 5,000 times, creating 5,000 possible combinations across the five chemicals. The 5,000 summed residues form the distribution for cumulative exposure, which is expressed in chlorpyrifos equivalents.

This computerized simulation was conducted for each food, creating a single residue distribution that is the random summation of residue values strategically extracted from each of the distinct residue distributions. This final distribution of summed residue values is expressed in chlorpyrifos equivalents. Eight such distributions were created, one each for apples, oranges, grapes, beans, lettuce, tomatoes, peaches, and peas.

The committee used two assumptions for nondetected residues: (1) that they were zero and (2) that they were present at the LOQ. The LOQ data used by the committee in this exercise were provided by the FDA. Since the FDA does not record the LOQ for each sample tested, it estimated an average LOQ of 0.01 ppm for all chemicals and foods analyzed in this study and proposed that this value be used in the committee's analysis.

Exposure Analysis

The objective of this exposure analysis is to produce a distribution of possible person-day exposures based on the food consumption data for 2-year-old children, including 1,831 person-day intake values for eight foods and eight separate residue distributions representing cumulative exposure—one for each of the eight foods. Person-day exposures are estimated by applying the following method.

1. Intake of food 1 by person 1 on day 1 is multiplied by some randomly extracted value from the residue data set (specific to food 1). The result is stored as an exposure value.
2. The process is repeated for *n* foods, still for person 1 on day 1.
3. The exposure values derived from *n* foods for one person-day are summed.
4. Steps 1 through 3 are repeated 5,000 times by using the strategic simulation to extract the residue data points from each summarized food-specific residue distribution of the residue data.
5. The 5,000 exposure values for person 1 on day 1 are stored and summarized as counts within exposure intervals.
6. Steps 1 through 5 are repeated for 1,831 person-days, producing 9,155,000 person-day exposure values, all expressed in chlorpyrifos equivalents.
7. The counts within exposure intervals are plotted as a frequency distribution.
8. The proportion of the sample falling above the RfD is estimated. The RfD for chlorpyrifos is 0.003 mg/kg bw/day.

A second exposure analysis was conducted to determine the sensitivity of exposure estimates to assumptions regarding the transfer of residues from raw to processed foods. Exposure was estimated for the same eight foods as above, and then for three juices (i.e., apple, orange, and grape).

The committee assumed that all residues in apples, grapes, and oranges were transferred unchanged to their juices, consistent with EPA practice (Peterson and Associates, Inc., 1992). This provides a maximum exposure estimate that is useful in the absence of statistically reliable data on the effects of processing.

Summary

The results of this analysis using five pesticides and eight selected unprocessed foods, excluding potent ChE inhibitors, and assuming that nondetectable residues are actually equal to zero are shown in Figure 7-10. (Results with nondetects set equal to the detection limit of 0.01 ppm are similar and are excluded for simplicity of presentation.) On the basis of these results, the RfD of 0.003 mg/kg bw/day (3 μg/kg bw/day) would be exceeded on approximately 1.3% of the person-days for children considered, representing approximately 120,000 of the 9.1 million person-days simulated (Figure 7-10).

Conclusions regarding the at-risk population are more difficult to reach. If one assumes that these simulated person-day exposure values are an accurate estimation of daily exposure for this population, then one must also assume that the consumption and residue data are also accurate representations. The committee does not believe that the data used do accurately represent the current status of those pesticide residues on foods because of the age of the consumption data, sample sizes, and the methods used by the FDA in interpreting residue values beneath an "action" or legal tolerance level. However, these data sets are the best now available

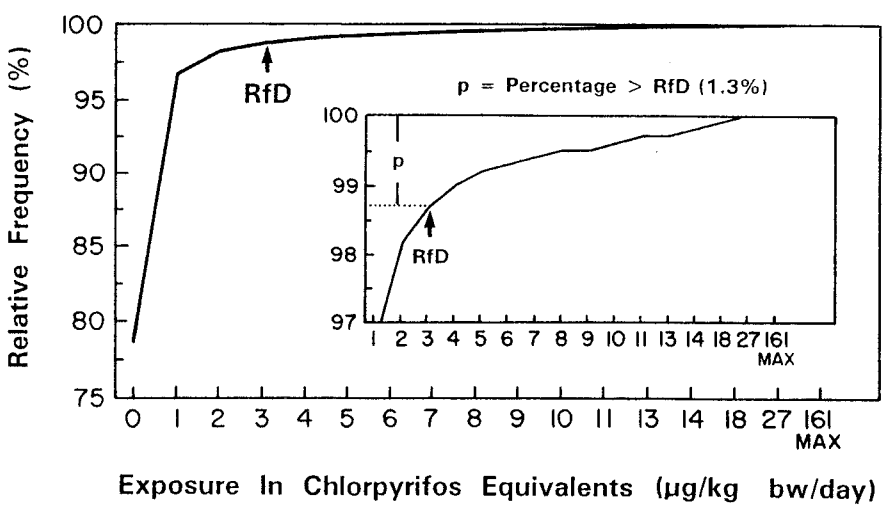


FIGURE 7-10 Exposure of 2-year-old children to organophosphate pesticides—1,831 2-year-old person-day food intake values. Foods: apples, oranges, grapes, beans, tomatoes, lettuce, peaches, and peas. Chemicals: acephate, chlorpyrifos, dimethoate, disulfoton, and ethion. Strategic simulation: 5,000 exposure values generated per person-day; cumulative distribution is summary of 9,115,000 simulated exposures.

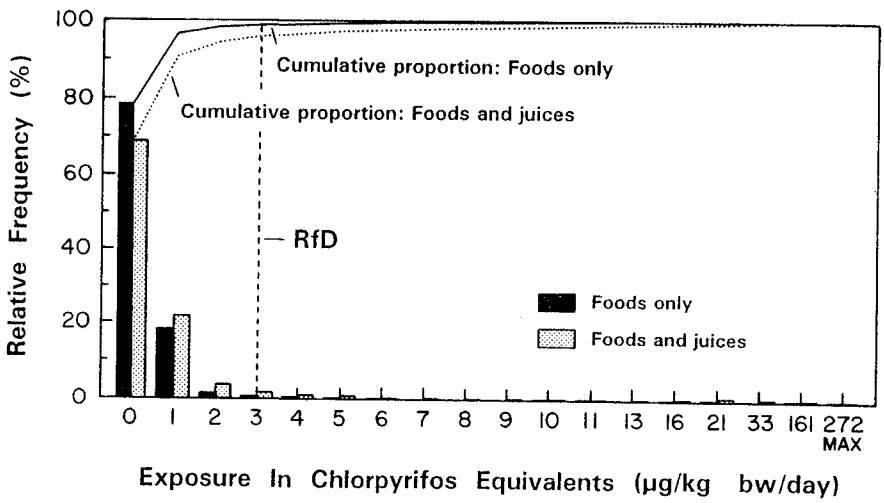


FIGURE 7-11 Exposure of 2-year-old children to organophosphate pesticides, including fruit juices. Strategic simulation: 5,000 exposure values generated per person-day; cumulative distribution is summary of 9,115,000 simulated exposures.

for a variety of chemicals and foods. (See Chapters 5 and 6 for an extensive examination of the limitations of the data.)

Although only 1.3% of the estimated person-day exposures were above the RfD, 3,584 person-days were more than ten times above the RfD, and the maximum exposure was ten times the RfD. These are clearly low probability events within a population of 3.5 million 2-year-old children; even a one-in-a-million event would occur 3.5 times per day. Even though these estimates are limited by the poor quality of the residue sampling data, they identify both a potential concern and an appropriate methodology for estimating exposure in large populations.

The results of the analysis are shown in Figure 7-11. The primary finding of this analysis is a shift in the distribution to higher residue levels. The percentage of the sample above the RfD rose from 1.3% to 4.1%, primarily because of the large intakes of apple juice and orange juice among 2-year-old children.

The committee concluded that this method is a workable and useful mechanism for assessing exposure, i.e., standardizing residue values by toxicity equivalents, combining these values based on either allowable or actual detected combinations of residues, and simulating exposures by combining residue values with actual reported intake values and summing exposures across foods within person-days. As with the other examples presented in this report, however, this discussion should be regarded as

an assessment of methodology rather than a specific attempt to characterize the proportion of children at risk. The method could also be used to study possible combinations of residues for any class of chemicals believed to have a common adverse effect, including cancer, where the end point of concern is not a site-specific tumor but, rather, the probability of a tumor occurring.

NONDIETARY EXPOSURE TO PESTICIDES

Although it was not generally within the committee's charge to examine exposures to pesticides by routes other than dietary, the committee wishes to point out that infants and children are subject to such exposures from a variety of sources. These sources should not be overlooked when attempting to estimate the total exposure of infants and children to pesticides and are therefore briefly summarized in this section.

In January 1990 EPA published the *Nonoccupational Pesticide Exposure Study* (NOPES). One of the study's primary objectives was to assess the relative contribution of each source to overall exposure to certain pesticides. Among their findings, the NOPES researchers concluded that (1) "house dust may be a source of exposure to pesticides via dermal contact, ingestion, and inhalation of suspended particulates, especially for infants and toddlers"; (2) "acute dermal exposures that occur during application events may contribute substantially to total exposure"; and (3) that, for the pesticides they examined, exposure from drinking water appeared to be minimal (EPA, 1990). Thus, exposure from all sources—not just ingestion—must be considered when estimating total exposure and risk to children.

Exposure via Parents

The child's first exposure to pesticides begins in utero, where chemicals may cross over the mother's placenta. Several studies have suggested an association between parental exposure (occupational and otherwise) to pesticides and childhood cancers. Researchers from the Children's Cancer Study Group (a cooperative clinical trials group with approximately 100 members and affiliate institutions in the United States and Canada) conducted a case-control study of occupational and household exposures of parents of 204 children with acute nonlymphoblastic leukemia (ANLL) (Buckley et al., 1989). Their most consistent finding was an association of ANLL risk when both mother and father had been exposed to pesticides.

Human birth defects possibly associated with prenatal occupational exposure to the organophosphate oxydemeton-methyl were published in 1989 by Romero et al. (1989). Gordon and Shy (1981) used ecologic data

to explore simultaneous maternal exposure to multiple agricultural chemicals in Iowa and Michigan and found an increased risk for facial clefts among offspring.

Lowengart and associates (1987) conducted a case-control study of leukemia patients 10 years and younger and the occupational and home exposures of their parents. With 123 matched pairs, a statistically significant association was seen between leukemia and the use of home and garden pesticides by either parent. In a cross-sectional study of 2,463 parents employed in the Central Valley area of California, limb reduction defects were observed to occur more frequently among the offspring of agricultural workers (relative risk of 2.3) at rates that were in excess of nationally established norms (Schwartz et al., 1986).

As is seen with lead and asbestos, children are at risk from toxicants that their parents unwittingly bring home on their clothes and other apparel. Children of parents employed in agricultural settings may be exposed to these "take-home" pesticides when the work clothing of their parents is washed with other family clothes (Formoli, 1990).

Exposure Through Air

Outdoor Air

In April 1990 the State of California canceled the permits for all uses of the soil fumigant 1,3-dichloropropene (commonly referred to as Telone). This action was prompted by an air monitoring program in the Central Valley that measured levels at a school in the area as high as 160 $\mu g/m^3$ (L. Baker and J. Behrmann, Air Resources Board, Sacramento, Calif., personal commun., 1990). Concentrations of 0.2 $\mu g/m^3$ of Telone in air are associated with cancer risks of 1 in 100,000 over 70 years, according to the EPA and the California Department of Health Services (CDHS).

Telone is usually applied undiluted to the soil around vegetable and tobacco crops to control nematodes and insects. Exposure to the vapor causes irritation of the mucosa and respiratory tract. The chemical is absorbed through intact skin, and systemic toxicity may follow cutaneous exposure as well as inhalation or ingestion of the compound (Flessel et al., 1978). Telone has been classified as a probable carcinogen in humans by the EPA (EPA, 1986) and is on the State of California's "Proposition 65" list of chemicals known to cause cancer.

Because pesticides are usually finely dispersed as droplets or particles at the time of application, aerial drift may cause them to be carried away from the target area where they were applied (Matsumura and Madhukar, 1984). Air sampled in areas where pesticides are used may contain residues either as vapors or bound to particles. Recent studies have found pesticide

residues suspended in fog. Respiratory absorption of chemicals tends to be more rapid than absorption through other routes of exposure, because of the abundant blood supply in and the thinness of the alveolar membrane (Matsumura and Madhukar, 1984).

Exposure to pesticide residues from ambient air sources is generally higher in areas close to agricultural lands and in communities surrounding pesticide manufacturing factories. Where urban and suburban developments are interspersed within agricultural lands, movement of the more volatile chemicals present potentially significant human exposure. Episodes of illnesses in communities near agricultural areas have been reported simultaneously with applications of the fumigants methyl bromide and chloropicrin (Murray et al., 1974; CDHS, 1980; Goldman et al., 1987).

Cotton defoliants have also been associated with a higher incidence of acute health symptoms among residents living near cotton fields. Tributyl phosphorotrithioate, trade name DEF, is a defoliant of primary concern from a public health perspective (Scarborough et al., 1989). DEF is quite stable and has been detected at very low concentrations in the ambient air of residential and urban areas of cotton-growing counties, even at times of the year when cotton is not being sprayed. However, formulation impurities of butyl mercaptan or dibutyl disulfite may be the causative agents rather than the parent defoliant. In a small northern California community, an increase in health symptoms such as headaches, runny nose, and asthma attacks was reported by residents living adjacent to a potato field that had been treated with the organophosphate pesticide ethoprop. Ethoprop, like DEF, releases a strong-smelling mercaptan gas (Ames and Stratton, 1991). Organophosphates and other commonly used pesticides have been detected in ambient air in California's Central Valley, but generally in such low concentrations that they are unlikely to contribute significantly to the total exposure of humans.

Arthur et al. (1976) measured the levels of several organophosphate and organochlorine pesticides in the Mississippi delta and found that the residue levels were the highest in August and September. In rural areas, where residue levels were higher than in urban areas, the highest concentrations were found where spraying was reported (Arthur et al., 1976).

Indoor Air

The widespread use of pesticides such as flea bombs and insecticide sprays and foggers in the home exposes children to pesticides in their indoor environment. Most home-use products contain either organophosphates or carbamates as their active ingredients, both of which are cholinesterase-inhibiting compounds. These compounds affect the nervous system and at low doses may cause a variety of cholinergic symptoms such

as drooling, excessive urination, or diarrhea (Berteau et al., 1989). For six pesticides (chlordane, heptachlor, aldrin, chlorpyrifos, diazinon, and gamma-BHC) analyzed in EPA's NOPES study, the mean air exposures were always or often higher than the estimated dietary exposure for the same compounds (EPA, 1990).

Fenske et al. (1990) measured chlorpyrifos (Dursban) concentrations following its application for flea treatment in a carpeted apartment. They found that the chlorpyrifos vapors measured in the infant breathing zone (25 cm above the carpet) were substantially higher than those measured in the sitting adult's breathing zone. Time-weighted averages for the 24-hour postapplication period in the infant breathing zone were 41.2 and 66.8 $\mu g/m^3$ for ventilated and nonventilated rooms, respectively; this is substantially higher than the interim guideline of 10 $\mu g/m^3$ proposed by the National Research Council's Committee on Toxicology for chlorpyrifos in indoor air following termiticide treatments (NRC, 1982). In addition, air concentrations increased from the time of application up to 5 to 7 hours later. The authors suggested that the treated carpet served as a source of volatilized chlorpyrifos and that although open windows provided mixing and dilution of air 1 m above the carpet, concentrations near the floor were affected much less (Fenske et al., 1990).

The short- and long-term health effects of exposure to commonly used home-use pesticide products are largely unknown. In assessing risk for infants in chlorpyrifos-treated homes, based on several conservative assumptions, Berteau et al. (1989) calculated an absorbed dose of 2.68 mg/kg. Fenske et al. (1990) found that the total estimated absorbed chlorpyrifos dose for infants exceeded the EPA's no-observed-effect level (NOEL) of 0.03 mg/kg/day in each case. The NOEL for chlorpyrifos is based on measurable changes in plasma acetylcholinesterase.

The indoor use of pesticides in public buildings such as schools and day-care centers leads to an additional source of exposure for children. In one episode, employees of a school for mentally handicapped children became ill within hours of entering a building that had been treated for roaches 3 days earlier and had not been ventilated. No students were admitted into the building until 14 days after the incident, when air levels of the pesticides used (dichlorvos and propoxur) had decreased to an acceptably safe level (White et al., 1987). An air analysis indicated that the levels of dichlorvos in the air were decreasing over time, but at a much slower rate than was expected from the data provided by the manufacturer.

Chlordane was the leading compound for controlling termites in homes for several years. Although it has since been canceled by the EPA for use as a termiticide, research demonstrates that chlordane air levels decline

very slowly with time (Menconi et al., 1988). In a cross-sectional epidemiological investigation of 85 chlordane-treated households containing a total of 261 people, investigators found a dose-response relationship between chlordane levels in home indoor air and incidence of migraines, sinusitis, and bronchitis (Menconi et al., 1988). Cases of more serious health effects such as neuroblastoma, acute leukemia, and aplastic anemia have been associated with exposure to chlordane (Infante et al., 1978).

Pentachlorophenol (PCP) is commonly used as a wood preservative and has become the second most heavily used pesticide in the United States (Cline et al., 1989). It is used in wood homes and on playground equipment. Studies indicate that PCP is virtually ubiquitous in the environment, and measurable residues of PCP are found in most people (Hill et al., 1989). Hill et al. (1989) found PCP in 100% of urine samples taken from 197 Arkansas children. Researchers at the Centers for Disease Control (CDC) found that mean serum PCP levels were 10 times higher in residents of PCP-treated log homes than in the controls (40 ppb compared to 420 ppb) and that serum levels for children in the log homes were "significantly higher" than those for their parents (Cline et al., 1989).

Exposure via Contaminated Surfaces

Home-Use Products

Indoor insecticide sprays and foggers may persist on carpets, floors, and other surfaces in the home. Young children, particularly those wearing only diapers, may be exposed playing on previously sprayed surfaces. In 1980 an 11-day-old infant suffered respiratory arrest in a hospital waiting room. Pesticide poisoning was suspected because tests showed his red blood cell cholinesterase levels to be depressed to 50% of normal low baseline levels. Around the time of his birth, the child's home had been treated with chlorpyrifos; the chemical was subsequently found on dish towels, food preparation surfaces, and the infant's clothing (Dunphy et al., 1980).

Pet Products

Flea control products may persist on the pet's fur and be transferred to children during contact with the animal. Flea control products commonly used in veterinary clinics, pet stores, and other commercial establishments include carbaryl, chlorfenvinphos, chlorpyrifos, dimethyldichlorovinyl phosphate (DDVP), fenthion, malathion, phosmet, and propoxur (Ames et al., 1989).

Playground Equipment

Wooden playground equipment is another source of pesticide exposure because of the various kinds of wood preservatives used to prevent microbial and insect attacks. A 1987 California survey estimated that approximately 20% of all wooden structures in parks were treated with chemical preservatives. Some wood preservatives—PCP, chromium, boric acid, creosote, and arsenic—can induce adverse skin reactions such as contact dermatitis, hyperkeratosis, and, in the extreme case, skin cancer (CDHS, 1987).

Exposure via Medications and Personal Products

Another important route of exposure involves the direct application of insect repellents and pediculocides to children's skin. These include such compounds as *N,N*-diethyl-*m*-toluamide, lindane, and malathion. Lanolin, used by some breastfeeding mothers on their nipples, is also a concern because of the pesticides it can contain.

N,N-Diethyl-*m*-toluamide

N,N-Diethyl-*m*-toluamide, commonly called Deet, is the active ingredient in numerous commercially available insect repellents. Although insect repellents can provide great personal benefit, rare adverse reactions can occur. Since 1961, at least six cases of systemic toxic reactions from repeated cutaneous exposure to Deet have been reported. Six girls, ranging in age from 17 months to 8 years, developed behavioral changes, ataxia, encephalopathy, seizures, and/or coma after repeated cutaneous exposure to Deet; three died (Oransky et al., 1989). Neurobehavioral analysis showed strong correlation between Deet exposure and affective symptoms, insomnia, muscle cramps, and urinary hesitation (McConnell et al., 1987).

In August 1989 the New York State Department of Health investigated five reports of generalized seizures temporally associated with topical use of Deet. Four of the patients were boys from 3 to 7 years old (Oransky et al., 1989).

Lindane and Malathion

For almost 30 years the pesticide lindane (a chlorinated hydrocarbon) has been used in a shampoo for the treatment of head lice (Taplin and Meinking, 1988). Concern has been raised about potential central nervous

system damage from exposure to lindane. In particular, cases of central nervous system toxicity have been reported from accidental ingestion as well as from single percutaneous exposures (Lee and Groth, 1977). One author reported two instances in which lindane lotion was given orally to children with scabies because of a lack of communication in one case and a language barrier in the other (Taplin and Meinking, 1988). Malathion has been recommended as a preferable treatment over lindane (Taplin et al., 1982; Fine, 1983).

Lanolin

Lanolin, a derivative of sheep's wool, is commonly used as an ointment to treat sore, cracked skin. Mothers who breastfeed frequently use it on their nipples, and it is sometimes applied directly to children's skin. The organophosphate pesticides diazinon and chlorpyrifos and several organochlorine pesticides such as dieldrin have been found at measurable levels in lanolin. The U.S. Food and Drug Administration identified 16 pesticides in lanolin it sampled in 1988. The principal source of these residues is the wool from sheep treated with a pesticide dip to control parasite infestations in the fleece (Cade, 1989). The fat-soluble organophosphate pesticide diazinon presented the greatest concern because of its frequent occurrence (21 of 25 samples) and the high levels identified (up to 29.2 ppm). (T. Levine, EPA, personal commun., 1988).

Occupational Exposures

In agricultural communities, children are often directly exposed to pesticides when they accompany their parents in the field or work there themselves (Pollack et al., 1990). In 1980, some 19 farm workers suffered organophosphate poisoning after working in a cauliflower field (Whorton and Obrinsky, 1983). Five of the workers were 18 years old or younger; three of those were between the ages of 9 and 15 years.

Exposure via Accidental Ingestion

Accidental poisonings are all too common among children. In one study of 37 children who had been hospitalized at Children's Medical Center in Dallas as a result of organophosphate or carbamate pesticide poisoning, ingestion of a liquid was the most common (73%) mechanism of exposure. Zwiener and Ginsburg (1988) reported that most poisonings took place in the home and were the result of careless storage of the original container or placement in unmarked or uncovered containers.

CONCLUSIONS AND RECOMMENDATIONS

Like other members of the general population, infants and children are exposed to pesticide residues in their diets. Estimation of dietary intakes requires information on both food consumption patterns and residue levels in food. The purpose of this chapter has been to demonstrate methods for estimating exposure to pesticides in the diet. The committee was guided by previous work on exposure estimation by the National Research Council (NRC, 1988, 1991a,b). Infants and children are also exposed to pesticides by nondietary routes, including air and contaminated surfaces such as rugs and playground equipment. Although a detailed analysis of nondietary routes of exposure to pesticides is outside the scope of this report, it is important for risk assessment purposes to consider the total exposure from all media. The following are the conclusions of the committee.

Conclusions

- Pesticide residues are present in the diets of infants and children. Estimation of dietary intakes of pesticides by infants and children requires information on both food consumption patterns and residue levels in food.

- Accurate estimation of dietary intake of pesticides by infants and children is difficult due to the limited amount of data on food consumption patterns of infants and children (Chapter 5) and limitations in the available data on pesticide residues (Chapter 6).

- Dietary exposures to pesticide residues can vary widely. Since most pesticide residues in foods are below the analytical limit of quantification, with comparatively few high residue levels, the distribution of dietary exposure to pesticides includes many low intakes. Some degree of positive skewness may be observed due to the occurrence of high consumption or high residue levels.

- To estimate dietary exposure to pesticides for infants and children, the committee combined probability distributions of food consumption with probability distributions of residue levels in order to obtain a probability distribution of individual exposures. The use of probability distributions for exposure assessment provides a more complete characterization of human exposure to pesticide residues in food than the use of summary statistics such as means or upper percentiles of exposure.

More accurate estimates of upper quantiles of the exposure distribution

can be obtained by pointwise multiplication of the residue and consumption distributions than by multiplying the quantiles obtained from the residue and consumption distributions, separately. Moreover, the probability distribution approach based on 1-year age groupings of children provides useful information on differences in exposure patterns for children 1 to 5 years of age.

- Average daily ingestion of pesticide residues is an appropriate measure of exposure for chronic risk assessment, whereas actual individual daily ingestion is more appropriate for acute risk assessment.

Since chronic toxicity is often related to long-term average exposure, the average daily dietary exposure to pesticide residues may be used as the basis for risk assessment with delayed irreversible chronic toxic effects. To take into account different food consumption patterns among individuals, the distribution of average daily dietary intake of pesticides should be examined within the population of interest. Since acute toxicity is more often mediated by peak exposures occurring within a short period (e.g., over the course of a day or even during a single eating occasion), individual daily intakes are of interest for risk assessment for acute toxic effects. Examination of the distribution of individual daily intakes for persons within the population of interest reflects both day-to-day variation in pesticide ingestion for specific individuals as well as variation among individuals. This distribution can be used to estimate the number of person-days in a given period during which intake will exceed a specified level, such as the acceptable daily intake (ADI), or reference dose (RfD).

- At present, there is a relatively limited amount of information on food consumption patterns of infants and children. To obtain accurate estimates of the distribution of individual intakes, more elaborate and more intensive consumption monitoring protocols are required.

- Because residue monitoring surveys conducted for compliance purposes are expected to lead to higher residue levels than those present in the general food supply, assessment of human exposure should normally be based on surveillance surveys. In using surveillance data, however, consideration needs to be given to regional differences in pesticide use and resultant residue levels.

The committee acknowledges that pesticide food surveillance data are generated by randomly sampling food items from the distribution system. The purpose of this sampling is to ensure agricultural compliance with acceptable pesticide use practices. This sampling is broad-based and often not focused only on pesticides actually used. Pesticide field trial data are generated under strictly controlled conditions of use. These data better reflect actual levels at the time of harvest when it is known that a specific

pesticide has been used. Each data source is used for purposes other than identifying actual dietary exposures, although both are useful in attempting to estimate these exposures.

- Frequently, the levels of pesticide residue in foods are below the analytical limit of quantification (LOQ). Since the actual residue level in such cases may lie anywhere between zero and the LOQ, there is some uncertainty about actual exposures in such cases.

For example, replacing the residue measurements below the LOQ with zero yields lower exposure estimates than substituting the LOQ for the unknown residue level. Notable differences occur when the analytical method is insensitive, the LOQ is high, or a large proportion of residues lies below the LOQ.

- The concentration of pesticide residues in foods may increase or decrease during food processing.

Changes in residue levels that occur during the processing of food are especially important in assessing the exposures of infants and young children, who consume large quantities of single processed foods, such as fruit juices, milk, and infant formula. In addition to the data accumulated by the food industry (Chapter 6), studies by pesticide manufacturers such as those furnished to the committee on the fate of residues during processing need to be conducted for most pesticides that produce detectable residues in food.

- Specific pesticides can be applied to more than one crop and, hence, appear on a number of food commodities. Residues of several pesticides may also appear on a single food commodity.

- Intake of multiple pesticides with a common acute toxic effect can be estimated by converting residues for each chemical to equivalent units of one of the compounds. The standardized residues can then be summed to estimate total residue levels in toxicity equivalence factors, and then combined with consumption data to construct a probability distribution of total exposure to all pesticides having a common mechanism of action.

Certain classes of pesticides such as cholinesterase inhibitors act by a common toxic mechanism. To properly evaluate the potential health effects of exposure to such pesticides, it is important to consider the total exposure to all pesticides in the class.

- Children are exposed to pesticides by nondietary routes.

Occupational exposure of the parent could result in exposure of the child in utero, in the home environment, or in the occupational setting of the parents. Pesticide residues have been detected in outdoor and indoor air, on contaminated surfaces, and in medications and personal products.

• When less than 100% of a given crop is treated with a particular pesticide, consideration might be given to adjusting exposure estimates according to the percentage of crop acreage treated. This adjustment can result in substantial reductions in estimates of exposure. This adjustment will be appropriate when the percentage of the crop treated is similar in different regions of the country, or when the crop is uniformly distributed throughout the country. Such adjustments should not be considered in the case of pesticides inducing acute toxic effects, since peak exposures are of importance in this case.

When these adjustments are used to adjust national data, they may result in averages that do not account for regional differences in pesticide use. It is therefore important that exposure estimates that have not been adjusted for acreage treated be presented and that such adjustments be critically examined.

Recommendations

The following recommendations were developed by the committee.

• Probability distributions based on actual data rather than simple summary statistics such as means or percentiles should be used to characterize human exposure to pesticide residues on food.

The advantage of using probability distributions rather than summary statistics to characterize exposure is that variation in individual food consumption patterns and residue levels in food are taken into account. This will require the collection of more detailed data on food consumption and residue levels as discussed in Chapters 5 and 6, respectively, but will provide more statistically robust estimates than the agency currently develops.

• The distribution of *average daily exposure* of individuals in the population of interest is recommended for use in chronic toxicity risk assessment; the distribution of *individual daily exposures* is recommended for evaluating acute toxic effects.

This recommendation is based on the committee's observation that chronic toxicity is typically related to long-term average exposure, whereas acute toxicity is more often mediated by peak exposures occurring within a short period, either over the course of a day or even during a single meal.

• If appropriately designed and conducted, surveillance studies of pesticide residues in food provide unbiased data on residue levels in food products. Field trials are also useful sources of information on pesticide residues in food. Such studies should be continued in order to expand the data base for evaluating dietary exposures to pesticides.

Surveillance studies based on random samples designed to provide a representative picture of residue levels in food are required to obtain unbiased information on dietary exposure to pesticides.

- The committee recommends that research to reduce the uncertainty in estimates of dietary exposure to pesticides be encouraged. Specifically, the development of improved analytical methods for residue analyses and statistical methods for imputing residue levels below the LOQ can lead to improved estimates of pesticide exposure.

All analytical methods for measuring pesticide residue levels in food are subject to an LOQ. Results below the LOQ may be as low as zero or as high as the LOQ itself, thereby imparting uncertainty regarding actual human exposure levels. This uncertainty will be reduced if more sensitive methods with lower LOQs are developed. Such technological improvements should be encouraged even in the absence of other pressures for more sensitive analytical methods.

Statistical methods for use with censored data (i.e., based on specific assumptions) can be used to impute residue levels below the LOQ, provided that the percentage of the residue data lying below the LOQ is not large. The use of such methods can reduce the uncertainty in resulting estimations of human exposure.

- When using multiresidue scans to detect different compounds in one scan of one food sample, all results should be recorded together. This will make possible more accurate evaluation of exposure distributions for multiple chemicals.

- The committee does not recommend the routine application of adjustments for the percentage of the crop treated in estimating dietary exposure to pesticides.

Adjustments for acreage treated are appropriate only under certain conditions. For example, such adjustments may be used when there is little regional variation in acreage treated, or when the crop is uniformly distributed at the national level.

- To determine total dietary exposure to a particular pesticide, intakes from all foods on which residues might be present need to be combined.

Many pesticides are approved for use on more than one crop. In addition, a single crop may be used in the production of a variety of processed foods. To estimate the total dietary exposure to a particular pesticide, it is important to consider the contribution of all foods on which residues might occur.

- To properly evaluate the potential risk from exposure to multiple pesticides with common mechanisms of action, it is necessary to develop

measures of total exposure to pesticides within the same class that reflect the overall toxicity of all pesticides combined.

Since the combined effect of pesticides acting by a common mechanism can be greater than the individual effect of any single pesticide, it is important to develop risk assessment methods that address the total risk from exposure to all pesticides within the same class. One possible approach is to establish toxicity equivalence factors based on no-observed-effect levels as was done for organophosphates in this chapter.

- Because infants and children are subject to nondietary sources of exposure to pesticides, it is important to consider total exposure to pesticides from all sources combined.

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EXHIBIT 3

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

Note to Reader

Background: As part of its effort to involve the public in the implementation of the Food Quality Protection Act of 1996 (FQPA), which is designed to ensure that the United States continues to have the safest and most abundant food supply. EPA is undertaking an effort to open public dockets on the organophosphate pesticides. These dockets will make available to all interested parties documents that were developed as part of the U.S. Environmental Protection Agency's process for making reregistration eligibility decisions and tolerance reassessments consistent with FQPA. The dockets include preliminary health assessments and, where available, ecological risk assessments conducted by EPA, rebuttals or corrections to the risk assessments submitted by chemical registrants, and the Agency's response to the registrants' submissions.

The analyses contained in this docket are preliminary in nature and represent the information available to EPA at the time they were prepared. Additional information may have been submitted to EPA which has not yet been incorporated into these analyses, and registrants or others may be developing relevant information. It's common and appropriate that new information and analyses will be used to revise and refine the evaluations contained in these dockets to make them more comprehensive and realistic. The Agency cautions against premature conclusions based on these preliminary assessments and against any use of information contained in these documents out of their full context. Throughout this process, If unacceptable risks are identified, EPA will act to reduce or eliminate the risks.

There is a 60 day comment period in which the public and all interested parties are invited to submit comments on the information in this docket. Comments should directly relate to this organophosphate and to the information and issues available in the information docket. Once the comment period closes, EPA will review all comments and revise the risk assessments, as necessary.

These preliminary risk assessments represent an early stage in the process by which EPA is evaluating the regulatory requirements applicable to existing pesticides. Through this opportunity for notice and comment, the Agency hopes to advance the openness and scientific soundness underpinning its decisions. This process is designed to assure that America continues to enjoy the safest and most abundant food supply. Through implementation of EPA's tolerance reassessment program under the Food Quality Protection Act, the food supply will become even safer. Leading health experts recommend that all people eat a wide variety of foods, including at least five servings of fruits and vegetables a day.

Note: This sheet is provided to help the reader understand how refined and developed the pesticide file is as of the date prepared, what if any changes have occurred recently, and what new information, if any, is expected to be included in the analysis before decisions are made. **It is not meant to be a summary of all current information regarding the chemical.** Rather, the sheet provides some context to better understand the substantive material in the docket (RED chapters, registrant rebuttals, Agency responses to rebuttals, etc.) for this pesticide.

Further, in some cases, differences may be noted between the RED chapters and the Agency's comprehensive reports on the hazard identification information and safety factors for all organophosphates. In these cases, information in the comprehensive reports is the most current and will, barring the submission of more data that the Agency finds useful, be used in the risk assessments.

Jack E. Housenger, Acting Director
Special Review and Reregistration Division

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

October 5, 1999

SUBJECT: Occupational/Residential Handler and Postapplication Residential Risk Assessment for Chlorpyrifos. DP Barcode: D259612. Case No. 818975. PC Code: 059101. Submission: S568580

FROM: Deborah Smegal, M.P.H./Risk Assessor
Re-Registration Branch 3
Health Effects Division (7509C)
Office of Pesticide Programs

and

Timothy Leighton, Environmental Health Scientist
Re-Registration Branch 4
Health Effects Division (7509C)
Office of Pesticide Programs

THRU: Steve Knizner, Branch Senior Scientist
Re-Registration Branch 3
Health Effects Division (7509C)
Office of Pesticide Programs

TO: Mark Hartman
Special Review and Reregistration Division (7508C)
Office of Pesticide Programs

EPA MRID Nos.: 40026001, 40094001, 43013501, 44167101, 44458201, 44444801, 44729401, 44729402, 44589001, 44739301

PHED: Yes, Version 1.1

EXECUTIVE SUMMARY

This document contains the occupational and residential exposure assessment for chlorpyrifos, resulting from the residential uses of chlorpyrifos products. Exposures are evaluated for occupationally-exposed Pest Control Operators (PCOs) and Lawn Care Operators (LCOs) at residential sites, residents who apply the chlorpyrifos products, and residential populations that may be exposed following pesticide application. Some products containing chlorpyrifos are intended primarily for homeowner use, while some are intended primarily or solely for PCO/LCO use. This memorandum addresses non-agricultural uses, focusing on residential sites. Agricultural, ornamental and animal premise uses are addressed elsewhere (memorandum from T. Leighton to D. Smegal, DP Barcode D259614, October 6, 1999).

Chlorpyrifos is an organophosphate insecticide used extensively in residential settings by both residents and PCOs. It is one of the top five insecticides used in residential settings. There are approximately 822 registered products containing chlorpyrifos on the market (REFs 9/14/99). Registered uses include a wide variety of food, turf and ornamental plants, as well as indoor product use, structural pest control, and in pet collars. It is used in residential and commercial buildings, schools, daycare centers, hotels, restaurants, hospitals, stores, warehouses, food manufacturing plants and vehicles. In addition, it is used as an adult mosquitocide. In 1998, Dow AgroSciences estimated that 70% of the urban chlorpryifos use involved termite control.

In June 1997, the registrants of chlorpyrifos voluntarily agreed to measures designed to reduce household exposure to chlorpyrifos, as part of a Risk Reduction Plan. This voluntary plan involved deletion of: indoor broadcast use, use as an additive to paint, direct application to pets (sprays, shampoos and dips), and indoor total-release foggers. The technical chlorpyrifos products have been amended to reflect the negotiated plan. The technical label limits end use product labeling to only those sites which are specified on its label. In addition, as part of this agreement, the registrants agreed to work with EPA to develop policies for a number of areas including:

- limiting household consumer use to only products packaged as ready-to-use;
- prohibiting use in inappropriate areas (e.g., toys, drapes, furniture);
- requiring PCOs to clean up spills and misapplications;
- requiring more training of PCOs and more supervision during application;
- reducing exposure by eliminating concentrates which require mixing;
- establishing specific protection measures for humans and pets during and immediately after application;
- revising labels to include appropriate intervals between treatment (e.g., to replace "use as necessary", currently on some labels);
- revising labels for safer termiticide and pet care products per PR notice 96-7 on all termiticide labeling and 96-6 on all pet care product labeling and support the Agency efforts to expedite these changes for other products; and
- accelerate education and training for PCOs on these measures to reduce risk and exposure, label improvements, and implementation of recent PR Notices 96-7 (for

termiticides) and 96-6 (for pet care products), and support the Agency efforts to expedite these changes for other products.

Chlorpyrifos, O,O-diethyl O-(3,5,6-trichloro-2-pyridyl) phosphorothioate, is an insecticide formulated as a wettable powder (containing 50% a.i.), emulsifiable concentrates (41.5-47%), dust (containing 0.1-7% a.i.), granular (containing 0.075%-2.5% a.i.), bait (containing 0.5% a.i.), flowables (containing 30% a.i.), impregnated material (containing 0.5-10% a.i.), pelleted/tableted (containing 0.5-1.0% a.i.), pressurized liquids (0.9-3.8% a.i.), and microencapsulated (0.5-20% a.i.). Dow AgroSciences states that formulations with concentrations greater than one pound a.i. per gallon (approximately 13% a.i.) are sold only to pest control or turf and ornamental professionals. Lower concentrations are available to homeowners from other suppliers for over-the-counter purchase. Except aerosols, granules and dusts, all formulated end-use products for application are diluted in water to a concentration of 1 percent a.i. or less (Dow AgroSciences 1998). However, HED is aware of at least one company that sells concentrated chlorpyrifos products (i.e., >13% up to 44.8% a.i.) to the public on the Internet (www.ADDR.com\~pestdepo\gizhome.html) as of September 15, 1999.

The toxicity endpoints used in this document to assess hazards include short-, intermediate- and long-term dermal and inhalation endpoints, and the acute oral endpoint. A route-specific short-term dermal no-observed adverse effect level (NOAEL) of 5 mg/kg/day from a 21-day dermal rat study has been identified based on plasma and red blood cell (RBC) cholinesterase (ChE) inhibition of 45% and 16%, respectively at 10 mg/kg/day (the lowest observed adverse effect level, LOAEL). Therefore, a dermal absorption adjustment is not necessary. The intermediate- and long-term dermal NOAEL is converted from an oral NOAEL of 0.03 mg/kg/day from a 2-year oral dog study using a 3 percent dermal absorption factor. Plasma and RBC ChE inhibition occurred in this study at a dose level of 0.1 mg/kg/day. Dermal absorption was estimated to be 3 percent based on the ratio of the oral lowest-observed-adverse effect level (LOAEL) of 0.3 mg/kg/day from the rat developmental neurotoxicity study (MRID Nos. 44556901, 44661001) to the dermal LOAEL of 10 mg/kg/day from the 21-day dermal study (MRID No. 40972801) for plasma and red blood cell cholinesterase inhibition. This absorption factor is comparable to the dermal absorption estimated from human data of 1-3% (MRID No. 00249203).

The short- and intermediate-term inhalation NOAEL is 0.1 mg/kg/day from two separate 90-day rat inhalation studies that did not observe effects at the highest dose tested. At higher oral doses of 0.3 mg/kg/day(LOAEL), 43% plasma and 41% RBC ChE were observed in animals. The lung absorption is assumed to be 100 percent or oral absorption. The long-term inhalation NOAEL is converted from an oral NOAEL of 0.03 mg/kg/day from the 2-year dog study, assuming that inhalation and oral absorption are equivalent. The acute oral NOAEL is 0.5 mg/kg/day from an acute oral rat study that observed 28-40% plasma cholinesterase inhibition 3-6 hours after dosing male rats with a single dose of 1 mg/kg/day (HIARC memorandum from D. Smegal to S. Knizner, March 4, 1999, Document number 013249). The acute oral NOAEL was used to assess short-term exposures resulting from incidental ingestion (i.e., hand to mouth exposures) of less than one week for children. This is considered appropriate because exposures and risks are calculated for the day of application, when residential exposures are expected to be greatest. Oral exposure was not evaluated for workers. The exposure duration for short-term assessments is 1 to 7 days.

Intermediate-term durations are 1 week to several months, and long-term exposures are durations greater than several months.

For dermal and inhalation risk assessment, risk estimates are expressed in terms of the Margin of Exposure (MOE), which is the ratio of the NOAEL selected for the risk assessment to the exposure. For occupationally exposed workers, MOEs > 100 (i.e., 10x uncertainty factor for interspecies extrapolation and 10x uncertainty factor for intraspecies variability) do not exceed HED's level of concern. For residential populations, MOEs > 300, which includes an additional 3x Food Quality Protection Act (FQPA) safety factor do not exceed HED's level of concern. The acute population adjusted dose (aPAD) used to assess short-term oral exposures is 0.0017 mg/kg/day, which is the acute oral NOAEL divided by an uncertainty factor of 300.

Multiple exposure studies were conducted by the registrant and submitted to the Agency that evaluate exposures to PCOs/LCOs/residential handlers and residents following application of chlorpyrifos products. These data include biological monitoring, passive dosimetry and environmental measurements. These data, along with the Pesticide Handlers Exposure Database (PHED) Version 1.1, were used to assess potential PCO/LCO exposures resulting from handling and applying chlorpyrifos in residential settings. Postapplication residential exposures were assessed using primarily the registrant-submitted data. In the absence of data, the Draft Standard Operating Procedures (SOPs) for Residential Exposure Assessments (December 18, 1997) were used to estimate exposures. Obviously, exposures associated with all uses of chlorpyrifos products have not been monitored. Therefore, the available data were used to evaluate similar uses (i.e., lawn studies used to evaluate yard and ornamental sprays, residential crack and crevice exposure data used to evaluate similar treatments in other buildings).

HED is in the process of revising the residential exposure assessment SOPs. This process may identify specific areas of further concern with respect to chlorpyrifos and exposure to the general population. For example, some of the secondary exposure pathways that EPA is currently addressing include exposures resulting from residue tracked into homes from outdoor use, indoor dust, and spray drift. In a recent study, polycyclic aromatic hydrocarbons (PAHs) that are abundant in house dust were shown to increase the toxicity of chlorpyrifos in vitro, particularly at low levels (i.e., 2-50 μM PAHs with 1-180 nM chlorpyrifos-oxon, a metabolite of chlorpyrifos that inhibits acetyl cholinesterase) (Jett et al. 1999). Currently, there are no SOPs available to evaluate these potential exposure pathways. These scenarios however, may be evaluated in the future pending revisions to the residential SOPs.

There is insufficient use information and exposure data to assess exposure resulting from use in vehicles (i.e., planes, trains, automobiles, buses, boats) and other current label uses such as treatment of indoor exposed wood surfaces, supermarkets, restaurants, theaters, furniture, and draperies. However, HED has concern for these uses based on the scenarios assessed within this document.

Risk and Uncertainty Characterization

Occupational/Residential Handler Risks

The following scenarios result in MOEs that exceed HED's level of concern (i.e., MOE less than 100 and 300 for occupational and residential pesticide handlers, respectively):

- Indoor Crack and Crevice Treatment by a PCO and residential applicator;
- Broadcast Turf Treatment by a LCO (intermediate and long-term applicator, mixer/loader) and short-term residential mixer/loader/applicator;
- Spot Treatment of Turf by a residential mixer/loader/applicator;
- Application of Insecticidal Dust Products by a PCO and residential applicator;
- Application of Granular Formulations by a LCO and residential applicator (by hand, belly grinder or push-type spreader);
- Termiticide Treatments for Pre-Construction by a PCO;
- Termiticide Treatments for Post-Construction by a PCO;
- Paintbrush Applications by a residential applicator; and
- Ornamental Application by a residential mixer/loader/applicator.

The following scenarios result in MOEs greater than 100 and 300 that do not exceed HED's level of concern for occupational or residential pesticide handlers, respectively:

- Ready-to-Use Formulated product (Ant Stop) containing 0.5% ai chlorpyrifos (residential handler), and
- Mixer/loader of lawn care products wearing PPE (LCO).

The results of the PCO/LCO handler assessment in residential settings for intermediate and/or long-term exposure scenarios indicate that most of the MOEs are less than 100, and therefore exceed HED's level of concern. The only intermediate-term scenario that results in a MOE consistently above 100 is lawn care professionals that wear PPE and mix and load lawn products (total MOEs 190-820). The majority of risks were estimated based on chemical-specific biomonitoring studies submitted by Dow AgroSciences (i.e., indoor crack and crevice treatment, broadcast turf application, and pre- and post-construction termiticide treatment) in which the PCOs wore label-specified personal protective equipment (PPE). Several of these studies did not apply the product at the maximum label rate, or only evaluated exposures for a few hours (i.e. 1-3 hours) of the work day, and consequently could underestimate exposures and risks to PCOs. Overall, the exposures and risks for LCOs/PCOs based on the chemical-specific biomonitoring studies are considered to be central tendency estimates because they evaluated less than a full day's exposure at the maximum label rate or they exclude accidental exposure (e.g., exposure resulting from a broken hose). In the absence of chemical-specific data, LCO/PCO exposures were estimated using data from PHED or the Draft Residential SOPs. The PHED data used for the mixer/loader for lawn treatment, and granular application (hand, belly grinder and push-type spreader) scenarios are representative of the chlorpyrifos uses as the surrogate data were monitored for the same uses.

The results of the residential handler assessment for short- term exposure scenarios indicate that eight of the nine scenarios evaluated have total MOEs that exceed HED’s level of concern

defined by a target MOE of 300. The only short-term scenario that results in a MOE above 300 is the use of a 0.5% ready-to-use formulated product. The residential handler MOEs ranged from 3 to 250 for dermal risk, from 120 to 14,700 for inhalation risk, and from 3 to 250 for total risk for the typical and maximum label-recommended use rates. For a number of scenarios, multiple evaluations were conducted using application rates less than the maximum label rate, or application using different equipment or methods (i.e., ornamental treatment via low pressure hand wand and hose-end sprayer, and granular application via hand, belly grinder and push-type spreader) to assist in risk mitigation and management decisions. MOEs for a few products evaluated at the minimum application rate were greater than 300 (i.e., crack and crevice spot treatment and ornamental application), and therefore do not exceed HED's level of concern. Due to an absence of chemical-specific homeowner applicator studies, the majority of residential applicator risks were estimated based on the data from the Draft Residential SOPs (i.e., indoor crack and crevice treatment, broadcast turf application, granular formulation application, paintbrush application, and treatment of ornamentals). In all cases, it was assumed that residents wore short pants, short sleeves, and no gloves, in accordance with current Agency policy. Only one of the residential handler scenarios was evaluated using chemical-specific data submitted by Dow AgroSciences.

Postapplication Residential Risks

The following scenarios result in MOEs less than 300 that exceed HED's level of concern:

- Broadcast Turf Treatment Using a Liquid or Granular Formulation;
- Yard Sprays;
- Indoor Crack and Crevice Treatment;
- Pet Collar Products; and
- Termiticide Treatments for Basement, Plenum and Slab Construction Homes (some of the MOEs for children exceed HED's level of concern).

While the following scenarios result in MOEs predominantly greater than 300 that do not exceed HED's level of concern for postapplication residential exposures:

- Aerial and ground-based fogger adult mosquitocide application; and
- Termiticide treatment (crawl space homes).

The results of the residential postapplication exposure scenarios indicate that seven of the eight scenarios evaluated have MOEs that are less than 300, and therefore exceed HED's level of concern. MOEs ranged from 7.5 to 3700 for total risk. The only scenario that resulted in a MOE consistently above 300 was from the aerial and ground-based fogger adult mosquitocide applications (MOEs are 2300 and 3600 for children and adults, respectively). The MOEs following termiticide treatment of crawlspace homes were above 300, however, treatment of other construction type homes for termites resulted in MOEs below 300 for children. The majority of residential postapplication risks were estimated based on chemical-specific studies submitted by Dow AgroSciences (i.e., crack and crevice treatment of the kitchen and bathroom, broadcast treatment of turf with chlorpyrifos spray and granules, and termiticide treatment). The

exposure and risk estimates based on the chemical-specific studies are considered to be reasonable estimates (i.e., arithmetic average exposure was used to calculate risk). Because these studies were conducted in adults, conservative assumptions were used to estimate child exposures. However, because adult activity patterns differ from children, i.e., hand-to-mouth activity, some of the registrant-submitted chemical-specific studies could under-estimate a child's exposure (e.g., lawn studies are not designed to reflect any potential for incidental ingestion of residues from treated turf, soil and/or granules). In the absence of chemical-specific data, exposures were estimated based on data from the Draft Residential SOPs (i.e., indoor crack and crevice treatment, and pet collar uses), which are considered to result in high-end risk estimates. Scientific literature studies, the AgDrift Model and the Draft Residential SOPs were used to evaluate adult mosquitocide uses.

No data are available to evaluate the postapplication residential exposures and risks associated with the use of insecticidal dust products indoors. In addition, there are no recommended procedures for evaluating these products in the Residential SOPs. Nevertheless, HED has concerns about the use of these products based on the low MOEs calculated using a study in the scientific literature for residents or workers that could apply these products. HED recommends that the registrant provide additional information on the potential postapplication residential exposures associated with these products.

1.0 INTRODUCTION

This document is organized as follows:

2.0 BACKGROUND

Purpose

This document evaluates the potential health effects of occupational and residential exposure to chlorpyrifos, resulting from the residential uses of chlorpyrifos products. Exposures are evaluated for occupationally-exposed Pest Control Operators (PCOs), Lawn Care Operators (LCOs) residents who apply the chlorpyrifos products, and residential populations that may be exposed following pesticide application. This information will be incorporated into the Chlorpyrifos Reregistration Eligibility Decision Document (RED).

Criteria for Conducting Exposure Assessments

An occupational and residential exposure assessment is required for an active ingredient if (1) certain toxicological criteria are triggered and (2) there is potential exposure during use or to persons entering treated sites after application is complete. Both criteria are met for chlorpyrifos.

Summary of Toxicological Endpoints

The Hazard Identification Committee memos, dated June 2, 1999 and March 4, 1999, indicate that there are toxicological endpoints of concern for chlorpyrifos. The endpoints, and associated uncertainty factors used in assessing the risks for chlorpyrifos are presented in Table 1.

Table 1
Chlorpyrifos Hazard Endpoints, Uncertainty Factors and MOEs

EXPOSURE SCENARIO	DOSE (mg/kg/day)	ENDPOINT	STUDY	MOE for Workers	MOE for Residents
Acute Dietary (oral)	NOAEL=0.5 UF = 100 FQPA = 3	plasma cholinesterase inhibition at peak time of inhibition (3-6 hours post exposure) at 1 mg/kg.	Blood Time Course Study	NR	300
Short-Term (Dermal)	Dermal NOAEL =5	Plasma and RBC cholinesterase inhibition of 45 and 16%, respectively at 10 mg/kg/day. (Dermal absorption factor not necessary)	21-day dermal rat study	100	300
Intermediate- and Long-Term (Dermal)	Oral NOAEL =0.03	Plasma and RBC cholinesterase inhibition at 0.1 mg/kg/day. (Use 3% dermal absorption)	2 year dog study	100	300
Short-,and Intermediate-Term (Inhalation)	Inhalation NOAEL= 0.1	Lack of effects in 2 rat inhalation studies at the highest dose tested. >40% plasma and >40% RBC cholinesterase inhibition following oral doses of 0.3 mg/kg/day (100% lung absorption assumed)	Two 90 day rat inhalation studies	100	300
Long-Term (Inhalation)	Oral NOAEL= 0.03	Plasma and RBC cholinesterase inhibition at 0.1 mg/kg/day (Assume inhalation and oral absorption equivalent)	2 year dog study	100	300

NR = Not Relevant
UF = Uncertainty Factor
MOE = Margin of Exposure
RBC = Red blood cell

As shown on Table 1, the short-term dermal NOAEL is 5 mg/kg/day from a 21-day dermal rat study, based on plasma and red blood cell (RBC) cholinesterase (ChE) inhibition of 45% and 16%, respectively at 10 mg/kg/day. Therefore, no dermal absorption factor adjustment is necessary. The intermediate- and long-term dermal NOAELs and long-term inhalation NOAEL are 0.03 mg/kg/day based on plasma and RBC ChE inhibition in a 2 year dog study. Because an oral NOAEL was selected, a dermal absorption factor of 3%, and a 100% default inhalation absorption factor (i.e., inhalation and oral absorption are equivalent) were used. Dermal absorption was estimated to be 3 percent based on the ratio of the oral lowest-observed-adverse

effect level (LOAEL) of 0.3 mg/kg/day from the rat developmental neurotoxicity study (MRID Nos. 44556901, 44661001) to the dermal LOAEL of 10 mg/kg/day from the 21-day dermal study (MRID No. 40972801) for plasma and red blood cell cholinesterase inhibition. This absorption factor is comparable to the dermal absorption estimated from human data of 1-3% (MRID No. 00249203).

The short- and intermediate-term inhalation NOAEL is 0.1 mg/kg/day based on lack of effects in two rat inhalation studies at the highest dose tested. At higher oral doses of 0.3 mg/kg/day >40% plasma and >40% RBC ChE were observed in animals. The acute oral NOAEL is 0.5 mg/kg/day from an acute oral rat study that observed 28-40% plasma cholinesterase inhibition 3-6 hours after dosing male rats with a single dose of 1 mg/kg/day (HIARC memorandum from D. Smegal to S. Knizner, March 4, 1999, Document number 013249). The acute oral NOAEL was used to assess short-term exposures resulting from incidental ingestion (i.e., hand to mouth exposure) of less than one week. This is considered appropriate because exposures and risks are calculated for the day of application, when residential exposures are expected to be greatest.

Summary of Use Pattern and Formulation

At this time some products containing chlorpyrifos are intended primarily for residential use, while some are intended primarily or solely for PCO/LCO use. Both occupational/PCO/LCO (non-agricultural) and residential use are evaluated in this document. Agricultural uses are addressed elsewhere.

Types of Pesticide/Targeted Pest/Use Sites

Chlorpyrifos is an organophosphate insecticide used extensively in residential settings by both residents and pest control operators (PCOs). It is one of the top five insecticides used in residential settings. There are approximately 822 registered products containing chlorpyrifos on the market (REFs 9/14/99). Registered uses include a wide variety of food, turf and ornamental plants, as well as indoor product uses, structural pest control, and in pet collars. It is used in residential and commercial buildings, schools, daycare centers, hotels, restaurants, hospitals, stores, warehouses, food manufacturing plants and vehicles. In addition, it is used as an adult mosquitocide. In 1998, Dow AgroSciences estimated that 70% of the urban chlorpryifos use involved termite control.

Formulation Types and Percent Active Ingredient

Chlorpyrifos, O,O-diethyl O-(3,5,6-trichloro-2-pyridyl) phosphorothioate, is an insecticide formulated as a wettable powder (containing 50% a.i.), emulsifiable concentrates (41.5-47%), dust (containing 0.1-7% a.i.), granular (containing 0.075%-2.5% a.i.), bait (containing 0.5% a.i.), flowables (containing 30% a.i.), impregnated material (containing 0.5-10% a.i.), pelleted/tableted (containing 0.5-1.0% a.i.), pressurized liquids (0.9-3.8% a.i.), and microencapsulated (0.5-20%

a.i.). Dow AgroSciences states that formulations with concentrations greater than one pound a.i. per gallon (approximately 13% a.i.) are only to pest control or turf and ornamental professionals. Lower concentrations are available to homeowners from other suppliers for over-the-counter purchase. Except aerosols, granules and dusts, all formulated end-use products for application are diluted in water to a concentration of 1 percent a.i. or less (Dow AgroSciences 1998). However, HED is aware of at least one company that sells concentrated chlorpyrifos products (i.e., >13% up to 44.8% a.i.) to the public on the Internet (www.ADDR.com\~pestdepo\gizhome.html) as of September 15, 1999.

Method and Types of Equipment Used for Mixing/Loading/Applying

- Handgun (LCO): Broadcast turf application
- Backpack/Low Pressure Handwand Equipment : crack and crevice treatment; spot treatment of turf; ornamental application
- Hose End Sprayer: Broadcast turf treatment, ornamental application
- Termite-injection equipment: subterranean termite control
- Belly-grinder equipment or a push type spreader: turfgrass
- Paintbrush: Treatment of infested wood

3.0 OCCUPATIONAL AND RESIDENTIAL EXPOSURE

3.1 Handler Exposures & Assumptions

EPA has determined that there is a potential exposure to mixers, loaders, applicators, or other handlers during usual residential use-patterns associated with chlorpyrifos. Based on the use patterns and potential exposures described above, 11 PCO/LCO/residential handler exposure scenarios were identified for chlorpyrifos.

Mixer/loader/applicator (M/L/A) exposure data for chlorpyrifos were required for a reregistration data call in (DCI) issued September 18, 1991 during the reregistration process, since one or more toxicological criteria had been triggered. Requirements for applicator exposure studies are addressed by Subdivision U of the Pesticide Assessment Guideline. Applicator exposure data were required previously by the Agency. The Pesticide Handlers Exposure Database (PHED), Version 1.1 was used for several scenarios. In addition, studies from the scientific literature were used for other situations.

The following studies monitoring PCO/LCO/residential application of chlorpyrifos were submitted by the registrant.

- MRID No./Accession No. 40026001. Vaccaro, J.R. (1986) Evaluation of Airborne and Whole Body Exposure of Lawn Care Specialists to Chlorpyrifos During Routine Treatment of Turf.

- MRID No. 44444801. Vaccaro, J.R. et al. (1997). Determination of Exposure and Dose of General Pest Control Operators to Chlorpyrifos during Routine Applications of

Dursban Pro® Insecticide to Crack/Crevices and Spots. November 25, 1997. Laboratory Project Study ID: HEH 785.

- MRID No. 44729401. Barnekow, D.E, and Shurdut, B.A. (1998). Evaluation of Workers' Exposure to Chlorpyrifos During the Use of Dursban Pro® Insecticide Concentrate for Broadcast Turf Applications. November 10, 1998. Laboratory Project Study ID: HEA 97089.

- MRID No. 44739301. Barnekow, D.E, Cook, W.L., Meitl, T.J., and Shurdut, B.A. (1999). Exposure to Chlorpyrifos Whilt Applying a Ready to Use Formulation. January 14, 1999. Laboratory Project Study ID: HEA 97046.

- MRID No. 44729402. Barnekow, D.E, and Shurdut, B.A. (1998). Evaluation of Workers' Exposures to Chlorpyrifos During the Use of Dursban® TC Termiticide Concentrate for Post-Construction Termiticide Applications. October 9, 1998 (original) and December 22, 1998 (amended). Laboratory Project Study ID: HEA 97054.

- MRID No. 44589001. Murphy, P.G., Beard, K.K., Chambers, D.M., Huff, D.W., Marino, T.A., Melichar, M., and Vaccaro, J.R. (1997). Evaluation of Workers' Exposures to Chlorpyrifos During the Use of Dursban® TC Termiticide Concentrate for Pre-Construction Termiticide Applications. December 15, 1997.

HED reviewed each of these studies and used the registrant-submitted data to estimate exposures to handlers/PCOs/LCOs applying chlorpyrifos-products in residential settings. A brief summary of each study is provided below, with reference to HED's memorandum that provides a more detailed review and analysis of the study. It should be noted that a number of the registrant-submitted studies conducted biomonitoring by measuring urinary concentrations of the primary chlorpyrifos metabolite 3,5,6-trichloro-2-pyridinol (3,5,6-TCP), to estimate chlorpyrifos exposures. Prior to the studies, baseline urinary 3,5,6-TCP concentrations were determined in the study volunteers, and these baseline measurements were subtracted from the exposure-related 3,5,6-TCP concentrations measured in the biomonitoring study. It is important to note that most individuals in the U.S., and nearly all the subjects in the Dow AgroSciences biomonitoring studies had low levels of urinary 3,5,6-TCP prior to study commencement, indicating a baseline exposure to chlorpyrifos, chlorpyrifos methyl or their metabolite 3,5,6-TCP, which most likely is attributed dietary sources.

In the absence of chemical-specific monitoring data, data obtained from PHED Version 1.1 were used to assess handler exposures for regulatory actions. PHED was designed by a task force of representatives from the U.S. EPA, Health Canada, the California Department of Pesticide Regulation, and member companies of the American Crop Protection Association. PHED is a software system consisting of two parts--a database of measured exposure values for workers involved in the handling of pesticides under actual field conditions and a set of computer algorithms used to subset and statistically summarize the selected data. Currently, the database contains values for over 1,700 monitored individuals (i.e., replicates).

Users select criteria to subset the PHED database to reflect the exposure scenario being

evaluated. The subsetting algorithms in PHED are based on the central assumption that the magnitude of handler exposures to pesticides are primarily a function of activity (e.g., mixing/loading, applying), formulation type (e.g., wettable powders, granulars), application method (e.g., aerial, groundboom), and clothing scenario (e.g., gloves, double layer clothing).

Once the data for a given exposure scenario have been selected, the data are normalized (i.e., divided by) by the amount of pesticide handled resulting in standard unit exposures (milligrams of exposure per pound of active ingredient handled). Following normalization, the data are statistically summarized. The distribution of exposure values for each body part (e.g., chest, upper arm) is categorized as normal, lognormal, or "other" (i.e., neither normal or lognormal). A central tendency value is then selected from the distribution of the exposure values for each body part. These values are the arithmetic mean for normal distributions, the geometric mean for lognormal distributions, and the median for all "other" distributions. Once selected, the central tendency values for each body part are composited into a "best fit" exposure value representing the entire body.

The unit exposure values calculated by PHED generally range from the geometric mean to the median of the selected data set. To add consistency and quality control to the values produced from this system, the PHED Task Force has evaluated all data within the system and has developed a set of grading criteria to characterize the quality of the original study data. The assessment of the data quality is based on a number of observations and the available quality control data. While data from PHED provide the best available information on handler exposures, it should be noted that some aspects of the included studies (e.g., duration, acres treated, pounds of active ingredient handled) may not accurately represent labeled uses in all cases. HED has developed a series of tables of standard unit exposure values for many occupational scenarios that can be utilized to ensure consistency in exposure assessments. This surrogate exposure guide serves as the basis for this assessment. Best available grades are assigned to the unit exposures as follows: matrices with grades A and B data and a minimum of 15 replicates; if not available, then grades A, B and C data and a minimum of 15 replicates; if not available, then all data regardless of the quality and number of replicates. Data confidence are assigned as follows:

High	=	grades A and B and 15 or more replicates per body part;
Medium	=	grades A, B, and C and 15 or more replicates per body part; and
Low	=	grades A, B,C, D and E or any combination of grades with less than 15 replicates.

There are three basic risk mitigation approaches considered appropriate for controlling occupational exposures. These include the use of engineering controls, administrative controls, and the use of personal protective equipment (PPE). Engineering controls are recommended for occupational hazards wherever feasible, because they have the least continual human implementation or intervention necessary in achieving decreased exposure levels. Occupational handler exposure assessments are typically completed by HED using a baseline exposure scenario and, if required, increasing levels of risk mitigation (PPE and engineering controls) to achieve an appropriate margin of exposure. The baseline clothing/PPE ensemble for occupational exposure scenarios is generally an individual wearing long pants, a long-sleeved shirt, no chemical-resistant gloves (there are exceptions pertaining to the use of chemically-resistant gloves, footwear and

aprons and these are noted), and no respirator. The first level of mitigation generally applied is PPE. As reflected in the calculations that follow, PPE may involve the use of an additional layer of clothing, chemical-resistant gloves, and/or a respirator. The next level of mitigation considered in assessing exposure and risk is the use of appropriate engineering controls which, by design, attempt to reduce or eliminate the potential exposure. Examples of commonly used engineering controls include enclosed tractor cabs, closed mixing/loading/transfer systems, and water-soluble packets. [Note: Administrative controls may include methods such as lowering application rates for handler exposure scenarios.]

For chlorpyrifos, a typical baseline scenario was not evaluated for PCOs/LCOs because it was assumed they would wear the label-specified PPE.

Occupational/Residential Handler Exposure/Risk Assessment

The following 11 PCO/LCO/residential application scenarios were considered:

(1) Indoor Crack and Crevice or Spot Application

Commercial Applicator (MRID No. 44444801)

The registrant submitted a study that characterizes exposures to professional pest control operators (PCO) during application of 0.29% Dursban Pro® (EPA Reg No. 62719-166) on cracks, crevices, and spot treatment of residential and commercial buildings. The equipment used for spraying the product was a 2-gallon, hand pressurized B&G sprayer. A total of ten professional male PCOs from three state-wide and local pest control companies were evaluated. Five of the ten volunteers performed a second replicate for a total of fifteen replicates. Each volunteer was dressed in long cotton underwear, a cotton overall with long sleeves and long pant legs, cotton socks, chemically-resistant shoes and protective gloves during the mixing process. Eye protection was used by the PCOs when chlorpyrifos was sprayed overhead. HED evaluated this study in DP Barcode 241777 and D241838 (Memorandum from D. Smegal to M. Hartman, April 19, 1999).

Dermal exposure was quantified using passive dosimetry (long cotton underwear, cotton coveralls with long sleeves and long pant legs, and cotton socks; hand washes; and head patches). Inhalation exposure was measured using a personal air pump attached to the test subject's belt. The pump was connected with a cassette containing a polyvinyl chloride filter and a cellulose support pad (37-mm diameter, 0.8-µm pore size) followed by a Chromosorb 102 vapor collection tube to evaluate inhalation exposures in the breathing zone of workers.

The amount of active ingredient (ai) handled per replicate ranged from 0.09 g to 31.04 g (mean = 9.20 g; S.D. = 9.77 g). The volume applied per replicate ranged from 0.02 gallons to 2.8 gallons (mean = 0.84 gal.; S.D. = 0.81 gal.). The sampling time per replicate ranged from 248 to 591 minutes (mean = 378 minutes). Of the sampling time, 2.3 percent (12 minutes) to 43 percent (154 minutes) was used for actual spraying activities (mean = 21 percent, or 76 minutes).

The data were used to estimate dermal and inhalation unit exposures (μg/ lb ai) based on the

worker-specific amount handled (lb ai) per day, and the worker-specific total dermal or inhalation exposure based on the dosimetry measurements. The mean dermal and inhalation unit exposures were then used to calculate the total dermal and inhalation doses for three scenarios (average, minimum and maximum) based on the range of chlorpyrifos (lb ai) handled by the PCOs during the 15 replicates. The amount (lb ai) handled per worker varied significantly and ranged from 0.0002 to 0.0684 lb ai, with a mean of 0.02 lb ai.

A summary of the dermal and inhalation dose estimates are presented on Table 2. Because dermal and inhalation unit exposure data sets are lognormally distributed, the current HED policy is to use the geometric mean for assessing exposure. As shown on Table 2, the total dermal absorbed dose ranges from 0.005 to 1.75 μg/kg/day, with a geometric mean of 0.51 μg/kg/day. The dose estimates resulting from inhalation range from 0.0015 to 0.52 μg/kg/day, with a geometric mean of 0.15 μg/kg/day. This study demonstrates that on average 71% of the total exposure to PCOs during crack and crevice treatment results from dermal exposure, while inhalation exposure contributes on average approximately 29% of the total dose. The dose estimates from this study were used to assess long-term exposures to a PCO.

The exposure data partially meet the criteria specified in Subdivision U (currently referred to as Series 875 Group A). There is a large variation in the results, due primarily to the large range of chlorpyrifos ai handled (0.09 to 31.04 g), volume applied per replicate (0.02 to 2.8 gallons), sampling time (248 to 591 minutes or 4 to 9.85 hours), spray time (12 to 154 min) and percent chlorpyrifos handled (0.05 to 0.53%). In fact, only two of the fifteen replicates reflect the maximum recommended label concentration of 0.5% chlorpyrifos; an average of 0.29% chlorpyrifos was handled by the fifteen PCOs. In addition, it is possible that different tasks/activities associated with pesticide application in residential and commercial locations contributed to the range of exposures. However, the impact of applicator activities can not be determined due to an absence of study details. Despite the limitations, the data collected in this study are of sufficient scientific quality to be used in the RED document.

Residential Application

In the absence of chemical-specific data, short-term doses to residents that could treat their homes with a crack and crevice product in an aerosol can were evaluated using data from PHED V1.1, and the Residential SOPs. It was assumed that a residential applicator would wear short-sleeves, short pants and no gloves, that an average applicator weighs 70 kg, and applies the entire contents of a 16 ounce aerosol can that contains 1% ai chlorpyrifos (w/w, 0.16 oz or 4.5 g) (EPA Reg. 026693-00003) as a high end estimate for a heavy infestation, and the application of a 16 oz can of a 0.5% ai chlorpyrifos (EPA Reg 239-2619) to represent more typical homeowner use. In addition, an assessment was conducted for a spot treatment, where a homeowner could apply 2 oz of a 0.5% ai product. The estimated doses are presented in Table 2. There is medium confidence in the dermal and inhalation unit exposure estimates from PHED, which are based on 30 dermal replicates of ABC grades, 15 hand replicates of grade A, and 30 inhalation replicates of grade ABC. The representativeness of the PHED data are excellent, as the surrogate study monitored exposures resulting from an insecticide aerosol can while treating baseboards in a kitchen.

(2) Broadcast Turf Application (MRID No. 44729401)

LCO Applicator Exposures

Exposure estimates were derived from a chemical-specific Dow AgroSciences study in which workers were monitored during commercial lawn care application. HED evaluated this study in DP Barcode D252357 (Memorandum from D. Smegal to M. Hartman, April 15, 1999). This study characterizes exposures to lawn care operators (LCOs) that apply an average of 183 gallons of 0.12 percent Dursban Pro (EPA Reg No. 62719-166) by broadcast applications to turf for an average of 6 hours (range of 4.4-8.2 hours). Exposures were estimated based on both dosimetry measurements and biomonitoring of urinary 3,5,6-TCP (the primary metabolite of chlorpyrifos). The study examined exposures to 15 lawn care insecticide applicators from two different companies in Ohio, that each treated 11-15 turf blocks (one block equals approximately 6,500 ft^2). The total area of treated turf ranged from 74,740 to 97,500 square feet (mean of 95,983 ft^2), while the total amount of chlorpyrifos handled ranged from 1.57 to 2.95 lb ai chlorpyrifos (mean of 2.17 lb ai). In addition, the workers unloaded and reloaded the hose following application to each lawn (i.e., repeated 15 times per replicate). This study does not characterize exposures associated with mixing and loading the insecticide. It was assumed that lawn care professionals could treat lawns for both intermediate and long term durations.

Each LCO wore pre-laundered cotton coveralls, a pre-laundered cotton socks, cotton briefs, and cotton T-shirts (undergarment); and a hat with affixed denim patches. At the end of the application, these dosimeters were collected from each applicator. The coverall and undergarments were sectioned into pieces representing arm, leg, and torso regions. Patches were affixed to the hat to serve as a surrogate for face, head and neck exposure. In addition, each LCO wore chemically-resistant nitrile gloves and knee high chemically-resistant boots (note that knee-high boots are not required by the label).

The total absorbed doses estimated from dosimetry range from 0.21 to 2.24 µg/kg/day, with a mean of 0.88±0.62 µg/kg/day. Approximately 33 percent of the absorbed doses resulted from inhalation and 67 percent from dermal exposure. The total absorbed dose estimated from biomonitoring ranged from 0 to 4.84 µg/kg/day, with an arithmetic mean of 0.65 ± 1.43 µg/kg/day (this average includes seven of the 15 workers that had exposures of zero because the exposure contribution from the application could not be distinguished from the high baseline chlorpyrifos exposure based on pre-study urinary 3,5,6-TCP concentrations). The geometric mean dose for workers who had exposure above baseline levels (n=8) is 0.4 µg/kg/day. In accordance with HED policy, the geometric mean is used to assess exposures because the biomonitoring data are lognormally distributed. The mean values are in somewhat good agreement with the estimates from dosimetry. The biomonitoring arithmetic average for the eight workers who had exposures above baseline was 1.23 µg/kg/day (i.e., excludes the seven workers with no exposure from lawn treatment). The registrant speculated that the highest exposure of 4.84 µg/kg (for OH05) was from a secondary source because 67% of the 3,5,6-TCP was excreted on day 5 post exposure. However, this value was included in the average dose because each volunteer was instructed to avoid chlorpyrifos for 10 days prior and 5 days following the study.

Pre-exposure baseline chlorpyrifos doses ranged from 0.2 to 3.73 µg/kg with a mean of 1.54 µg/kg, despite the fact that workers were instructed to avoid chlorpyrifos exposure 10 days prior to the study initiation. The high baseline chlorpyrifos dose makes it difficult to interpret the biomonitoring results. For example, seven of the fifteen workers had exposure levels (based on urinary 3,5,6-TCP) less than baseline levels, and therefore, their exposure from broadcast turf application is probably within the seven worker-specific baseline range (0.94 to 3.73 µg/kg), and not zero as concluded by the registrant.

The analysis of blood samples drawn from each applicator 24 and 48 hours post exposure indicated that no significant depression in plasma and red blood cell cholinesterase activity, relative to pre-study activity levels, occurred to the applicators after the application of the Dursban Pro insecticide. All of the plasma and red blood cell cholinesterase activities were within the reference range for the laboratory of 1,000 to 3,500 and 5,300 to 10,000 international units (IU)/ liter (L), respectively except for the plasma pre-exposure level for volunteer OH15 (352 IU/L). It should be noted, however, that in animals peak cholinesterase inhibition occurs 3-6 hours post exposure. In addition, the prior exposure of many of these PCOs may have resulted in suppressed baseline cholinesterase levels.

The lower leg (calves) coverall samples contained approximately 80% of the total coverall chlorpyrifos, despite that only 9% of the dermal dose was attributed to the sock dosimeters. However, it should be noted that each worker wore knee high chemical resistant footwear during application (only chemical resistant footware is required by the label, not knee high footwear). In addition, the exposure from hand washes represented 11% of the total dermal exposure, despite the fact that each worker wore chemically-resistant gloves.

The majority of the exposure data meet the criteria specified in Series 875 Group A. The applications used in this study represented 50% of the maximum rate for treatment of subsurface feeding insects. For example, the study applied 0.12% ai at 2 gallons/1000 ft^2, while the label allows up to approximately 0.12% ai at 4 gallons/1000 ft^2. Therefore, it is possible that this study underestimates the actual exposures to LCOs that apply the maximum label rate for subsurface soil broadcast treatment. For comparison purposes, dose estimates were also calculated based on the adjusted flow rate of 4 gallons/1000 ft^2, as shown on Table 2. The flow-rate adjusted dose estimates are two times higher than the estimated biomonitoring exposures, with a geometric mean of 0.8 μg/kg/day.

LCO Mixer/Loader Exposures

Because the biomonitoring study did not evaluate exposures for mixer and loading activities, these scenarios were evaluated using PHED V1.1. Two unit exposures for a mixer/loader handling liquid were evaluated and are presented in Table 2. One for a single layer of clothing and gloves, and the second for two layers of clothing and gloves. There is high confidence in the dermal and inhalation unit exposure estimates from PHED.

Residential Application

HED has no data monitoring chlorpyrifos exposures to residents during broadcast or spot treatment of turf. Therefore, exposures were evaluated based on data obtained from the Residential SOPs (also from PHED V1.1) for mixing/loading and application activities. This assessment evaluates both the broadcast and spot treatment of turf, which are assumed to be short-term scenarios for residents. For the broadcast treatment, it was assumed that a resident would use a hose end sprayer to treat 0.5 acre/day of turf, which represents the mean to upper-percentile range of the distribution of lawn size, with Dursban 1-12 Insecticide (EPA Reg No. 62719-56; 12.6% ai; 1 lb ai/gallon). For spot treatment of turf, it was assumed that a resident would use a low pressure handwand to treat 1000 ft^2 with the same chlorpryifos product. The dose estimates for residential use assume that individuals wear short pants, short sleeves and no gloves. For the hose-end sprayer, there is low confidence in dermal and inhalation unit exposure

estimates, which are based on 8 dermal and inhalation replicates of C grade data, and 8 grade E hand replicates. For the low pressure handwand (liquid/open pour), there is low confidence in dermal unit exposure estimates, which are based on 9-80 dermal replicates of ABC grade data, and 70 hand replicates of all grades. There is medium confidence in the inhalation unit exposure estimates, which are based on 80 inhalation replicates of ABC grade data. The label recommends diluting 3-12 oz of Dursban 1-12 Insecticide (12.6% ai; 1 lb ai/gallon) with1 to 3 gallons of water. As shown on Table 2, a range of dose estimates were calculated for broadcast treatment, assuming application at both the minimum and maximum dilution rates of 3 to12 oz/gallon/ water/ 1000 ft^2. The short-term dermal doses (not adjusted for absorption) range from 214 to 857 μg/kg, while the inhalation exposures range from 0.07 to 0.27 μg/kg/day. For spot treatment, the maximum application rate of 12 oz ai/gallon water 1000 ft^2 resulted in short-term dermal and inhalation doses of 134 and 0.04 μg/kg/day, respectively. These short-term dermal and inhalation dose estimates are presented on Table 2.

(3) Application of a Ready-To-Use Formulated Product (MRID No. 44739301)

Exposure estimates were derived from a chemical-specific registrant-submitted study in which 15 homeowners were monitored during the application of a ready-to-use formulated product, Ortho Ant Stop containing approximately 0.5% chlorpyrifos. HED evaluated this study in DP Barcode D252738 (Memorandum from D. Smegal to M. Hartman, April 29, 1999). In this study, homeowners applied five 24 oz. ready-to-use disposable bottles (with screw on tops) over a one hour duration to the outside foundation and perimeter of the house, and other areas (e.g., flower beds) where ants were present. A total of fifteen adult volunteers (nine females and six males) in the area of Indianapolis, Indiana were evaluated. The volunteers wore standard clothing that consisted of a short-sleeve coveralls with long pants, underwear, and a baseball style hat, but no gloves. Volunteers wore their own uncontaminated shoes. Each volunteer was instructed not to treat their homes or yards with chlorpyrifos containing products either immediately before, during or after the conduct of the study, and to avoid chlorpyrifos-containing products 10 days prior and 4 days after application. The amount of active ingredient (ai) handled per replicate ranged from 0.015 g to 0.038 g (mean = 0.033 g; S.D. = 0.006 g).

Exposures were estimated based on both dosimetry measurements and biomonitoring of urinary 3,5,6-TCP. Dermal exposure was quantified using passive dosimetry [cotton underwear (T-shirt, briefs or women's underwear), short-sleeve cotton coveralls with long pant legs, and hand washes; and a baseball style hat]. Inhalation exposure was measured using a personal air pump attached to the test subject's belt. The pump was connected by tygon tubing with a 37-mm mixed cellulose ester filter (0.8-µm pore size) connected to a Chromosorb 102 vapor collection tube to evaluate inhalation exposures in the breathing zone of volunteers.

The total absorbed dose estimated from passive dosimetry range from 0.03 to 0.86 µg/kg/day, with a mean of 0.25±0.25 µg/kg/day. Approximately 12 percent of the absorbed dose, as estimated from the passive dosimetry data, resulted from inhalation (mean 0.03 µg/kg/day) and 88 percent from dermal exposure (0.23 µg/kg/day). The total absorbed dose estimated from biomonitoring ranged from 0 to 1.9 µg/kg/day, with an arithmetic mean of 0.49 ± 0.59 µg/kg/day, and a geometric mean of 0.24 µg/kg/day. The mean values are in somewhat good agreement with the estimates from dosimetry. The biomonitoring results are slightly higher, but given that hand wash residues contribute on average 57% of the total dermal exposure, it is possible that the volunteers may have incidentally ingested chlorpyrifos as well (which would only be captured in

the biomonitoring results). Baseline chlorpyrifos pre-exposure ranged from 0.05 to 0.3 µg/kg with a mean of 0.12 µg/kg, despite the fact that volunteers were instructed to avoid chlorpyrifos exposure 10 days prior to the study initiation.

The geometric mean biomonitoring dose estimate of 0.24 µg/kg/day is used in this risk assessment in accordance with HED policy for lognormally distributed data sets. This dose estimate was divided into dermal and inhalation doses based on the passive dosimetry results, (i.e., 88% dermal and 12% inhalation), because there are different short-term inhalation and dermal endpoints for risk assessment. The resulting absorbed dose estimates used in the risk assessment are 0.029 µg/kg/day for inhalation and 0.21 µg/kg/day for dermal, as shown on Table 2. For short-term scenarios (such as residents), the absorbed dermal dose estimate from the biomonitoring results (absorbed dose) was further adjusted to an estimated dermal non-absorbed dose of 7 µg/kg/day (using a 3% dermal absorption factor) for direct comparison with the short-term dermal toxicity endpoint. These dose estimates represent a central-tendency to high-end scenario for residential applicators, who are more likely to apply one can of product rather than the five cans used in the study, but could wear shorts rather than long pants.

This study met most of the requirements contained in the Series 875 Group A, Applicator Exposure Monitoring Test Guidelines, and the data are useful for risk assessment.

(4) Insecticidal Dust Product Application (Bulbous Duster or Shaker Can)

HED has no data monitoring exposures from chlorpyrifos application using a duster. Therefore, chlorpyrifos exposures were evaluated using a study in the scientific literature in which a dust formulation was applied to a home garden (Kurtz and Bode 1985). This analysis is presented in a memo from D. Jaquith to Chlorpyrifos file, June 11, 1996 entitled Documentation of Applicator Exposure Assessment for Chlorpyrifos Reregistration Eligibility Document--Application in the Residential Environment. Although chlorpyrifos dust products are not registered for garden use, this study is considered to represent the best surrogate data available because it measures exposure per quantity of product handled. For this assessment, both a residential applicator and utility workers (i.e., during application of product to underground wires or cables) were evaluated. It was assumed that a homeowner could dispense a 10 oz can of a 1% ai product (2.83 g ai) (EPA 62719-54) to treat a heavily infested home, while it was assumed a worker could handle a more concentrated product (Rainbow Ko Fire Ant Killer, 7% ai, EPA Reg 13283-17), which is sold in both 4 oz and 100 oz containers (7.9 and 198.4 g ai, respectively). The label notes that the 4 oz container treats 1 sq ft^2, while the 100 oz container treats up to 100 ft^2. It was assumed that a residential applicator would be exposed short-term (i.e., 1-7 days), and that a worker could be exposed both short- and intermediate-term (i.e., 7 days to several months).
In the study, 24, 15-minute replicates were available for individuals that dispensed 190 to 220 g of a 5 percent carbaryl dust product (9.5-11 g ai or 0.021-0.024 lb ai) using a shaker can to corn and beans. Measurements were taken of the total deposition of the material on the skin/clothing surfaces. The product was applied for 15 minutes, enough time to treat an average home garden or a heavily infested home. The total potential dermal exposure, measured using total deposition was 11 mg per 15 minute treatment (5.0×10^3 mg/lb ai). Respiratory exposure was not measured.

There are no data adequate to determine the amount of protection that clothing offers to dust formulations. Therefore, HED assumed that areas covered by clothing offer 50 percent

protection and that gloves offer 90 percent protection. HED estimated exposure for workers based on total deposition, wearing long pants, long sleeves, and gloves to be 4.5 mg per 15 minutes (or 4.5 mg/10 g ai carbaryl) and total deposition for residents wearing long pants, short sleeves with no gloves to be 4.9 mg per 15 minutes (or 4.9 mg/10 g ai carbaryl). These data were normalized to g ai chlorpyrifos handled to assess an in home dust treatment. Therefore, residential chlorpyrifos exposure was estimated to be 1.4 mg ai (i.e., 4.9 mg/10 g ai carbaryl * 2.83 g ai chlorpyrifos), while worker exposure was estimated to range from 3.6 to 89 mg ai chlorpyrifos for a 4 oz and 100 oz container, respectively (i.e., 4.5 mg/10 g ai carbaryl * 7.91 or 198.4 g ai chlorpyrifos). As shown on Table 2, the resulting short-term dermal dose for residents is 20 μg/kg/day, while the short- and intermediate- term dermal doses to workers range from 51 to 1275 μg/kg/day. These exposure estimates are considered to be conservative because the quantity of chlorpyrifos dust used indoors by residents is likely to be much less than the quantity of dust products typically used in gardens.

(5) Granular Formulation Application by Hand

HED has no data monitoring exposures from chlorpyrifos application of granular formulation by hand (EPA Reg. 62715-14, 62715-210). Therefore, exposures were evaluated based on data obtained from PHED V1.1. for LCOs, and the Residential SOPs for residential applicators (also from PHED V1.1). The unit exposure estimates for LCOs assume workers wear chemical-resistant gloves plus long-sleeve shirt and long pants. There is medium confidence in the dermal and inhalation unit exposure estimates, which are based on 16 dermal, 15 hand, and 16 inhalation replicates of ABC grade data. It should be noted that the PHED unit exposure estimates are based on a single study in which a test subject wearing chemical-resistant gloves spread the granular formulation around the outside of the residence and over 90 percent of the samples contained no detectable material. The dose estimates for residential use assume that individuals wear short pants, short sleeves and no gloves. There is also medium confidence in the unit exposure estimates for residential exposure, which are based on 16 dermal, hand and inhalation replicates each of ABC grade data. It was assumed that an average application dispensed is 0.0459 lbs of active ingredient, which assumes a LCO or homeowner treats 1000 ft^2 of turf with an active granular formulation at 2 lb ai/acre. It was assumed that a LCO could apply a granular formulation for durations greater than 7 days and up to several months (i.e., intermediate term), while a resident is more likely to apply a granular formulation once or twice a season (i.e., short-term).

(6) Loading Granular Formulation and Applying with Belly-Grinder Equipment

HED has no data monitoring exposures from chlorpyrifos application of granular formulation using a belly-grinder. Therefore, exposures were evaluated based on data obtained from PHED V1.1. for LCOs, and the Residential SOPs for residential applicators (also from PHED V1.1). The unit exposure estimates for LCOs assume workers wear chemical-resistant gloves plus long-sleeve shirt and long pants. There is low confidence in the dermal unit exposure estimates, which are based on 29 to 45 dermal replicates of ABC grade, and 20 hand replicates of all grades of data. There is high confidence in the inhalation unit exposure estimates which are based on 40 replicates of AB grade data. The unit exposure estimates for residential use assume that individuals wear short pants, short sleeves and no gloves. There is also medium confidence in the dermal unit exposure estimates for residential exposure, which are based on 20 to 45 dermal, and 23 hand replicates each of ABC grade data. There is high confidence in the inhalation unit

exposures, which are based on 40 replicates of AB grade data. Similar to the scenario discussed above, it was assumed that an average application dispensed is 0.97 lbs of active ingredient based on a DAS-submitted study of a granular formulated product (MRID 44167101). In addition, this was the average amount of active ingredient handled in the 55 replicates for application of granular bait in the studies cited in PHED. It was assumed that a LCO could apply a granular formulation for durations greater than 7 days up to several months (i.e., intermediate term), while a resident is more likely to apply a granular formulation once or twice a season (i.e., short-term).

(7) Loading Granular Formulation and Applying with a Push-Type Spreader

HED has no data monitoring exposures from chlorpyrifos application of granular formulation using a push-type spreader. Therefore, exposures were evaluated based on data obtained from PHED V1.1. for LCOs, and the Residential SOPs for residential applicators (also from PHED V1.1). The unit exposure estimates for LCOs assume workers wear chemical-resistant gloves plus long-sleeve shirt and long pants, while residents are assumed to wear short pants, short sleeves and no gloves. There is low confidence in the dermal unit exposure estimates for LCOs and residential applicators due to inadequate replicate numbers, which are based on 0 to 15 dermal replicates of C grade data, 0 hand replicates for LCOs and 15 hand replicates each of C grade data for residents. There are no head, neck or hand replicates for the LCO clothing scenario. For residents, a 50 percent protection factor was used to back calculate a short-sleeved scenario from the long sleeved data. There is high confidence in the inhalation unit exposure estimates for both LCOs and residents, which are based on 15 replicates of B grade data. Similar to scenario discussed above, it was assumed that an average application dispensed is 0.97 lbs of active ingredient based on a DAS-submitted study of a granular formulated product (MRID 44167101). In addition, this was the average amount of active ingredient handled in the 55 replicates for application of granular formulation in the studies cited in PHED. It was assumed that a LCO could apply a granular formulation for durations greater than 7 days up to several months (i.e., intermediate term), while a resident is more likely to apply a granular formulation once or twice a season (i.e., short-term).

(8) Pre-Construction Termiticide Use for Subterranean Termite Control (Mixing/Loading and Applying) (MRID No. 44589001)

Exposure estimates were derived from a chemical-specific study submitted by Dow AgroSciences in which workers were monitored during application of chlorpyrifos, as the termiticide Dursban® TC (43.2% ai) (EPA Reg. 62719-47), during pre-construction termiticide treatments. HED evaluated this study in DP Barcode D247635 (Memorandum from J. Cruz to M. Hartman, May 24, 1999). This study quantified exposures to a mixer/loader/applicator (M/L/A) during mixing/loading/application and tarp pulling processes.

The M/L/A performed an open-pour mixing/loading task in which a PCO loaded Dursban® TC concentrate into a mixing tank containing the appropriate amount of water. After mixing, the diluted product was sprayed onto the soil using a hand-held sprayer and then two workers (tarp pullers) laid the untreated plastic tarp over the treated soil prior to pouring the concrete foundation.

The product was diluted to a nominal rate of 1% (actual 1.44%) prior to application. All

applications were made with a low pressure spray equipment fitted with a hand-held hose-end sprayer or spray wand fitted with a shrouded rose nozzle. The flow rates at which the spray was applied to the sites varied depending on the truck, but in general applications were between 8 to 12 gallons/minute. There were 17 M/L/A replicates, representing at least three hours exposure time. There were16 tarp puller replicates each representing 6-7 minutes. Each worker completed 8 tarp pulling replicates in less than one hour. M/L/A wore long underwear, a long sleeved shirt, long pants, and PPE consisting of rubber boots, tyvek or cotton coveralls, and arm-length gloves (note the label only requires a single layer of clothes; the coveralls and arm-length gloves are not required). Each worker removed their PPE after the spray operation was concluded. The tarp pullers wore a long sleeved shirt, long pants socks, leather and/or rubber boots, and a hat. In addition, one half (8) of the workers wore arm-length chemical resistant gloves, while the other half (8) did not wear gloves.

Dermal exposure was quantified using whole body dosimeters, and hand washes. For M/L/A, each participant wore a whole body dosimeter consisting of a long sleeved shirt and pants which were segmented and analyzed to determine potential exposures for the arms, upper legs, lower legs and torso. In addition, an undergarment consisting of one-piece cotton long underwear was collected to determine the penetration of chlorpryifos through outer clothing onto skin. Note that M/L/A replicates also wore a Tyvek (9 replicates) or cotton (8 replicates) coverall on top of the whole body dosimeter as personal protective clothing. A hat with a denim patch was analyzed to quantify head, neck, and face surface deposition.

Air samples were collected using a personal air sampling pump connected to a 37-mm GN-4 filter in series with a Chromosorb 102 tube. The filters were used to collect particulates while sorbent tubes were used to trap vapors. Samples were analyzed using GC-ECD.

As shown on Table 2, the average dermal absorbed dose (assuming a 3% dermal absorption rate) for the M/L/A wearing a single layer of clothes is 1.57 μg/kg/day, while the average inhalation dose is 0.45 μg/kg/day, based on passive dosimetry. The average dermal absorbed dose for the M/L/A wearing a double layer of clothes is 0.477 μg/kg/day, while the average inhalation dose is 0.45 μg/kg/day, based on passive dosimetry. These exposure estimates are for a 3 hour exposure measured in the study.

As shown on Table 2, the average dermal absorbed dose for the tarp pullers contacting one tarp without gloves is 0.081 μg/kg/day, while the average inhalation dose is 0.015 μg/kg/day, based on the passive dosimetry measurements. In addition, it was assumed that a worker could pull 8 tarps in one work day, which the study evaluated for construction of townhouses, or other homes under construction in close proximity. Therefore, the average 7 minute exposure for each tarp was multiplied by a factor of 8. The average dermal absorbed dose for the tarp pullers contacting eight tarps without gloves is 0.644 μg/kg/day, while the average inhalation dose is 0.122 μg/kg/day. The average dermal absorbed dose for the tarp puller wearing arm-length chemical-resistant gloves and contacting one tarp is 0.023 μg/kg/day, while the average inhalation dose is 0.021 μg/kg/day based on passive dosimetry. The average dermal absorbed dose for the tarp puller wearing arm-length chemical-resistant gloves and laying eight tarps is 0.177 μg/kg/day, while the average inhalation dose is 0.168 μg/kg/day based on passive dosimetry. It was assumed that these workers could be exposed for more than several months a year (i.e., long term).

(9) Post Construction Termiticide Use (Mixing/Loading and Applying) for Subterranean

Termite Control (MRID No. 44729402)

Exposure estimates were derived from a chemical-specific study submitted by Dow AgroSciences in which workers were monitored during application of chlorpyrifos, as the termiticide Dursban® TC (43.9% ai) (EPA Reg. 62719-47), during post-construction termiticide treatments. HED evaluated this study in DP Barcode D252357 (Memorandum from G. Bangs to M. Hartman and D. Smegal, April 29, 1999). This study quantified potential pesticide applicator inhalation, dermal, and biological exposure to chlorpyrifos. Post-construction treatments were applied to various construction styles of residential housing (i.e., slab-on-grade, basement, crawlspace and combinations thereof) in Virginia, Alabama, and Georgia. The applicators applied termiticide at a rate of approximately 4 gallons of ~1 percent a.i. dilution (range 0.71-1.24%) per 10 linear feet to an average of 124 gallons per structure (range 40-325 gallons). Mixer/loader/applicator exposures during actual structural work using hand held spray gun or injection rod were monitored by passive dosimetry and limited biomonitoring of volunteer PCO. During applications, the PCOs wore the label-required protection, including a cotton coverall, chemically resistant nitrile gloves, a hat, protective eyewear and a half-facepiece respirator (if working in confined spaces). During mixing/loading, subjects wore additional PPE that consisted of chemically resistant footwear and an extra (second) coverall or a chemically resistant apron. There were a total of 15 replicates representing 9 different volunteers, from 3 companies in three cities. The study was conducted in compliance with most, but not all, OPPTS guidelines. The biomonitoring was very limited (5 replicates) and mixing/loading exposures were not measured separately from application exposures.

Higher inhalation exposures were encountered in basement and crawlspace applications than during slab treatments. The arithmetic mean inhalation dose is 1.48 μg/kg/day (normalized 70 kg body weight), and ranged from 0.17 to 3.18 μg/kg/day normalized body weight (N=14). The geometric mean dose is 0.91 μg/kg/day. The arithmetic mean value is based on data from 14 replicates because the fifteenth replicate had an unusually high dermal dose (50 μg/kg) resulting from an accident with a broken hose. Average inhalation exposure/hour (average 6.62 hours worked) was 15 μg/hr, with a range of 1.67 to 25.84 μg/hr.

During crawlspace treatments, workers experienced the greatest amount of dermal exposure to the head/neck (~48 percent of the dermal exposure on average). During slab and basement treatments, workers experienced the highest levels of dermal exposure to the legs (~63 percent and ~51 percent respectively on average). During basement treatments, exposure to the hands was greatest (~23 percent of total dermal exposure on average), however the number of application replicates was low (N=3). The arithmetic average dermal absorbed dose (N=14) based on passive dosimetry was 3.28 μg/kg/day with a range of 0.45 to 13.85 μg/kg/day, and excluding the 49.9 μg/kg/day dose due to one replicate being sprayed by a broken hose. The geometric mean absorbed dermal dose is 2.48 μg/kg/day, including the individual sprayed with a broken hose. These values utilize the current HED dermal absorption factor of three percent.

The total mean dose, calculated by addition of average inhalation and absorbed dermal doses, was estimated to be 4.76 μg/kg/day (normalized 70 kg body weight; N=14; range: 0.82 to 16.7 μg/kg/day), with inhalation representing 31 percent and dermal representing 69 percent of total dose measured via passive dosimetry. Total estimated dose (dermal and inhalation) for the 15th replicate was 50.50 μg/kg/day, which may be considered a typical worst-case exposure because it represents an equipment malfunction (i.e., broken hose).

Total mean absorbed chlorpyrifos dose of 4.27 μg/kg/day measured via the biological monitoring of the five workers in Georgia is slightly higher than the total absorbed chlorpyrifos dose calculated as the sum of 3 percent of total potential dermal dose (corrected for dermal absorption; measured via passive dosimetry) and potential inhalation dose for the same 5 replicates (3.24 μg/kg/day). Total absorbed dose was estimated directly by biomonitoring of the chlorpyrifos metabolite 3,5,6-TCP in the urine samples of five volunteer applicators at the Georgia location (it is unclear why the fifth replicate had the same weight as another, unless one volunteer was monitored for 2 days). The volunteers were told to avoid chlorpyrifos exposure for ten days before the exposure application and for five days after the exposure. Each applicator collected all the urine voided on the day before application, the day of application, and for four consecutive days after initial exposure. The urine was collected at 12-hour intervals. The first day's collection was used as the baseline for correcting exposure calculations. The baseline chlorpyrifos ranged from 0.39 to 3.4 μg/kg(actual body weight)/day, with a mean of 1.1 μg/kg/day. The difference in estimated absorbed dose levels between biomonitoring and passive dosimetry may be due to various factors, including: incidental oral exposure to chlorpyrifos; field spike recovery from coveralls was consistently low (mean = 22 % ± 13%), so losses may not have been fully accounted for, or; subjects participating in biological monitoring experienced exposure to chlorpyrifos outside the study setting. (Note: the low field recovery data were factored into the dose estimates).

In at least three cases (replicates AL03, GA13, GA14), significantly more ai was reportedly applied than was handled, and the study report does not explain how that is possible (i.e., did the applicators use other, previously prepared solution in addition to their own?). In order to analyze the unit dose per pound ai handled, the average of the pounds "handled" and "applied" was utilized. A range of unit dose based on passive dosimetry was obtained by applying the mean exposure of the 14 replicates to the high (32.7 lb), low (4.0 lb), and mean (10.72 lb) amount of material handled.

(10) Paintbrush Application

HED has no data monitoring exposures to chlorpyrifos resulting from a paintbrush application to treat insect-infested wood. Therefore, exposures were evaluated based on data obtained from the Residential SOPs for residential applicators (also from PHED V1.1). These data represent a worker painting a bathroom with a fungicide-treated latex paint. PCOs were not evaluated for this scenario because they are assumed to treat larger surfaces of wood with rollers or a spray, rather than a paintbrush. The unit exposure estimates for residential use assume that individuals wear short pants, short sleeves and no gloves. There is low to medium confidence in the dermal unit exposure estimates for residential exposure, which are based on 14 to 15 dermal replicates of grade C data, and 15 hand replicates of B grade data. There is medium confidence in the residential inhalation unit exposure estimates, which are based on 15 inhalation replicates of C grade data. HED conducted two evaluations, a worst case scenario that assumed an individual could apply one gallon of diluted chlorpyrifos product (as Dursban 1-12 Insecticide; EPA Reg No. 62719-56) to treat a large wood-infested area, and a more typical scenario which assumed the application of a quart of diluted product for a localized wood infestation. The label recommends diluting 5.33 oz of Dursban 1-12 Insecticide (12.6% ai; 1 lb ai/gallon) with 1 gallon of water. The resulting short-term dermal and inhalation dose estimates for the worst case scenario are 140 and 0.17 μg/kg/day, respectively, while the typical scenario doses estimates are 34 and 0.043 μg/kg/day, respectively. The dose estimates are presented on Table 2.

(11) Ornamental Application

HED has no data monitoring chlorpyrifos exposures to residents during mixing/loading or application to ornamentals (flowers, shrubs, evergreens, vines, shade and flowering trees and other ornamental plants). Therefore, exposures were evaluated based on data obtained from the Residential SOPs (also from PHED V1.1) for mixing/loading and application activities. This assessment evaluates application via both a low pressure handwand and a hose end sprayer, which are assumed to be short-term scenarios for residents. A range of exposure estimates were evaluated for both application methods, the minimum, typical and maximum dilution rates of 1 oz, 4 oz and 1 quart of product per 3 gallons of water. The maximum rate is recommended for beetles. It was assumed that a resident would apply 5 gallons of diluted Dursban 1-12 Insecticide (EPA Reg No. 62719-56; 12.6% ai; 1 lb ai/gallon), in accordance with the residential SOPs for treatment of ornamental trees. The unit exposure estimates for residential use assume that individuals wear short pants, short sleeves and no gloves. For the hose-end sprayer, there is low confidence in dermal and inhalation unit exposure estimates, which are based on 8 dermal and inhalation replicates of C grade data, and 8 grade E hand replicates. For the low pressure handwand (liquid/open pour), there is low confidence in dermal unit exposure estimates, which are based on 9-80 dermal replicates of ABC grade data, and 70 hand replicates of all grades. There is medium confidence in the inhalation unit exposure estimates, which are based on 80 inhalation replicates of ABC grade data. As shown on Table 2, the dermal dose estimates range from 5.6 to 594 μg/kg/day, while the inhalation dose estimates range from 0.0018 to 0.18 μg/kg/day. The use of the low pressure handwand results in higher exposures.

Table 2 presents the exposure scenarios and exposure calculations using the above data sources for the residential uses of chlorpyrifos. Children are not included in this table since children would not be expected to apply this material, although they might be exposed after application.

3.2 Residential Postapplication Exposures & Assumptions

EPA has determined that there is potential exposure to the general public (adults and children) following applications at residential and public sites - indoors and outdoors. Postapplication exposure data were required for chlorpyrifos in a reregistration DCI issued September 19, 1991 during the reregistration process, since, at that time, one or more toxicological criteria had been triggered for chlorpyrifos. The dose estimates are presented in Tables 3 and 4.

The following studies were submitted by the registrant:

- MRID No. 40094001 Airborne Chlorpyrifos Concentrations Measured During and Following Applications of Dursban TC Insecticide to Residential Dwellings. GH-P 1310.

- MRID No. 430135-01 Vaccaro et al. 1993. Chlorpyrifos: Exposure to Adults and Children Upon Reentry to Domestic Lawns, Following Treatment with a Chlorpyrifos-Based Mixture. Study ID No. DECO-HEH2.1-1-182(121).

- MRID No. 441671-01 Vaccaro et al. 1996. Chlorpyrifos: Exposure to Adults and Children Upon Reentry to Domestic Lawns, Following Treatment with a Chlorpyrifos-Based Granular Insecticide.

- MRID No. 444582-01 Byrne et al. 1998. Residential Exposure to Chlorpyrifos from Reentry to Structures Treated with Crack and Crevice and Spot Applications of Dursban Pro.

HED reviewed each of these studies and used the registrant-submitted data to estimate exposures to adults and children in residential settings. A brief summary of each study is provided below, with reference to HED's memorandum that provides a more detailed review and analysis of the study. As noted previously, a number of the registrant-submitted studies conducted biomonitoring by measuring urinary concentrations of the primary chlorpyrifos metabolite 3,5,6-trichloro-2-pyridinol (3,5,6-TCP), to estimate chlorpyrifos exposures. Prior to the studies, baseline urinary 3,5,6-TCP concentrations were determined in the study volunteers, and these baseline measurements were subtracted from the exposure-related 3,5,6-TCP concentrations measured in the biomonitoring study. It is important to note that most individuals in the U.S., and nearly all the subjects in the Dow AgroSciences biomonitoring studies had low levels of urinary 3,5,6-TCP prior to study commencement, indicating a baseline exposure to chlorpyrifos, which most likely is attributed dietary sources.

3.2.1 INDOOR POSTAPPLICATION EXPOSURES.

(1) Crack, Crevice and Spot Treatment of Kitchen and Bathroom (MRID 44458201)

Dow AgroSciences submitted a study designed to estimate chlorpyrifos exposure to adults conducting normal daily activities following treatment of the kitchen and bathroom of three houses with crack and crevice and spot applications of Dursban Pro insecticide (0.5% chlorpyrifos dilution with water) for cockroach control. HED evaluated this study in DP Barcode D242444 (Memorandum from D. Smegal to M. Hartman, December 3, 1998). Between 0.663 and 0.787 L of product (3.32 g to 3.94 g chlorpyrifos) was applied to the houses. Six adults (four women and two men), two from each of the three treated houses, were monitored 1 day pre-application and for 10 days postapplication via urine collection and analysis. The urine was analyzed for 3,5,6-TCP, the primary metabolite of chlorpyrifos. The volunteers were instructed to perform normal activities and to spend at least 12 hours per day inside the treated house. Air monitoring was conducted at two heights in the kitchen (site of application) and family room (adjacent room). In addition, deposition measurements and dislodgeable residues were collected in the family room and a bedroom of each treated house. Dislodgeable residues were measured on hard plastic toys (balls), and also on carpets in the family room and bedroom, to determine the amount of chlorpyrifos available for absorption.

Dislodgeable residues from the carpet and hard toy wipes in non-treated rooms were generally non detectable, indicating that the potential for dermal absorption is low. Based on the biomonitoring and environmental data collected in this study, the maximum one-day chlorpyrifos dose for the 6 adult volunteers, corrected for baseline exposure, is 0.39 μg/kg/day which is comparable to or less than estimated chlorpyrifos baseline doses of 0.1 - 0.86 μg/kg/day. The overall mean dose to the six volunteers is 0.18 μg/kg/day based on the biomonitoring data, while the mean baseline dose is 0.4 μg/kg/day. The method used to estimate exposures directly measures internal dose and does not differentiate between routes of exposure. However, the study results indicate that the predominant route of exposure is through inhalation.

Exposures to young children were estimated using air concentrations measured 15 inches above the floor, and conservative EPA default exposure assumptions (i.e., breathing rate, body weight and duration of exposure). Dermal and oral exposures were assumed to be negligible based on an absence of detectable dislodgeable residues in the carpet wipes or on hard plastic toy wipes in all three houses, except for a negligible quantity of residue detected on a hard ball in the family room of house #3. For example, if a child ingested the entire residue present on the toy, the resulting dose would be approximately 0.089 μg or 0.006 μg/kg, which is negligible relative to the estimated exposures from inhalation (10 -100 fold less). The estimated 10 day mean doses to children are 0.08, 0.28 and 0.22 μg/kg/day, while the highest one-day doses are 0.27, 0.76 and 0.61 μg/kg/day for houses #1, #2 and #3, respectively. These exposure estimates are also within the background range observed for adults. The one day exposure estimates are conservative, because they assume a child could spend 21 hours exclusively in the room with the highest detected concentration. However, this study did not evaluate chlorpyrifos residues on soft plush toys, which could also contribute to child exposure.

In conclusion, these data demonstrate that exposures to adults and children following crack, crevice and spot applications of chlorpyrifos in the kitchen and bathroom by a licensed applicator are comparable to typical background exposures levels. However, these data do not support the use of crack and crevice or spot treatment in bedrooms, living rooms, closets, day care centers, schools, playhouses, on furniture or draperies, or in other rooms that could result in higher exposure to individuals, particularly children. In addition, these data do not support the indoor application of up to 1% Dursban Pro for the treatment of exposed wood surfaces, voids and channels in damaged wood, wall voids, and junctions between wood and foundation that are currently listed on the label.

In addition, low air concentrations of chlorpyrifos were still present in all three homes 10 days post treatment, however some of the current labels allow re-treatment every 7 days. In one house, the highest daily average air concentrations were detected on the 6th day following chlorpyrifos treatment, indicating possible sinks and resuspension. The results of this assessment are presented in Table 3. This study has not addressed the possible cumulative effects of multiple treatments over time, although, additional information has been requested from the registrant to support a 7 day re-treatment interval as proposed in the Dow AgroSciences submission (MRID 44331901).

(2) Crack and Crevice Treatment of Other Rooms Using Residential SOPs

HED also assessed potential short-term exposures to adults and children using the Draft Residential SOPs (December 18, 1997), to supplement the evaluation of crack and crevice treatment based on the registrant-submitted biomonitoring study discussed above. This additional assessment was conducted due to the concerns that the registrant-submitted biomonitoring did not adequately evaluate exposures that could occur following treatment of baseboards and window and door frames in family rooms, bedrooms, living rooms or other treatments that could occur in schools, day care centers, playhouses, or the many other buildings listed on the labels.

The highest deposition residue detected in the family room of house #3 (room adjacent to treated kitchen) from the registrant-submitted biomonitoring study was used in this analysis (i.e., 2.298 μg/100 cm^2 collected one day postapplication). This assumption was considered reasonable,

although it would have been preferable to have actual residue data from the treated kitchen (these data were not provided). Exposures were estimated for both adults and children, assuming that 50% of the residue is available as dislodgeable residue. The standard default assumptions recommended in the Residential SOPs were used, which include: body weights of 70 and 15 kg for adults and children, respectively, transfer coefficients of 48,000 and 8,700 cm^2 for adults and children, respectively, exposure time of 8 hours for contact with carpet and 4 hours for contact with surfaces, child hand surface area of 350 cm^2, and a frequency of entire hand to mouth activity of 1.56 times/hour. Inhalation exposures were not calculated using the SOPs, because comprehensive air monitoring was conducted in the registrant-submitted biomonitoring study, and HED believes inhalation exposures were adequately characterized. The estimated doses for dermal and oral exposures are presented on Table 3. As shown on the table, the estimated doses are significantly higher than those estimated from the biomonitoring study, suggesting that dermal and oral exposures are of concern in rooms treated with chlorpyrifos.

Scientific Literature on Indoor Broadcast Application

In 1998, scientists at Rutgers University published a study that evaluated exposure to children following a single broadcast use of chlorpyrifos in two apartments by a licensed pesticide applicator (Gurunathan et al. 1998). The Gurunathan et al. (1998) study evaluates a broadcast application, a method which the registrant voluntarily canceled in 1997, that raises some exposure issues not fully addressed by a crack and crevice application study discussed above (MRID No. 44458201). For example, the broadcast study detected chlorpyrifos residues in plush toys placed in treated rooms one hour after application, whereas, the crack and crevice study only measured dislodgeable residues from carpets and hard plastic toys 1 hour to 10 days post-treatment that were placed in untreated rooms (i.e., bedroom and family room) prior to treatment. In addition, the broadcast study accounted for the frequent hand-to-mouth activity of children based on videotaping, which the crack and crevice study could not adequately address because it estimated adult exposures (whose activity patterns are different) based on biomonitoring data. Gurunathan et al. (1998) measured chlorpyrifos in air, plastic and plush toys, and in dust in and on smooth surfaces. This study estimated child doses of 208 μg/kg/day (or 634 μg/kg/day for high hand to mouth contact) based on environmental measurements and conservative exposure assumptions. However, these exposure estimates were not validated by actual measurements of absorbed doses based on urinary excretion of 3,5,6-TCP (as was done for the crack and crevice study discussed above). The study concluded that dermal and oral exposures via toys and other surfaces may present greater risk than inhalation, and that potential inhalation exposure was negligible. In addition, this study observed continued deposition on surfaces in treated rooms 2 weeks postapplication, and demonstrated that chlorpyrifos may adhere to objects brought into a room hours or days after pesticide application. Peak deposition on surfaces (of plastic toys) occurred 36 hours postapplication (0.043 μg/cm^2). The authors suggest that the current labels specifying a re-entry time for residents of 1-3 hours based on air measurements may be inadequate, and that routine application could lead to the accumulation in toys or other sorbant surfaces (i.e., pillows). The authors recommend that toys should not be stored in open rooms at least one week after broadcast application of chlorpyrifos.

HED evaluated this study, and concluded that it significantly overestimates the typical child doses resulting from currently registered indoor uses. In addition, the estimates in this study are significantly higher than those estimated based on a broadcast application biomonitoring study

submitted by the registrant (MRID No. 42008401), and reviewed by HED (memo from D. Jaquith to D. Edwards, DP Barcode: D168824, August 18, 1995). For example, HED estimated child doses of 23 μg/kg/day on day one and 14 μg/kg/day on day two following a broadcast application. The following is a list of refinements that need to be considered, or uncertainties that exist in the Gurunathan et al. (1998) study:

- A total of 12 g of chlorpyrifos was applied directly to entire floor surfaces of each room, which is approximately three times more than the amount applied for crack and crevice treatment (3.32-3.94 g based on the study above).
- The toys (plush and plastic) were placed directly on treated surfaces 1 hour postapplication, which enhances the quantity of chlorpyrifos sorbed to the toys, relative to the amounts found from air deposition in the crack and crevice study. Current registered uses (i.e., crack and crevice) are not likely to result in toys contacting treated areas.
- A hexane-methanol solvent was applied to the dresser surfaces and was used in the wipe samples, while hexane was used to extract dust and toy residues. The solvent enhances chemical availability from the surfaces resulting in higher residue measurements than are likely to be absorbed by an individual contacting or handling these surfaces/toys.
- The bioavailability of chlorpyrifos in the toys (i.e., amount available for absorption) was not addressed, as noted by the study authors.
- The exposure estimates assumed that children touch a contaminated surface 366 times/hour and put their contaminated hand in their mouth 70 times/hour. However, it is unlikely that chlorpyrifos concentrations are replenished on the entire hand surface every time a child touches a surface.
- The hand surface area and inhalation rate used to estimate child exposures are higher than EPA's recommended values in the Draft Residential Exposure SOPs or the Exposure Factors Handbook (i.e., study used 400 cm^2 for hand surface area and 12 m^3/day for inhalation rate compared to the mean EPA-recommended values of 350 cm^2 and 8.3 m^3/day, respectively).

The Agency concludes that the screening-level estimate derived in this study can be better refined using values from the EPA's Exposure Factors Handbook, conducting biomonitoring to determine absorbed dose, and using more realistic sampling methodologies.

(3) Pet Collar Uses

A number of pet collars are currently registered. HED has no chemical-specific data that evaluate exposures to individuals from the use of pet flea collar products. Therefore, HED conducted this analysis in accordance with HED's 1997 Draft SOPs for Residential Exposure Assessments. However, a pet collar exposure study is underway at Mississippi State University by Dr. Janice Chambers. HED evaluated pet collars that contained 3-9% ai chlorpyrifos, considered to be representative of these products, in DP Barcode D2532246 (Memorandum from D. Smegal to J. Rowland, March 1, 1999). These collars are sulfodene scratchhex flea and tick collar for cats 4306-16 and Zema 11 month collar for dogs 45087-40. Exposures were estimated assuming that one percent (0.01) of the active ingredient applied to the pet to be available for dermal and inhalation exposure from handling flea collars. This assumption is based on the best professional judgement of the OPP/HED staff and is assumed to be an upper-percentile value. For this analysis, a range of exposure estimates were calculated. One estimate assumed that exposure was equally divided between the inhalation and dermal routes (i.e., 50% dermal and

50% inhalation), while the other assumed that exposure was exclusively through dermal contact. In addition, EPA-recommended default mean body weights of 70 kg for adults and 15 kg for children age 1-6 years of age were used to estimate dose.

Additional refinements were incorporated into this analysis to account for the duration of exposure (i.e., labeled efficiency of the product is 11 months or 330 days), and to account for the amount of chlorpyrifos that could be dermally absorbed through the skin of humans. A dermal absorption factor was used because the long-term dermal no-observed-adverse effect level (NOAEL) used to calculate MOEs is based on an oral two-year dog study and route-to-route extrapolation. This refinement assumes steady-state exposure to chlorpyrifos. Dermal absorption was estimated to be 3 percent based on the ratio of the oral lowest-observed-adverse effect level (LOAEL) of 0.3 mg/kg/day from the rat developmental neurotoxicity study (MRID Nos. 44556901, 44661001) to the dermal LOAEL of 10 mg/kg/day from the 21-day dermal study (MRID No. 40972801) for plasma and red blood cell cholinesterase inhibition. This absorption factor is comparable to the dermal absorption estimated from human data of 1-3% (MRID No. 00249203). The dose estimates and MOEs for two pet collar products for each age class are presented in Table 3.

(4) Residential Treatment for Subterranean Termite Control (MRID No. 40094001)

A study submitted by the registrant (MRID No. 40094001) was used to determine the respiratory exposures of the residents of homes treated with chlorpyrifos (0.5-1% Dursban TC) for subterranean termite control. Thirty two homes, 8 each of plenum, crawlspace, slab, and basement construction, were treated at several different locations throughout the country. Applications were made by licensed professional applicators using conventional equipment and following the label instructions. Air in the kitchen, one bedroom, and the basements of basement construction homes was monitored before treatment and at various intervals after application for one year.

Treatment of homes with chlorpyrifos for subterranean termite control appears to result in a slightly increased exposure over background levels soon after treatment. Exposures return to background levels within a few days after the application for slab, crawlspace, and the first floor rooms of basement homes. Basements showed higher concentrations of the chemical than first floor rooms. The concentrations in basements declined slowly over time, reaching first floor levels within one year after application. Treatment of plenum structures appears to result in airborne concentrations in first floor rooms that are slightly higher than those observed in other construction types. These increased levels return to background within a few months after application.

Adults and children were assumed to be in the residence for 16.4 and 21 hours per day, respectively based on EPA default assumptions. The resulting respiratory doses are presented in Table 4. As shown on Table 4, the maximum 1 year average air concentrations ranged from 0.11 to 0.29 $\mu g/m^3$ in the study submitted by the registrant. These concentrations represent the average of the highest detected concentration from 8 homes. However, studies in the published literature measured slightly higher air concentrations (average of kitchen and bedroom) of 1.32-3.13 $\mu g/m^3$ at 1 year postapplication, and similar concentrations of 0.1 to 0.3 $\mu g/m^3$ up to 8 years postapplication in homes of similar construction (slab and crawl construction) (Wright et al. 1988, 1994).

It should be noted that all of these studies only evaluate exposures resulting from treatment of soil outside the home, and do not evaluate the potentially higher exposures that could result from indoor treatment of a termite infestation.

(5) Insecticidal Dust Products

No data are available to evaluate the postapplication residential exposures and risks associated with the use of insecticidal dust products indoors. In addition, there are no recommended procedures for evaluating these products in the Residential SOPs. Nevertheless, HED has concerns about the use of these products based on the relatively low MOEs calculated for residents or workers that could apply these products. HED recommends that the registrant provide additional information on the potential postapplication residential exposures associated with these products.

3.2.2 OUTDOOR POSTAPPLICATION EXPOSURES

(6) Lawn Treatment using a Liquid Spray (MRID No. 43013501)

Residential exposures following lawn treatment with a liquid chlorpyrifos spray were quantified based on a chemical-specific biomonitoring study submitted by Dow AgroSciences (MRID No. 43013501). HED's review of this study is presented in memo D197713 from D. Jaquith to L. Propst entitled "Review of study measuring environmental levels of and exposure to chlorpyrifos following lawn care treatment" dated June 17, 1996. In this study, eight volunteers performed activities intended to mimic a child walking/running, sleeping, crawling, and sitting on the turf following a broadcast treatment with 0.29 percent liquid chlorpyrifos spray (as Dursban Turf Insecticide). The insecticide was applied at the maximum label rate of 3 ounces per 1000 ft^2. The activities were performed for a period of four hours, beginning when the turf had dried, four hours after application, however only two of the hours consisted of direct dermal contact with the lawn. Exposures were monitored by measurement of urinary 3,5,6-TCP concentrations. Dislodgeable residues were monitored over the 48 hour period following drying of the turf, and were determined by dragging a weighted patch ("DOW Sled") over the treated surface at various time intervals. It must be recognized that the “Sled” dosimeter represents new technology and that the relationship between dragging a denim patch and transfer to actual human skin has not been established. No data are available for further dissipation after 48 hours, making extended exposure analyses impossible. Due to the design of the biological monitoring study, it was not possible to derive separate exposure values for subsequent days.

The registrant attempted to address the issue of possible exposure of children through hand/oral contact following contact with a treated surface by washing the hands and assuming that all of the material rinsed from the hands was available for oral ingestion. The oral exposure, however, was adjusted for hand surface area (i.e., a child’s hand is 41% of an adults hand). There are no quantitative data addressing the possible exposure via the hand/oral route currently available. The assumption was considered to provide a reasonable estimate of exposure via this route.

As shown on Table 3, for adults, the mean total estimated dose, corrected for baseline, is 6.3 μg/kg/day with a range of 3.5 to 10.1 μg/kg/day for a single exposure event immediately after drying of the treated turf. The extrapolated mean dose estimate for a 1-6 year old child is 10 μg/kg/day with a range of 7.9 to 13 μg/kg/day. This extrapolation to child may underestimate

exposure because it neglects incidental ingestion of soil, and/or mouthing grass.

(7) Lawn Treatment using a Granular Product (MRID No. 44167101)

In addition, residential exposures following lawn treatment with chlorpyrifos were quantified for a granular insecticide (MRID No. 44167101). HED's review of this study is provided in memo D233282 from D. Smegal to M. Hartman entitled "Exposure of Individual to chlorpyrifos following Turf Treatment with a Granular Product", dated November 18, 1998. In this study, nine volunteers performed activities intended to mimic a child walking/running, sleeping, crawling and sitting on turf following application of a granular formulation of 0.5% chlorpyrifos at a rate of 1.8 lb active ingredient (ai) per acre. The activities were identical to those evaluated in the liquid lawn study discussed above. The activities occurred for a four hour period postapplication, although only two of the hours consisted of direct dermal contact with the lawn.

Absorption of chlorpyrifos was determined by monitoring the amount of metabolite 3,5,6-TCP excreted in the urine over an average of 5.5 days following exposure. Based on the biomonitoring and environmental data collected in this study, the mean total dose to 8 adults (4 male and 4 female), corrected for baseline exposure is 1.4 μg/kg/day with a range of 0.56 to 3.7 μg/kg/day. The extrapolated estimate of a child's dose (1-6 yrs old) based on the adult data is a mean of 2 μg/kg/day, with a range of 0.75 to 5.1 μg/kg/day. The method used to estimate exposures directly measures internal dose and does not differentiate between routes of exposure. This extrapolation to child may underestimate exposure because it neglects incidental ingestion of granules or soil. In addition, the exposures may be underestimated for individuals that follow the label because deposition measurements indicate that only 75% of the theoretical recommended label rate was applied to the field where exposure activity occurred. However, the amount applied is within the typical variation for the equipment used.

(8) Mosquitocide Uses

HED evaluated potential postapplication bystander exposure to chlorpyrifos from the mosquito control applications. Chemical-specific data are not available. Therefore, literature studies, the AgDrift Model (V1.0) that was developed by the Spray Drift Task Force, and the Residential SOPs were used to develop a screening-level assessment. The use of the literature and Ag Drift Model is consistent with the assessment that was developed in the fenthion RED. No proprietary data from the model library were used in this assessment. The purpose of these model calculations is to refine the turf deposition factor for aerial application of chlorpyrifos in mosquito control public heath treatments. Details of this analysis are presented in DP Barcode D252022, Memorandum from J. Dawson and D. Smegal to S. Knizner and M. Hartman, April 6, 1999.

HED evaluated potential postapplication exposures to adults and child residents entering treated lawns following ground-based fogger Mosquitomist One ULV (EPA Reg. 8329-24) mosquito control uses. Potential exposures were estimated because of the concern for the residues that may be deposited during the ultra low volume (ULV) ground-based fogger applications in the vicinity of residential dwellings or other recreational areas (e.g., schools, playgrounds, parks, athletic fields). Exposure from ULV aerial applications of Mosquitomist One was evaluated and determined to be negligible. This assessment has been developed to ensure that the potential exposures are not underestimated and to represent a conservative model that encompasses potential exposures received in other recreational areas (e.g., school playgrounds, parks, athletic

fields). The evaluated scenarios that could result in postapplication are as follows:

- Dermal exposure from residues deposited on turf (adult and child);
- Incidental non-dietary ingestion of residues deposited on lawns from hand-to-mouth transfer (toddler);
- Ingestion of treated turfgrass (toddler); and
- Incidental ingestion of soil from treated areas (toddler).

Chemical-specific data for mosquito uses are not available. Therefore, the equations and assumptions used for each of these four scenarios were taken from the Draft SOPs for Residential Exposure Assessments guidance document. Although the SOPs were initially developed for direct turf applications, the models are used in this assessment to determine if there is a potential concern using a screening level approach (i.e., tier 1). In addition to the use of the SOPs, the unique nature of the mosquito control uses requires additional information in determining the deposition rate of chlorpyrifos (i.e., amount of ai deposited on residential turf). The determination of the deposition rates are consistent with HED's assessment developed in the fenthion RED. HED did not calculate airborne concentrations and complete an inhalation-based risk assessment because of the infinite dilution that is anticipated in an outdoor application and based on the very low application rate. The dose estimates for adults and children, by pathway, are presented on Table 3.

(9) Yard and Ornamental Sprays

Yard Application

The potential exposures associated with chlorpyrifos-containing yard and ornamental products were evaluated based on a comparison to the exposures associated with liquid and granular insecticidal products for turf (MRID No. 43013501, for liquid insecticide, and 44167101 for granular insecticide). Details of this evaluation are presented in HED Review DP Barcode D2532246 (Memorandum from D. Smegal to J. Rowland, March 1, 1999).

A typical yard and ornamental spray product recommends that a 5.3% ai chlorpyrifos product be diluted at a rate of 4 oz/15 gallons of water, and applied to 500 ft^2 of yard (Ortho® Lawn Insect Spray, EPA Registration No. 239-2423, 1996). In the absence of product density information, the density of water (8 lb/gal) was assumed to estimate a total application rate of 0.0265 lb ai /1000 ft^2 (1.15 lb ai/acre). Therefore, this product application rate is approximately 3.5 times less than the application rate for the liquid turf product of 0.0937 lb ai/1000 ft^2 (i.e., 4.1 lb ai/acre) (MRID No.43013501), and approximately 64 percent of the application rate for the granular product of 0.0413 lb ai/1000 ft^2 (MRID No. 44167101).

Another turf and ornamental product recommends that a 24.64% ai chlorpyrifos product be applied from 1.5- 6 oz/1,000 ft^2 of yard (Dursban® 2E, EPA Registration No. 9404-66). This product contains 2 lb ai/gallon of chlorpyrifos. Therefore, the product application rate would range from 0.023 to 0.0936 lb ai/1,000 ft^2 (1.0 to 4.1 lb ai/acre), which is similar to the liquid and granular turf application rates.

By analogy, therefore, exposures resulting from the use of these yard insect sprays are expected to be similar or less than those resulting from the lawn insecticides. Average doses for adults are

expected to range from 1.4 to 6.3 μg/kg/day for a four hour exposure the day of product application, but only two hours consisted of direct dermal contact with the treated turf. Extrapolated mean doses to children are expected to range from 2 to 10 μg/kg/day. Exclusive ornamental use is expected to result in lower exposures; however, because the labels allow both yard and ornamental uses, the yard use (which results in the higher potential exposures) has been evaluated.

4.0 OCCUPATIONAL AND RESIDENTIAL RISK CHARACTERIZATION

Margins of exposure (MOEs) for occupational and residential exposure were calculated for short-term (one to seven days), intermediate-term (one week to several months), and long-term exposure (several months to lifetime), depending on the scenario. The MOE is calculated by dividing the NOAEL by the daily exposure. The NOAELs presented on Table 1 were used to calculate risks.

The acceptable margin of exposure (MOE) is 300 for oral, dermal and inhalation exposures for all residential populations, including infants and children (including residents). This factor includes 10X for interspecies extrapolation, 10X for intraspecies variation and a 3X Food Quality Protection Act (FQPA) factor. The acceptable MOE for commercial PCOs is 100 for all routes of exposure.

A total MOE is also calculated because there is a common endpoint (i.e., cholinesterase inhibition). Route-specific data are available for the dermal, inhalation and oral routes of exposure, therefore, the following reciprocal MOE calculation is used:

$$MOE_{Total} \quad \frac{1}{\frac{1}{MOE_{(Oral)}} + \frac{1}{MOE_{(Dermal)}} + \frac{1}{MOE_{(Inhalation)}}}$$

4.1 Risk and Uncertainty Characterization of Handler Exposures

MOEs for occupational and residential handler exposure were calculated for short-, intermediate and long-term exposure. Table 2 presents the exposure scenarios and exposure calculations using the above data sources for the non-agricultural occupational uses of chlorpyrifos. Children are not included in this table since children would not be expected to apply this material, although they might be exposed after application.

(1) Indoor Crack and Crevice Treatment. The long-term MOEs for PCOs were calculated based on passive dosimetry measurements obtained from a chemical-specific registrant-submitted study in which 0.29% Dursban Pro® was applied using a 2-gallon, hand pressurized B&G sprayer. As shown on Table 2, the mean dermal and total MOEs are less than 100 and exceed HEDs level of concern (range from 17 to 59, with total MOEs of 13 and 45) for PCOs that could handle more than 0.02 lb ai per day (the average quantity in the study). Inhalation MOEs are above 100 (197 to 20,000), except for PCOs that handled the maximum quantity in the study (0.0684 lb ai) (MOE is 58). However, the total MOE is 4500, and does not exceed HED's level of concern if a minimal quantity of 0.0002 lb ai chlorpyrifos is handled. Risks were calculated for the full range of exposures evaluated in the registrant-submitted study because there is insufficient information available on the distribution of actual product used by PCOs during crack, crevice and

spot treatments. It should be noted that these risk estimates are based on PCOs that wore a double layer of clothes, chemically-resistant boots and gloves and eye protection.

These risk estimates represent an average scenario because only two of the 15 worker replicates reflect the maximum recommended label concentration of 0.5%; an average of 0.29% chlorpyrifos (as Dursban Pro®) was handled by the fifteen PCOs. In addition, as noted previously, there was a large variation in exposure results due primarily to the range of chlorpyrifos ai handled (0.09 to 31.04 g), volume applied per replicate (0.02 to 2.8 gallons), sampling time (248 to 591 minutes or 4 to 9.85 hours), spray time (12 to 154 min) and percent chlorpyrifos handled (0.05 to 0.53%). In addition, it is possible that different tasks/activities associated with pesticide application in residential and commercial locations contributed to the range of exposures. However, the impact of applicator activities can not be determined due to an absence of study details.

The short-term exposures and MOEs for a resident that could apply a crack and crevice aerosol spray to their home were evaluated using PHED V1.1., in the absence of chemical-specific data. As shown on Table 2, the total MOEs are less than 300 for the application of an entire 16 oz can of 1% ai or 0.5% ai chlorpyrifos (100 and 200, respectively), and therefore exceed HEDs level of concern. The total MOEs are due primarily to dermal exposure. These risk estimates are conservative, and assume that a resident will apply an entire 16 oz aerosol can in one day. In addition, HED evaluated a spot treatment, assuming the application of 2 oz of a 0.5% ai product. The resulting total MOE is 1600 and does not exceed HED's level of concern.

(2) Broadcast Turf Applications

Lawn Care Professional

The intermediate and long-term exposures and MOEs were based on a chemical-specific registrant-submitted study that evaluated exposures to 15 lawn care applicators based on both passive dosimetry measurements and biomonitoring of urinary TCP. The geometric mean dose estimate of 0.4 μg/kg/day, used in this assessment is based on the biomonitoring results, which are considered to be more reliable that the passive dosimetry results. However, because the biomonitoring data do not differentiate between route of exposure, only a total exposure estimate and MOE could be calculated. The total MOE of 75 for the lawn care applicator exceeds HEDs level of concern (i.e., less than 100). In addition, risks were calculated for potential chlorpyrifos exposure at the maximum label-recommended application rate of 4 gallons/1000 ft^2 for subsurface soil treatment, because the study only evaluated an application rate of 2 gallons/1000 ft^2. This results in an approximate MOE of 38, which also exceeds HED's level of concern. These risks are based on workers that wore a single layer of clothes, chemically-resistant knee-high boots and gloves and a hat.

Because there is insufficient information to determine if lawn care professionals are exposed for intermediate (7 days- several months) or long-term durations, the long-term toxicity endpoints were conservatively used to calculate the MOEs based on the biomonitoring results for applicators. However, the intermediate and long-term dermal endpoints, and long-term inhalation endpoints are identical (30 μg/kg/day) because they are based on the same chronic oral dog study.

Risks were also evaluated for a mixer/loader that could handle liquids using surrogate exposure

data obtained from PHED, Version 1.1. As shown on Table 2, the total intermediate, and long-term MOEs for both the application rates (2 gallons/1000 ft^2 and 4 gallons/1000 ft^2) are above 100 (range from 190 to 820) and therefore, do not exceed HED's level of concern. The MOEs are dominated by dermal exposure. The MOEs for mixer/loader activities, which are based on route-specific PHED data, were calculated for both intermediate- and long-term exposures using the appropriate toxicity values (i.e., the intermediate and long term inhalation endpoints of 100 and 30 μg/kg/day, respectively). In conclusion, MOEs do not exceed HED's level of concern for mixer/loaders that wear the label-specified PPE.

Residential Applicator

The short-term total MOEs for residents that mix/load and apply chlorpyrifos to their lawns range from 6 to 37, and therefore exceed HED's level of concern for residents (MOEs less than 300). This assessment evaluated both broadcast and spot treatment using the hose end sprayer, and low pressure handwand, respectively, and used exposure assumptions recommended in the Residential SOPs because of the lack of chemical-specific information. The majority of the exposure results from dermal exposure, as all the inhalation MOEs exceed 300.

As noted previously, there is low confidence in dermal and inhalation unit exposure estimates for the hose-end sprayer scenario. In addition, there is low confidence in dermal unit exposure estimates, and medium confidence in the inhalation unit exposure estimates for the low pressure handwand. These MOEs are based on central tendency exposure estimates of the unit exposure, area treated, and body weight, and a central to upper-percentile assumptions for the application rate recommended in the Residential SOPs. Therefore, these MOEs are considered to be representative of central tendency to high-end estimates.

(3) Ready-to-Use Formulated Product. The short-term doses and MOEs were based on a chemical-specific registrant-submitted study that evaluated exposures to 15 homeowners based on both passive dosimetry measurements and biomonitoring of urinary TCP. The geometric mean of the lognormally-distributed dose is estimated to be 0.24 μg/kg/day. This assessment is based on the biomonitoring results, which are considered to be more reliable that the passive dosimetry results. However, because the biomonitoring data do not differentiate between route of exposure, and the short- and intermediate-term toxicity endpoints are different for dermal and inhalation exposure, the passive dosimetry results were used to segregate the total exposure estimate. As discussed previously, based on the dosimetry data approximately 88% of the total dose was from dermal exposure, while approximately 12% was from inhalation.

As shown on Table 2, the resulting absorbed dose estimates used in the risk assessment are 0.029 µg/kg/day for inhalation and 0.21 µg/kg/day for dermal. For short-term scenarios (such as residents), the absorbed dermal dose estimate was further adjusted to an estimated dermal dose (non-absorbed) of 7 µg/kg using a 3% dermal absorption factor for direct comparison with the short-term dermal toxicity endpoint. The resulting combined dermal and inhalation MOEs are above 300 for a resident (590), and therefore do not exceed HED's level of concern. These exposure estimates represent a central-tendency to high-end scenario for residents, who are more likely to apply one can of product rather than five cans in a given day, but could wear shorts, rather than long pants.

(4) Insecticidal Dust Products. Due to an absence of chemical-specific data the exposures and risk estimates resulting from use of insecticidal dust products were evaluated using a scientific study that provided exposure estimates (i.e., deposition) per quantity of dust product handled. As discussed previously, the data were normalized for chlorpyrifos exposure. As shown on Table 2, the short-term MOEs for both residents and utility workers (i.e., treating underground wires) that could apply dust products are below 100 and 300, respectively, and therefore exceed HED's level of concern (250 for residents and 0.8 to 98 for workers depending on quantity handled and duration of exposure). These estimates could overestimate exposures and risks because they are based on a study that evaluated a 15-minute application of a 5% dust formulation to the garden (Kurtz and Bode 1985). The residential MOEs are central tendency to high end and assume the application of an entire 10 oz can of a 1% ai product. The worker MOEs are central tendency for application of a 4 oz can (7% ai), and high end for the application of a 100 oz container (7% ai) of dust product. Because the study did not measure inhalation exposure, the exposure estimates and MOEs do not account for this exposure pathway, which could result in an underestimation of risk.

(5) Granular Formulation by Hand. Due to an absence of chemical-specific data, the exposures and risks resulting from hand application of granular formulation were evaluated using data from PHED V1.1 and the residential SOPs. As shown on Table 2, the intermediate-term total MOE for a LCO (20) and the short-term total MOE for a resident (17) are less than 100 and 300, respectively and therefore, exceed HED's level of concern. The risk estimates are driven by dermal exposure. As noted previously, there is medium confidence in the unit exposure estimates from PHED that are based on a single study in which a test subject wearing chemical-resistant gloves spread the granular formulation around the outside of the residence and over 90 percent of the samples contained no detectable material.

(6) Granular Formulation Application with Belly Grinder. Due to an absence of chemical-specific data, the exposures and risks resulting from the belly grinder application of a granular formulation were evaluated using data from PHED V1.1 and the residential SOPs. As shown on Table 2, the total intermediate-term MOEs for a LCO (7) and the short-term MOEs for a residential applicator (3) are less than 100 and 300, respectively and therefore exceed HEDs level of concern. The risks are dominated by dermal exposure. As noted previously, there is low and medium confidence in the dermal unit exposure estimates for LCOs and residents, respectively, and high confidence in the PHED inhalation unit exposure estimates used to evaluated LCOs and residents.

(7) Granular Formulation Application with Push-type Spreader. Due to an absence of chemical-specific data, the exposures and risks resulting from the push type-spreader application of granular formulation were evaluated using data from PHED V1.1 and the residential SOPs. As shown on Table 2, the total MOEs for both a LCO (54) (intermediate-term) and residential applicator (110) (short-term) are less than 100 and 300, respectively and therefore exceed HEDs level of concern. The risk estimates are driven by dermal exposure. The inhalation MOEs for both LCOs and residents are 1150, and therefore do not exceed HEDs level of concern. As noted previously, there is low confidence in the dermal unit exposure estimates from PHED and high confidence in the PHED inhalation unit exposure estimates.

(8) Pre-Construction Termiticide Treatment. The long-term doses and MOEs were based on a chemical-specific registrant-submitted study that evaluated exposures to

mixer/loader/applicators (M/L/A) and tarp pullers based on dermal passive dosimetry measurements and air monitoring. As shown on Table 2, the mean doses to M/L/A resulting from a 3 hour exposure resulted in MOEs that exceed HED's level of concern of 100 (range 15-33) regardless of clothing (one or two layers). (Note the label requires only one layer of clothing, and does not require forearm length gloves, as worn by the workers). The MOEs a tarp puller were also below 100 for a tarp puller that could contact 8 tarps in one day (as was done in the study), and exceeded HED's level of concern even when the worker wore forearm-length chemical resistant gloves (range of 39-87). However, the MOEs are above 100 for workers that could lay only one tarp (approximately 7 minute duration), with and without gloves (range from 310 to 690). These exposures and MOEs are considered low-end estimates for workers that wore a double layer of clothing and forearm length gloves (not required by the label) and central tendency estimates for the workers that wore single layer of clothing and forearm length gloves (only regular gloves required by the label). These data could underestimate risks to a worker that is exposed for more than 3 hours per day or applies a 2% dilution spray to treat utility poles and fences (because the study applied a 1% ai diluted product).

(9) Post-Construction Termiticide Treatment. The long-term doses and MOEs were based on a chemical-specific registrant-submitted study that evaluated exposures to 15 PCOs mixing, loading and applying a chlorpyrifos product based on both passive dosimetry measurements and biomonitoring of urinary TCP. Because the biomonitoring measurements were only available for 5 individuals, the risks were calculated using both biomonitoring and dosimetry results. As shown on Table 2, the arithmetic mean biomonitoring dose is 4.27 μg/kg/day and the resulting total MOE is 7 and therefore, exceeds HED's level of concern. The geometric mean absorbed dermal and inhalation dose estimates based on the passive dosimetry are 2.48 and 0.91 μg/kg/day, respectively. The dosimetry dose estimates also result in MOEs that exceed HEDs level of concern (range from 12 to 33, with a total MOE of 9). It should be noted that during application the workers wore the label-specified PPE which includes long pants, long sleeve shirt, chemically resistant gloves, eye protection, a hat and a half face-piece respirator in confined spaces. In addition, during mixing and loading the workers also wore a second layer of clothes or apron and chemically resistant boots. These dose estimates and MOEs are considered central-tendency values and exclude exposure to a worker whose hose broke during the study, resulting in a dose that was ten times greater than the mean dose of the other 14 workers. In addition, these risks could underestimate exposures to workers that handle more concentrated solutions of 2% allowed on the label to treat utility poles and fences because the workers in the study applied a 1% diluted product.

(10) Paint Brush Applications. Due to an absence of chemical-specific data, the exposures and risks resulting from a paintbrush application to treat insect-infested wood by a resident were evaluated using data from the residential SOPs for both a worst case (1 gallon product) and typical scenario (1 quart product). As shown on Table 2, the total short-term MOEs for both scenarios are below 100 (35 and 140, respectively) and therefore, exceed HED's level of concern. The risks are dominated by dermal exposure. The inhalation MOEs are well above 300 (590 and 2300, respectively). There is low to medium confidence in the dermal unit exposure estimates and medium confidence in the inhalation unit exposure estimates. The unit exposure estimates recommended by the residential SOPs are central tendency (i.e., unit exposure values and body weight). Therefore, the MOEs for the typical case of 1 quart are considered to be a central tendency values, while the worst-case estimates are considered to be high end values.

(11) Ornamental Application. The exposures and risks to residents during the mixing/loading and application of chlorpyrifos to ornamentals were evaluated using the residential SOPs, due to an absence of chemical-specific data. As shown on Table 2, the total short-term MOEs based on application via the low pressure handwand and hose end sprayer are below 300 (range from 8 to 270), and therefore exceed HED's level of concern. However, the total MOE is greater than 300 (880) if only the minimum rate (1 oz product/3 gallons of water) is applied to ornamentals via the hose end sprayer. These estimates are considered central tendency to high-end values. As noted previously, there is low confidence in dermal and inhalation unit exposure estimates for the hose-end sprayer. For the low pressure handwand, there is low confidence in dermal unit exposure estimates, and medium confidence in the inhalation unit exposure estimates.

4.2 Risk and Uncertainty Characterization of Postapplication Residential Exposures

To calculate the potential risk to persons from postapplication exposures to chlorpyrifos HED used the NOAELs discussed previously. Average body weights of 70 and 15 kg were assumed for adults and children, respectively. As noted previously, the registrant submitted four studies addressing residential postapplication exposures. These studies were used to estimate exposures and risks to residents. One study evaluated residential exposures following crack, crevice and spot treatment of the kitchen and bathroom for cockroach control. Two additional studies, evaluated lawn application (liquid and granular), while another study monitored air levels for one year following termiticide treatment. Where relevant, exposure estimates were based on biological monitoring data (i.e., lawn studies, crack and crevice study) and hand/oral exposure derived from handwash data (i.e., lawn studies). Other exposures were calculated based on environmental measurements (i.e., termiticide use). In the absence of data, the Draft Residential SOPs were used to estimate exposures and risks. The risk estimates are presented in Tables 3 and 4.

HED is in the process of revising the Residential Exposure Assessment SOPs. This process may identify specific areas of further concern with respect to chlorpyrifos and exposure to the general population. For example, some of the secondary exposure pathways that EPA is currently addressing include exposures resulting from residue tracked into homes from outdoor use, indoor dust, and spray drift. In a recent study, polycyclic aromatic hydrocarbons (PAHs) that are abundant in house dust were shown to increase the toxicity of chlorpyrifos in vitro, particularly at low levels (i.e., 2-50 μM PAHs with 1-180 nM chlorpyrifos-oxon, a metabolite of chlorpyrifos that inhibits acetyl cholinesterase) (Jett et al. 1999). Currently, there are no SOPs available to evaluate these potential exposure pathways. These scenarios however, may be evaluated in the future pending revisions to the residential SOPs.

There is insufficient use information and exposure data to assess exposure resulting from use in vehicles (i.e., planes, trains, automobiles, buses, boats) and other current label uses such as treatment of indoor exposed wood surfaces, supermarkets, restaurants, theaters, furniture, and draperies. However, HED has concern for these uses based on the scenarios assessed within this document.

(1) Crack and Crevice Treatment of Kitchen and Bathroom. The risks to residents following crack and crevice and spot treatment were evaluated based on a chemical-specific registrant-submitted biomonitoring study that evaluated treatment in the kitchen and bathroom. In this study, biomonitoring results were within the typical pre-exposure baseline levels and HED

concluded that the dermal and oral doses were negligible based on dislodgeable residue data and toy wipe samples in rooms adjacent to treatment. Therefore, only passive dosimetry inhalation dose estimates based on air sampling were available. As shown on Table 3, short- and intermediate-term inhalation MOEs for doses following crack and crevice treatment range from 560 to 670 for adults to 130 to 360 for children. Only the child inhalation MOE for the maximum 1 day exposure exceeds HEDs level of concern of 300. As noted previously, the one day dose estimate for a child may be conservative because it assumes a child spends 21 hours exclusively in the room with the highest detected air concentration.

The Dow AgroSciences study only evaluated exposures following treatment of the kitchen and bathrooms, while the label for this and similar products allow use in bedrooms, living rooms, closets, schools, day care centers, etc that could result in higher risks to children. Also the Dow study only evaluated small hard ball toys, and not plush toys that could possibly act as a sink for chlorpyrifos (as shown in the published literature). In addition, the study only evaluated use of 0.29% Dursban Pro, which could underestimate exposure because the label recommends concentrations up to 0.5% Dursban Pro for indoor crack and crevice treatments, and up to 1% for the control of wood-infesting insects on wood surfaces, wall voids, and voids and channels in damaged wood.

Low air concentrations were still present 10 days post treatment, however the current labels allow re-treatment every 7 days. This study has not yet addressed the possible cumulative effects of multiple treatments over time. (This information has been requested from the registrant). In one house, the highest daily average air concentrations were detected on day 6 indicating possible sinks, or resuspension.

(2) Crack and Crevice Treatment of Other Rooms. Because the registrant-submitted study does not adequately address exposures associated with all the uses listed on this and similar product labels, HED also evaluated exposures using the Residential SOPs in conjunction with residue data from this biomonitoring study. The resulting MOEs are all less than 300, and therefore exceed HEDs level of concern. The SOP-calculated values are, however, considered conservative because they use high-end exposure assumptions (i.e., transfer coefficients, and exposure time for contacting a surface). Nevertheless, in the absence of additional data, these SOP-estimated MOEs suggest a health concern for crack and crevice treatment in schools, day care centers, playhouses or other rooms that children may occupy for extended periods of time.

(3) Pet Collar Uses. The residential SOPs were used to assess pet collar exposures due to an absence of chemical-specific data. Residential postapplication MOEs for both cat and dog pet collar products containing 3-9% ai chlorpyrifos are below 300 (MOEs range from 8 to 150) if long-term exposure is assumed to occur through both dermal and inhalation exposure. However, pet collar MOEs are all above 300 (range from 530-2500) if exposure is assumed to be exclusively through the dermal route, except for children exposed to the 9% a.i. dog collar (MOE is 140). Because the Residential SOPs were used to evaluate pet collar use, using conservative assumptions, it is likely that these values over estimate the true exposure and risk. However, at this time HED does not have information that could further refine these estimates. This analysis also does not evaluate potential oral exposures that could result from a child mouthing or chewing on the flea collar, although most labels explicitly state that children should not be allowed to handle or play with the flea collar. Scientists at the Mississippi State initiated a study in April 1999 to evaluate exposures from pet collars containing chlorpyrifos (Personal communication D.

Smegal with J. Scott Boone, Research Toxicologist, Center for Environmental Health Sciences, College of Veterinary Medicine, Mississippi State, March 17, 1999).

(4) Termiticide Treatment. Based on a chemical-specific registrant-submitted study, the short-, intermediate- and long-term MOEs for adult residents exposed to chlorpyrifos vapor concentrations for various time intervals following a subterranean termiticide control treatment are above 300 for crawlspace, basement, plenum and slab construction homes and range from 420 to 3700. Therefore, these MOEs do not exceed HEDs level of concern. In addition, the inhalation MOEs for a child in a crawlspace home are above 300 (410 to 770). However, some of the MOEs for children are below 300 for basement, plenum and slab construction homes (MOEs range from 130 to 1100). These MOEs maybe conservative because they assume a child spends 21 hours per day at home.

The Dow AgroSciences study measured air concentrations for up to one year postapplication in four types of homes (n=8/house type). The maximum one year average air concentrations ranged from 0.11 to 0.29 $\mu g/m^3$. Studies in the published literature measured slightly higher air concentrations (average of kitchen and bedroom) of 1.32-3.13 $\mu g/m^3$ at one year postapplication, and 0.1 to 0.3 $\mu g/m^3$ eight years postapplication in homes of similar construction (slab and crawl construction) (Wright et al. 1988, 1994). It should be noted that all of these studies evaluate exposures resulting from treatment of soil outside the home, and do not evaluate the potentially higher exposures that could result from indoor treatment of a termiticide infestation.

(5) Insecticidal Dust Treatment. No data are available to evaluate the postapplication residential exposures and risks associated with the use of insecticidal dust products indoors. In addition, there are no recommended procedures for evaluating these products in the Residential SOPs. Nevertheless, HED has concerns about the use of these products based on the relatively low MOEs calculated for residents or workers that could apply these products. HED recommends that the registrant provide additional information on the potential postapplication residential exposures associated with these products.

(6) Lawn Treatment with a Liquid Spray. A chemical-specific registrant-submitted biomonitoring study was used to assess residential exposure following lawn treatment with a liquid spray. The total short-term MOEs for adults and children exposed to lawn treated with 0.29% chlorpyrifos spray range from 7.5 to 9, and exceed HEDs MOE level of concern (i.e., MOE less than 300). Both the dermal and inhalation MOEs also exceed HEDs level of concern and range from 10 to 190. The oral MOE for children of 400 is not of concern.

(7) Lawn Treatment with a Granular Insecticide. A chemical-specific registrant-submitted biomonitoring study was used to assess residential exposure following lawn treatment for a granular insecticide. The total MOEs for adults and children exposed to lawn treated with a 0.5% granular formulation of chlorpyrifos range from 73 to 120, and also exceed HEDs MOE level of concern (i.e., MOE less than 300). The dermal MOEs, which range from 90 to 190, contribute most to the total MOEs, and also exceed HEDs level of concern. The inhalation MOEs range from 330 to 400 while the oral MOE for children is 6000.

It should be noted that the MOEs are based on central tendency dose estimates the day of treatment from state-of-the art biomonitoring studies, and therefore are not conservative. In fact, HED has concerns that the MOEs could be underestimated for young children because both lawn

studies did not adequately address incidental ingestion of soil/granules or the more frequent hand to mouth activity of children compared to adults. Oral exposures to children were estimated to be 41% of the residue on an adult's hands (based on a surface area conversion) from a one-time washing. In addition, exposures could be underestimated in some instances because these lawn-care products are used in residential areas, playgrounds, recreational areas, school yards, and golf courses, etc., and it was assumed that a child could be exposed to only one treated turf for 4 hours per day.

The Dow AgroSciences Studies (granular and liquid application) evaluated a 4 hour exposure immediately following treatment (or 4 hours after the liquid insecticide had dried). However, 2 of the hours were spent on a blanket (while sunbathing and picnicking). Also, due to the design of the biological monitoring studies, it was not possible to derive separate exposure values for subsequent days. Furthermore, transferable residue data were not available for the liquid lawn treatment beyond 48 hours after application, making extended exposure analyses impossible. In this study, there was no clear decline in residues during the 48 hours after the turf treated with liquid chlorpyrifos had dried, possibly because of technical problems associated with using a drag over a turfgrass medium. The registrant should conduct transferable residue studies on turf for a period of more than 48 hours and with more samples collected to allow the derivation of a regression for decline of transferable residues over time.

(8) Mosquitocide Use. In the absence of chemical-specific data, the scientific literature, AgDrift Model and the Draft Residential SOPs were used to assess chlorpyrifos as a mosquitocide. The resulting screening-level short-term MOEs for chlorpyrifos adult mosquito control uses indicate that MOEs are greater than 2300 for all postapplication exposure scenarios for adults and toddlers for the ground-based fogger mosquito control applications. Exposure resulting from aerial applications of Mosquitomist One ultra low volume (U.L.V) were evaluated and determined to be negligible.

(9) Yard and Ornamental Spray Treatment. By analogy, yard and ornamental spray products were evaluated and determined to result in comparable doses and short-term MOEs with the lawn care products based on label uses and application rates. Therefore, use of many of these products is likely to result in MOEs that exceed HEDs level of concern.

Table 2. Estimates of Exposures and Risks to Commercial Applicators and Residents Applying Chlorpyrifos in the Residential Environment

Application Scenario	Unit Exposure (μg/lb ai)		Lb ai Handled	Central Tendency Dose (μg/kg/day) (a)		MOE (b)		
	Dermal	Inhalation		Dermal	Inhalation	Dermal	Inhalation	Total
(1) Indoor Crack & Crevice Treatment								
Long term PCO with PPE (double layer clothes, chemically-resistant boots and gloves, eye protection) (c) **(0.29% Dursban Pro, EPA Reg. 62719-166)**	1790	532	Mean = 0.02	0.51	0.15	59	197	45
			Min = 0.0002	0.005	0.0015	5900	20000	4500
			Max = 0.0684	1.75	0.52	17	58	13
Short-term Residential Applicator (SS, SP, no gloves) (Residential SOPs) (p) **(EPA Reg 026693-00003 (1%), 239-2619 (0.5%))**	220000	2400	0.01 (1%ai at 16 oz)	31.4	0.34	159	292	100
			0.005 (0.5% ai at 16 oz)	15.7	0.17	318	584	200
			0.00063 (0.5% at 2 oz)	1.96	0.02	2540	4700	1600
(2) Broadcast Turf Application (Intermediate and Long-Term for PCOs; Short-Term for Residential Applicators) (0.12% Dursban Pro, EPA Reg. 62719-166 for PCOs and Dursban 1-12 Insecticide EPA Reg. 62719-56 for Residents)								
Applicator with PPE (d) (single layer clothes, chemically-resistant boots and gloves, hat)	NA	NA	Mean= 2.17 (1.57-2.95)	Total: 0.4 (biomonitoring)(j)		Biomonitoring: 75 (k)		
				0.8 (label max) (j)		Label Max: 38 (j, k)		
Mixer/Loader (liquid) (Single layer clothes, gloves)(i)	23	1.2	2.95 (l)	0.029(m)	0.05 (m)	1032	1980 (IT)	680 (IT)
							600 (LT)	380 (LT)
				0.058 (j)	0.1 (j)	516	990 (IT)	340 (IT)
							300 (LT)	190(LT)

Table 2. Estimates of Exposures and Risks to Commercial Applicators and Residents Applying Chlorpyrifos in the Residential Environment

Application Scenario	Unit Exposure (μg/lb ai)		Lb ai Handled	Central Tendency Dose (μg/kg/day) (a)		MOE (b)		
	Dermal	Inhalation		Dermal	Inhalation	Dermal	Inhalation	Total
Mixer/Loader (liquid) (double layer clothes, gloves)(i)	17	1.2	2.95 (l)	0.021(m)	0.05 (m)	1400	1980 (IT) 600 (LT)	820 (IT) 420(LT)
				0.042 (j)	0.1 (j)	700	990 (IT) 300 (LT)	410 (IT) 210(LT)
Residential Mixer/Loader/Applicator Broadcast with Hose End Sprayer (SS, SP, no gloves) (Residential SOPs)	30000	9.5	0.5 (min. 3 oz/gal)	214 (f)	0.07	23	1470	23
			2 (max 12 oz/gal)	857 (f)	0.27	6	368	6
Residential Mixer/Loader/Applicator Spot treatment with Low Pressure Handwand (SS, SP, no gloves) (Residential SOPs)	100000	30	0.094	134 (f)	0.04	37	2490	37
(3) Ready-to-Use Formulated Product (Ortho Ant Stop) (n)								
Short-term Residential Applicator (SS, LP, no gloves)	NA	NA	7.3E-5	7	0.029	714	3,448	590
(4) Insecticidal Dust Product (Shaker Can or Bulbous Duster)								
Residential Applicator (10 oz can of 1% ai chlorpyrifos; 2.83 g ai) (EPA Reg. 62719-66, 62719-54 and 192-171)								
Short-term Residential Applicator (SS, LP, no gloves)	2200000	NE	0.024	20 (f,o)	NE	250	NE	250

Table 2. Estimates of Exposures and Risks to Commercial Applicators and Residents Applying Chlorpyrifos in the Residential Environment

Application Scenario	Unit Exposure (μg/lb ai)		Lb ai Handled	Central Tendency Dose (μg/kg/day) (a)		MOE (b)		
	Dermal	Inhalation		Dermal	Inhalation	Dermal	Inhalation	Total
Worker (4 oz or 100 oz of 7% ai chlorpryifos; 7.91 or 198 g ai) (EPA Reg. 13283-17, Rainbow Kofire Ant Killer)								
Short-term Exposure (LS, LP, gloves)	2000000		0.024	51 (f,o) (4 oz)	NE	98	NE	98
				1275 (f,o) (100 oz)		3.9	NE	3.9
Intermediate-term Exposure (LS, LP, gloves)				1.5 (g,o) (4 oz)	NE	20	NE	20
				38 (g,o) (100 oz)		0.8	NE	0.8
(5) Granular Formulation (Hand Application) (PHED V1.1, Residential SOPs) (EPA Reg. 62715-14, 62715-210)								
LCO (LS,LP, gloves) (intermediate-term)	71000	470	0.0459	1.4 (g)	0.31	21	324	20
Residential Applicator (SS, SP, no gloves) (short-term)	430000	467	0.0459	282 (f)	0.31	18	327	17
(6) Granular Formulation (Belly Grinder) (PHED V1.1, Residential SOPs) (EPA Reg. 62715-14, 62715-210)								
LCO (LS,LP, gloves) (intermediate-term)	9300	62	0.97	3.9 (g)	0.9	8	120	7
Residential Applicator (SS, SP, no gloves) (short-term)	110000	62	0.97	1520 (f)	0.9	3	120	3
(7) Granular Formulation (Push-type Spreader) (PHED V1.1, Residential SOPs) (EPA Reg. 62715-14, 62715-210)								
LCO (LS,LP, gloves) (intermediate-term)	1270 (h)	6.3	0.97	0.5 (g)	0.09	57	1150	54
Residential Applicator (SS, SP, no gloves) (short-term)	3000	6.3	0.97	42 (f)	0.09	120	1150	110

Table 2. Estimates of Exposures and Risks to Commercial Applicators and Residents Applying Chlorpyrifos in the Residential Environment

Application Scenario	Unit Exposure (μg/lb ai)		Lb ai Handled	Central Tendency Dose (μg/kg/day) (a)		MOE (b)		
	Dermal	Inhalation		Dermal	Inhalation	Dermal	Inhalation	Total
Termiticide Treatments (PCOs with PPE)								
(8) Pre-Construction (1.44% ai chlorpyrifos as Dursban TC, EPA Reg. 62719-47) (Long-term) (e)								
M/L/A (single layer clothes; forearm length gloves) (3 hour average exposure) (dosimetry)	NA	NA	NA	1.57	0.45	19	67	15
M/L/A (double layer clothes; forearm length gloves) (3 hour average exposure) (dosimetry)	NA	NA	NA	0.477	0.45	63	67	33
Tarp puller (with forearm-length gloves) (dosimetry)	NA	NA	NA	1 tarp: 0.023	1 tarp: 0.021	1322	1430	690
				8 tarps: 0.177	8 tarps: 0.168	169	179	87
Tarp puller (without gloves) (dosimetry)	NE	NE	NE	1 tarp: 0.081	1 tarp: 0.015	373	1961	310
				8 tarps: 0.644	8 tarps: 0.122	47	245	39
(9) Post-Construction (1% ai chlorpyrifos as Dursban TC) (EPA Reg. 62719-47) (long-term) (r)								
Mixer/Loader/ Applicator (PPE =LS, LP, chemically resistant gloves, hat, eye protection and half facepiece respirator in confined spaces; during M/L: 2 layers clothes and chemically resistant shoes)	NA	NA	10.72 (4-32.7)	biomonitoring: 4.3		7		7
				Dosimetry: 2.5	Dosimetry: 0.91 (no protection)	12	33	9

Table 2. Estimates of Exposures and Risks to Commercial Applicators and Residents Applying Chlorpyrifos in the Residential Environment

Application Scenario	Unit Exposure (μg/lb ai)		Lb ai Handled	Central Tendency Dose (μg/kg/day) (a)		MOE (b)		
	Dermal	Inhalation		Dermal	Inhalation	Dermal	Inhalation	Total
(10) Paint Brush (Residential SOPs) (Short-term) (Dursban 1-12 Insecticide, EPA Reg. 62719-56)								
Residential Applicator (SS, SP, no gloves)	230000	284	0.0416 (1 gallon)	140 (f)	0.17	37	590	35
			0.0104 (1 quart)	34 (f)	0.043	148	2300	140
(11) Ornamental Application (Residential SOPs) (Short-term) (Dursban 1-12 Insecticide, EPA Reg. 62719-56)								
Residential Mixer/Loader/Applicator Low pressure Handwand (SS, SP, no gloves)	100000	30	0.013 (min. 1 oz/3 gal H20)	18.6 (f)	0.0056	269	17950	270
			0.05 (typical 4 oz/3 gal H20)	71 (f)	0.021	70	4670	69
			0.416 (max. 1 qt/3 gal H2O)	594 (f)	0.178	8	561	8
Residential Mixer/Loader/ Applicator Hose End Sprayer (SS, SP, no gloves)	30000	9.5	0.013 (min. 1 oz/3 gal H20)	5.6 (f)	0.0018	897	56700	880
			0.05 (typical 4 oz/3 gal H20)	21 (f)	0.0068	233	14700	230
			0.416 (max. 1 qt/3 gal H2O)	178 (f)	0.0565	28	1770	28

SS= short-sleeves; LS = long sleeves; LP= long pants, SP = short-pants; IT = intermediate term; LT = long term.
NA = Not applicable
NE = Not evaluated
M/L/A = Mixer/Loader/Applicator

(a) Average dose presented, unless otherwise specified. Range of exposure is presented in parentheses. Average dose (μg/kg/day) = average unit exposure (μg/lb ai) * Lb ai handled * dermal absorption factor (intermediate and long term) / 70 kg body weight. Data from PHED is the "best fit" mean exposure (i.e., geometric mean for lognormal distributions, arithmetic mean for normal distributions and median for other distribution types).

(b) MOE = NOAEL/ Dose, where the acute oral NOAEL is 500 μg/kg/day (1 day); short-term dermal NOAEL is 5000 μg/kg/day (less than 7 days), intermediate- and long-term dermal NOAELs are 30 μg/kg/day (greater than 7 days), short- and intermediate inhalation NOAEL is 100 μg/kg/day (1day to several months), and long-term inhalation NOAEL is 30 μg/kg/day (greater than several months). Acceptable MOE 100 for commercial PCOs and 300 for residents, which accounts for 10X for interspecies 10X extrapolation for intra-species variability and an FQPA factor of 3. Values rounded to two significant figures.

(c) Exposures based on MRID No. 444448-01 biomonitoring study of PCOs applying 0.29% ai chlorpyrifos wearing the label-specified PPE for crack and crevice applications; therefore no baseline is available. Dermal exposure already adjusted for 3% dermal absorption. The full range of exposures and

MOEs are reported, because there is insufficient information available on the distribution of actual product handled by PCOs in the field.

(d) Exposures based on MRID No. 447294-01, biomonitoring study using 0.12 Percent Chlorpyrifos Spray with PCOs wearing the label-specified PPE for turf application; therefore no baseline is available.

(e) Exposures based on registrant study MRID No. 44589001. Average exposure for M/L/A is 3 hours. Average 7 min exposure for tarp pullers were multiplied by 8, to assume a worker could pull 8 tarps in a work day.

(f) Short-term dermal dose does not adjust for dermal absorption because the short-term dermal NOAEL of 5 mg/kg/day is based on a 21-day rat dermal study.

(g) Intermediate-term dermal dose was adjusted for absorption assuming 3% dermal absorption for comparison with the intermediate-term oral NOAEL of 0.03 mg/kg/day.

(h) Unit exposures from PHED were adjusted to account for 90% protection from gloves.

(i) In the absence of chemical-specific data, surrogate unit exposures obtained from PHED, Version 1.1 were used.

(j) The biomonitoring study applied the 0.12% Dursban Pro (EPA Reg. 62719-166) at a rate of 2 gallons/1000 ft2, when the label recommends a maximum application rate of 4 gallons/1000 ft2 for subsurface soil treatment. Therefore, the exposures were conservatively adjusted upwards by a factor of 2 (i.e., normalized to the maximum rate) to account for the difference in application rate.

(k) The exposure estimates were compared to the intermediate and long-term dermal and long-term inhalation NOAEL of 30 μg/kg/day because there is insufficient information to determine if exposures are intermediate or long-term.

(l) Maximum quantity handled from biomonitoring study (MRID No. 44729401).

(m) Absorbed Dermal Dose (μg/kg/day) = Unit exposure (μg/lb ai) * amount handled (2.95 lb ai) * dermal absorption factor (0.03) / 70 kg body weight.

(n) Exposures based on biomonitoring data from MRID No. 44739301, using the geometric mean of 0.24 ug/kg. Passive dosimetry results were used to segregate exposure into dermal and inhalation components due to different toxicity endpoints (see text). Short-term dermal exposure was further adjusted using a 3% dermal absorption factor to obtain a dermal exposure estimate for comparison with the short-term dermal endpoint of 5000 ug/kg.

(o) Exposure estimates based on a study that evaluated the application of a dust product to a home garden (Kurtz and Bode 1985), where exposure was normalized for chlorpyrifos exposure. Exposures are predominantly dermal. See text.
Residential Handler Dose (μg/kg/day) =(deposition in study (4.9 mg/10 g ai carbaryl) * 2.83 g ai chlorpyrifos* 1000 μg/mg) / 70 kg
Worker Dose (μg/kg/day) =(deposition in study (4.5 mg/10 g ai carbaryl) * 7.91or 198 g ai chlorpyrifos* 1000 μg/mg) / 70 kg

(p) Exposure based on Residential SOPs, and assumes the application of a 16 oz aerosol can that contains 1% or 0.5% ai chlorpyrifos.

(q) Value based on the average amount of active ingredient handled in the 55 replicates of dispensing granular bait from the studies in PHED.

(r) Exposure estimates based on MRID No. 44729402. Biomonitoring results based on 5 individuals, dosimetry data based on 15 individuals.

Table 3. Estimates of Postapplication Exposures and Risks to Residents				
Reentry Scenario	**Central Tendency Dose (μg/kg/day) (a)**		**MOE (b)**	
	Adult (70 kg)	**Child (15 kg)**	**Adult**	**Child**
(1) Crack & Crevice Treatment of Kitchen and Bathroom (Dursban Pro EPA Reg. 62719-166) (c) (Short-and Intermediate-term)				
Maximum 1-Day Inhalation Exposure:	0.18 (0.075- 0.39)	0.76 (g)	560	130
10-Day TWA Inhalation Exposure	0.15 (g)	0.28 (g)	670	360
(2) Crack & Crevice Treatment of Other Rooms Using Residential SOPs (Dursban Pro, EPA Reg. 62719-166) (o) (Short-term)				
Dermal Exposure From Carpets (p)	56.5	53.4	88	94
Dermal Exposure From Surfaces (p)	28.2	26.7	177	187
Oral Exposure (f)	NE	1.67	NE	299
(3) Pet Collar Uses (11 month efficiency) (long-term)				
Dog: Collar (EPA No. 45087-40; 3.44 g ai)				
Dermal	0.022 (I)- 0.045 (h)	0.1 (I)- 0.21 (h)	670 (h) - 1300 (I)	140 (h) -290 (I)
Inhalation	0.74	3.47	40	9
Total Exposure (l)	0.76	3.6	39	8
Cat Collar (EPA No. 4306-16; 0.93 g chlorpyrifos)				
Dermal	0.006 (I)- 0.012 (h)	0.028 (I)- 0.056 (h)	2500 (h) - 5000 (I)	530 (h) -1100 (I)
Inhalation	0.20	0.93	150	32
Total Exposure (l)	0.206	0.96	150	31
(4) Termiticide Treatment (See Table 4)				
(5) Insecticidal Dust Products (Insufficient data to evaluate; see text)				
Broadcast Turf Application (Short-term)				
(6) 0.29 Percent Chlorpyrifos Spray (Dursban Turf Insecticide) (d)				
Inhalation	0.59	5	170	20
Dermal (k)	510	414	10	12
Oral	NE	1.26	NE	400
Total Absorbed Dose	6.3 (3.5-8.9)	10 (7.9-13)	9 (m)	7.5 (m)

Table 3. Estimates of Postapplication Exposures and Risks to Residents				
Reentry Scenario	**Central Tendency Dose (μg/kg/day) (a)**		**MOE (b)**	
	Adult (70 kg)	**Child (15 kg)**	**Adult**	**Child**
(7) Granular Formulation of 0.5% Chlorpyrifos (Dursban Insecticide) (e)				
Inhalation	0.3	0.25	330	400
Dermal	27	56	190	90
Oral	NE	0.085	NE	6000
Total Absorbed Dose	1.4 (0.56 - 3.7)	2 (0.75 - 5)	120 (m)	73 (m)
(8) Aerial and Ground-Based Fogger Adult Mosquitocide Application (Mosquitomist One EPA Reg. 8329-24) (n) (short-term)				
Dermal	1.38	1.3	3600	3800
Oral (hand to mouth)	NE	0.0816	NE	6100
Oral (Turfgrass Ingestion)	NE	0.0093	NE	54000
Oral (Soil Ingestion)	NE	0.000025	NE	2000000
Total Exposure	1.38	1.39	3600	2300
(9) Yard and Ornamental Sprays (Evaluated based on analogy to Lawn Products; see text)				

NE = Not evaluated because exposure not of concern for adults
TWA = Time weighted average.

(a) Average dose presented, unless otherwise specified. Range of doses is presented in parentheses

(b) MOE = NOAEL/ Dose, where the acute oral NOAEL is 500 μg/kg/day (1 day); short-term dermal NOAEL is 5000 μg/kg/day (less than 7 days), intermediate- and long-term dermal NOAELs are 30 μg/kg/day (greater than 7 days) (absorbed dose), short- and intermediate inhalation NOAEL is 100 μg/kg/day (1day to several months), and long-term inhalation NOAEL is 30 μg/kg/day (greater than several months). Acceptable MOE 300, which accounts for 10X for interspecies 10X extrapolation for intra-species variability and an FQPA factor of 3. Values rounded to two significant figures.

(c) MRID 44458201. Doses based on biomonitoring and environmental measurements.

(d) MRID 43013501. Doses based on oral, dermal and inhalation exposure based on biomonitoring and environmental measurements. Dose estimated for the day of application only. (See text). Child doses adjusted from original HED review to reflect 1-6 year old child (1.24 m3/day, 15 kg body weight and 0.41 child hand factor ratio relative to adult).

(e) MRID 44167101. Oral, dermal and inhalation dose based on biomonitoring and environmental measurements. Dose estimated for the day of application only. (See text). Dermal absorbed dose from biomonitoring data adjusted to dermal exposure, assuming 3% absorption factor, for direct comparison with dermal NOAEL of 5000 ug/kg from rat dermal study.

(f) Oral hand to mouth dose (μg/kg/day) = available surface residue (1.15E-2 μg/cm2) * surface area of hands (350 cm2) * frequency of hand contact (1.56 events/hr) * exposure time (4 hrs/day) / body weight (15 kg for a child)

(g) Estimate based on the air concentrations detected in house #2, which were higher than those detected in houses #1 and 3.

(h) Dose estimates modified from EPA Review DP Barcode: D253246 (D. Smegal to J. Rowland, March 1, 1999), based on body weight. Assumes 100% dermal exposure.

(i) Dose estimates modified from EPA Review DP Barcode: D253246 (D. Smegal to J. Rowland, March 1, 1999), based on body weight. Assumes 50% dermal exposure and 50% inhalation exposure.

(j) Mean dose is based on mean biomonitoring data. Assumes 100% inhalation exposure.

(k) Absorbed dermal dose readjusted to dermal exposure for direct comparison with the dermal NOAEL of

5000 μg/kg from the dermal rat study. Original HED review estimated absorbed dermal dose assuming a 1% dermal absorption factor.

(l) Total dose assuming 50% dermal and 50% inhalation exposure.

(m) Total MOE = 1 / [(1/MOE inhalation) + (1/MOE dermal) + (1/MOE oral)].

(n) Doses and MOEs based on the application rate of 0.01 lb ai/acre. Inhalation dose was considered to be negligible because of infinite dilution that is anticipated in an outdoor application and based on the very low application rate.

(o) Doses estimated using the highest deposition residue of 2.298 μg/100cm2 in the family room of house number (room adjacent to the treated kitchen). It was assumed that 50% of the residue is available as dislodgeable residue in accordance with the Residential SOPs.

(p) Dermal dose from carpet/surfaces (μg/kg/day) = [available surface residue (0.0115 μg/cm2) * TC (cm2/hr) [43,000 for adults and 8700 for children] * Exposure time (hr/day) [8 hrs/day for carpet and 4 hr/day for surfaces]] / body weight (70 kg for adults and 15 kg for a child).

Table 4
Indoor Chlorpyrifos Air Concentrations and Estimated Exposures and Risks to Residents After Subterranean Termite Control Treatment (a)

Construction Type	Air Concentration (μg/m3) (b)	Dose (μg/kg/day) (d)		MOE (e)	
		Adult (70 kg)	Child (15 kg)	Adult Male	Child
Crawlspace					
Day 1 (c)	0.31	0.046	0.15	2200	670
Day 7 (days 2-7)	0.33	0.049	0.16	2000	630
Day 30 (days 8-30)	0.26	0.039	0.13	2600	770
Day 90 (days 31-90)	0.34	0.05	0.16	2000	630
1 year (days 91-365)	0.15	0.022	0.073	1400	410
Basement					
Day 1 (c)	1.36	0.2	0.66	500	150
Day 7 (days 2-7)	0.77	0.11	0.37	910	270
Day 30 (days 8-30)	0.7	0.1	0.34	1000	290
Day 90 (days 31-90)	0.41	0.061	0.2	1600	500
1 year (days 91-365)	0.29	0.043	0.14	700	210
Plenum					
Day 1 (c)	1.6	0.24	0.77	420	130
Day 7 (days 2-7)	1.56	0.23	0.76	430	130
Day 30 (days 8-30)	1.37	0.2	0.66	500	150
Day 90 (days 31-90)	0.23	0.034	0.11	2900	910
1 year (days 91-365)	0.17	0.025	0.082	1200	370
Slab					
Day 1 (c)	0.87	0.13	0.42	770	240
Day 7 (days 2-7)	0.46	0.068	0.22	1500	450
Day 30 (days 8-30)	0.18	0.027	0.087	3700	1100
Day 90 (days 31-90)	0.32	0.047	0.15	2100	670
1 year (days 91-365)	0.11	0.016	0.053	1900	570

(a) Estimates were derived from a registrant-submitted air monitoring study (MRID No. 40094001)

(b) Air concentrations represent the mean value of the maximum detected concentration from 8 houses of similar construction type.

(c) Time weighted average of the 2, 4, 8 and 24 hour measurements.

(d) Dose calculated as follows: dose (μg/kg/day) = air conc (μg/m^3) * inhalation rate (m^3/day) * hours per day in house/24 hours * 1/body weight (kg). Assumptions are as follows: respiratory volumes of 15.2, and 8.3 m^3/day for an adults and 3 yr old child (average of male and female), respectively (Exposure Factors Handbook 1997 p. 5-24), and body weights of 70 and 15 kg, respectively. In addition, it assumes that adults and children spend 16.4 and 21 hours per day at home, respectively (Exposure Factors Handbook 1997 p.15-17, 15-147)

(e) MOE = NOAEL / dose, where short- and intermediate-term inhalation NOAELs =100 μg/kg/day (1 day to several months) and long-term inhalation NOAEL =30 μg/kg/day (several months to years). Acceptable MOE 300, which accounts for 10X for interspecies extrapolation, 10X for intra-species variability and an FQPA factor of 3. Values rounded to two significant figures.

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EXHIBIT 4

Dursban Announcement

06/08/2000

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY

WASHINGTON, D.C. 20460

Administrator Carol M. Browner
U.S. Environmental Protection Agency
Dursban Announcement

Remarks Prepared for Delivery
June 8, 2000
Washington, D.C.

Today, the Clinton-Gore Administration is announcing a major step to improve safety for all Americans from the health risks posed by pesticides. We are eliminating virtually all home and garden uses of Dursban – the most widely used household pesticide in the United States.

This action comes after completing the most extensive scientific review of the potential hazards from a pesticide ever conducted. This action -- the result of an agreement with the manufacturers -- will significantly minimize potential health risks from exposure to Dursban, also called chlorpyrifos, for all Americans, especially children.

This action is good news for the protection of our country's public health. It is good news for the environment. And it is particularly good news for children, who are among the most vulnerable to the risks posed by pesticides.

In 1993, the Clinton-Gore Administration went to Congress with a plan -- based on recommendations from the National Academy of Sciences -- to better protect our families from the risks of pesticides by making children's health the benchmark for safety. We did this because children are not just small adults. Their bodies are still developing and more susceptible to risks from toxic chemicals. They play on floors and in yards where pesticides have been applied. And they eat proportionately more food with respect to body weight than do adults. When our health and safety standards protect children, the entire public is protected.

Three years after Congress received this plan from the Clinton-Gore Administration, it unanimously passed the Food Quality Protection Act.

President Clinton signed it into law in 1996. Last summer EPA took the first actions under the new law against two pesticides that posed the greatest threats to children at that time: methyl parathion and azinphos methyl. Today we are taking action against chlorpyrifos – the most commonly used pesticide in homes, buildings and schools.

Dursban is the common trade name, but it is one product of more than 800 containing the chemical chlorpyrifos, which today's action affects. Chlorpyrifos is commonly found in many home-and-garden bug sprays. It is used in some treatments of termites. And it is used on some agricultural crops. It belongs to a family of older, riskier pesticides called organophosphates, some of which date back 50 years or more. The time has come to review these pesticides for safety, and, where the science dictates, remove those chemicals that pose an unreasonable threat to human health and move to newer, safer alternatives. That is what we are doing today.

With today's announcement, we are taking the fastest action possible for removing these household products from the market: This action:

-- will virtually eliminate home, lawn and garden uses by the end of the year.

-- It will virtually eliminate all termite-control uses in existing homes by the end of the year.

-- It will eliminate this year the use of chlorpyrifos for all sensitive areas, such as schools, day cares, parks, hospitals, nursing homes and malls by the end of the year.

-- It will eliminate or dramatically lower pesticide residues on several foods by next growing season.

-- And, finally, it will eliminate the use of chlorpyrifos as a termiticide for new home-and-building construction by the end of 2004.

I am pleased that the major manufacturers, Dow AgroSciences and others, have entered into this agreement to ensure that the risk reductions we are seeking will begin as quickly as possible.

Today's action is part of an overall commitment by the Clinton-Gore Administration to protect public health and the environment that begins with our children. The protection of children has guided the actions we've taken for cleaner air to breathe. The protection of children has guided the actions we've taken for safer drinking water. And the protection of children has guided the actions we are taking for safer pesticide use. Today's action represents another

significant step in safeguarding the health of our children, and therefore the health of all Americans.

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09/15/2009 Administrator Lisa P. Jackson, Remarks on Proposed Fuel Efficiency Rules for Autos, As Prepared

EXHIBIT 5

PESTICIDES IN THE AIR – KIDS AT RISK:
Petition to EPA to Protect Children From Pesticide Drift

Submitted on behalf of
United Farmworkers, Pesticide Action Network of North America,
Physicians for Social Responsibility, MomsRising,
Pineros y Campesinos Unidos del Noroeste,
Sea Mar Community Health Center,
California Rural Legal Assistance Foundation,
and Farm Labor Organizing Committee

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THE U.S. ENVIRONMENTAL PROTECTION AGENCY HAS FAILED TO PROTECT CHILDREN FROM PESTICIDE DRIFT AND MUST TAKE IMMEDIATE STEPS TO CORRECT THESE VIOLATIONS OF FEDERAL PESTICIDE LAWS

This petition asks the U.S. Environmental Protection Agency ("EPA") to remedy ongoing violations of its legal obligations to protect children from unsafe aggregate exposures to pesticides. Specifically, EPA has failed to protect children from exposure to toxic pesticides that drift from agricultural fields and contaminate areas where children congregate, such as homes, park, schools, and daycare centers. To ensure that children are protected from toxic pesticides as required by the law, this petition asks EPA to:

(1) expeditiously evaluate the exposure of children to pesticide drift and impose safeguards to ensure that children are protected from aggregate pesticide exposures, including pesticide drift;[1] and

(2) immediately adopt interim prohibitions on the use of toxic drift-prone pesticides such as organophosphates and n-methyl carbamates near homes, schools, parks, and daycare centers or wherever children congregate.

EXECUTIVE SUMMARY

I. CHILDREN ARE PARTICULARLY VULNERABLE TO PESTICIDES.

In 1993, the National Academy of Sciences ("NAS") released a pivotal study on the heightened vulnerabilities of children to pesticides. The study criticized EPA for treating children like "little adults" and for failing to address their unique susceptibility to pesticides and their exposures based on the foods they eat and their play and exploration activities. Children

[1] The term "drift" as used in this petition includes any airborne movement of pesticides away from a target site during and/or after application, including airborne movement of pesticide droplets, pesticide powders, volatilized vapor-phase pesticides, and pesticide contaminated soil particles.

are especially vulnerable to harm from pesticides because they are growing and developing, eat and drink more per body weight than adults, consume large amounts of certain foods, and engage in activities that increase their exposure such as frequently putting hands or objects into their mouths. NAS recommended that EPA revise its pesticide regulations to account for children's vulnerabilities, consumption patterns, and "exposures from all sources – not just ingestion . . . ."[2]

II. CONGRESS DIRECTED EPA TO ENSURE THAT CHILDREN ARE PROTECTED FROM PESTICIDES FROM ALL SOURCES BY THE END OF 2006.

Congress heeded the NAS recommendations and unanimously passed the Food Quality Protection Act in 1996. That law requires EPA to "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure" to pesticides.[3] "Aggregate exposure" includes "all anticipated dietary exposures and all other exposures for which there is reliable information," including pesticide drift exposures.[4] Congress gave EPA an August 2006 deadline to bring all pesticides used on foods into compliance with these protective mandates.

III. EPA IGNORED CHILDREN WHO ARE POISONED BY SPRAY DRIFT AND VOLATILIZATION DRIFT WHERE THEY LIVE, GO TO SCHOOL, AND PLAY.

To comply with the new FQPA requirements, EPA developed methods to estimate child and infant pesticide exposures from a variety of sources, including crawling and playing on treated lawns and carpets, putting their hands into their mouths, and playing with pets treated with flea shampoos. These assessments led EPA to cancel numerous home uses of pesticides because of excessive risks to children.

Pesticide drift is another significant route of exposure for children, particularly those who

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[2] NAS, Pesticides in the Diets of Infants and Children, at 307 (1993), Attachment 1 (hereinafter "NAS Report"). This and all attachments are located on the enclosed CD.

[3] 21 U.S.C. §§ 346a(b)(2)(C)(ii)(I), (II).

[4] 21 U.S.C. § 346a(b)(2)(A)(ii) (emphasis added); see also 21 U.S.C. § 346a(b)(2)(C)(vi).

live in agricultural areas. The 1993 NAS study on children's risks from pesticides found that agricultural pesticide drift can contribute to kids' overall pesticide exposure and that airborne pesticide residues are generally higher in areas close to agricultural lands.[5] The California Department of Pesticide Regulation has also documented harmful exposures to the public from pesticide drift. And the Washington State Pesticide Incident Reporting and Tracking Review Panel has found that "[e]xposure to pesticide drift is an important cause of documented pesticide-related illness in Washington."[6]

Inexplicably, EPA has failed to assess children's exposures to pesticides that drift from agricultural sites to homes, schools, daycares, parks, and other places where children may be exposed. EPA's failure comes despite its acknowledgment of its obligation to protect children from drift, which can cause acute poisonings as well as cancer, long-term reproductive and developmental disorders, and other chronic adverse effects. By failing to assess the risk to children who are exposed to agricultural pesticide drift, EPA maintains a double-standard that often provides some protections for kids from pesticides used in urban and residential settings, but leaves kids who live near agricultural sites unprotected and vulnerable to pesticide drift.

In failing to protect these forgotten children, EPA has violated the Food Quality Protection Act. The agency's failure also violates executive orders directing EPA to ensure that its programs do not have disproportionate adverse health impacts on children, minority, and low-income populations.

IV. EPA MUST TAKE IMMEDIATE STEPS TO COMPLY WITH ITS LEGAL DUTY TO PROTECT ALL CHILDREN FROM PESTICIDE DRIFT.

This petition asks EPA to take two immediate steps to comply with its legal duty to

[5] NAS Report at 308-09, Attachment 1.

[6] Washington State Pesticide Incident Reporting and Tracking Review Panel, Annual Report: 2005, at 81 (May 2007), Attachment 2.

protect all children from all pesticide exposures:

First, EPA must fully evaluate drift risks for all pesticides that have the potential to move from agricultural sites to areas where children congregate, such as homes, parks, schools, and daycare centers. Based on these assessments, EPA must limit or prohibit pesticide uses that result in children being exposed to unsafe levels of pesticide particles or vapors. In order to adequately protect children, EPA must correct its violations of the FQPA and executive orders more quickly than the current set of pesticide registration reviews, which are not scheduled to be completed until 2022.

Second, to protect children while it conducts the necessary drift exposure assessments and develops pesticide-specific protective measures, EPA should impose no-spray buffer zones for dangerous drift-prone pesticides around homes, schools, parks, daycare centers, and other places where children congregate. EPA has recognized that such buffer zones are an effective method in reducing risks associated with pesticide drift. These buffers should be required for all pesticides that have the potential to drift, including the two classes of widely used nerve toxins (organophosphates and n-methyl carbamates) that cause acute poisonings. EPA has found that young children are already exposed to these two classes of pesticides at or possibly in excess of maximum safe levels, without having considered the additional exposures from drift. EPA must take immediate steps to prevent the additional unassessed drift exposures from harming children while EPA completes the drift risk evaluations.

BACKGROUND

I. PESTICIDE DRIFT POSES SIGNIFICANT RISKS TO CHILDREN'S HEALTH.

A. Children Are More Susceptible to Harm From Pesticides Than Adults.

In 1993, the National Academy of Sciences ("NAS") published a pivotal study documenting the many ways pesticides pose especially severe risks to infants and children. In

particular, NAS found that pesticides pose heightened risks to children because "[i]nfants and children are growing and developing," "[t]heir metabolic rates are more rapid than adults," and "[t]here are differences in their ability to activate, detoxify, and excrete xenobiotic compounds."[7] Children are also at heightened vulnerability because they "eat and drink more than adults" in relation to their body weight, they consume large quantities of certain fruits and vegetables, and engage in risky behaviors "such as playing on floors or lawns or putting objects in their mouths."[8] The NAS Report found that EPA had failed to assess children's unique exposures to pesticides and their special susceptibilities to the adverse health effects of such exposures at various stages of development.[9] Recent EPA-funded research confirms that children can be much more vulnerable to pesticide exposure than adults.[10]

B. EPA Has Long Recognized That Drift Exposures Can Be Harmful to Children.

One of the many routes by which children are exposed to pesticides is through pesticide drift. In its 1993 Report on children and pesticides, NAS observed that "[e]xposure to pesticide residues from ambient air sources is generally higher in areas close to agricultural lands and in communities surrounding pesticide manufacturing factories" and also "movement of the more volatile chemicals present potentially significant human exposure."[11] To guard against harms

[7] NAS Report at 3, Attachment 1.

[8] EPA, Pesticides and Food: Why Children May be Especially Sensitive to Pesticides (Mar. 2008), Attachment 3.

[9] NAS Report at 3-7 (1993), Attachment 1.

[10] E.g., Centers for Children's Environmental Health & Disease Prevention Research, Exposures & Health of Farm Worker Children in California, Attachment 4; see also EPA, Children's Exposure to Pesticides and Related Health Outcomes, Attachment 5 (cataloguing studies indicating that children are less able to protect themselves from organophosphate poisoning because they have yet to fully develop the "PON1 enzyme," which is necessary to detoxify these chemicals.).

[11] NAS Report at 309, Attachment 1.

associated with pesticide exposures, NAS recommended that "exposure from all sources—not just ingestion—must be considered when estimating total [pesticide] exposure and risk to children."[12]

For decades, EPA has required pesticide labels to include general admonitions to avoid spray drift, but has recognized that this generalized label direction is inadequate to protect innocent bystanders such as children from pesticide drift. For example, the Worker Protection Standard ("WPS") regulations contain a provision generally requiring pesticide users to "assure that no pesticide is applied so as to contact, either directly or through drift, any worker or other person . . . ."[13] However, even with such general label directions, EPA found that numerous poisoning incidents were occurring each year and the current drift labeling was "inconsistent or inadequate and for many products unclear to applicators and others."[14]

In order to provide better protections from drift, EPA took two actions. First, it established a "Spray Drift Task Force" ("SDTF") charged with helping to develop "a generic spray drift database which is expected to be capable of satisfying spray drift data requirements for virtually all pesticide product registrations in the United States and Canada."[15] The SDTF ultimately developed an evaluation tool—called "AgDRIFT"—that can help estimate exposure from spray drift for individual pesticides.[16] Second, EPA published a notice proposing "improved and more consistent product label statements for controlling pesticide drift in order to

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[12] NAS Report at 307, 308-09, Attachment 1.

[13] 40 C.F.R. § 170.210(a).

[14] EPA, Pesticide Registration (PR) Notice 2001-X Draft: Spray and Dust Drift Label Statements for Pesticide Products, Attachment 6 (hereinafter "Draft Spray Drift PR Notice").

[15] EPA, Pesticide Registration (PR) Notice 90-3: Announcing the Formation of an Industry-Wide Spray Drift Task Force (Apr. 6, 1990), Attachment 7.

[16] SDTF, AgDRIFT Frequency Asked Questions (July 31, 2003), Attachment 8.

be protective of human health and the environment."[17] EPA explained why it believed new spray drift label language was necessary:

> EPA's position on pesticide drift is that applicators must not allow pesticide spray or dust to drift from the application site and contact people, animals, and certain sensitive sites, including structures people occupy . . . , parks and recreation areas, nontarget crops, aquatic and wetland areas, woodlands, pastures, or rangelands. The Agency believes this is prudent public policy. It sets high but appropriate standards for applicators to protect people and the environment. . . . EPA believes the suggested labeling in this Notice will reduce risks associated with pesticide drift without a significant reduction in product efficacy. Accordingly, EPA believes that these label statements will help ensure that the requirements of FIFRA are met and, specifically, that pesticides are used in a manner that does not result in "unreasonable adverse effects on the environment."[18]

EPA's proposal would have placed limits on application equipment, methods, and conditions, such as wind speeds to reduce drift exposures.[19]

Despite recognizing that current pesticide labels were "inadequate" to prevent harmful drift, EPA never finalized the proposal to impose greater safeguards to prevent spray drift. Nor has EPA implemented the drift mitigation outlined in the proposal when it registered or reregistered pesticide uses.

C. Documented Evidence Confirms That Pesticide Drift Harms Children.

In 2001, EPA expressed concern about the number of poisoning incident reports and concluded that existing pesticide restrictions are insufficient to prevent harmful spray drift.[20] Poisoning incident reports continue to show that pesticide drift poses significant risks to people. For example, in 2006, the Washington State Pesticide Incident Reporting and Tracking Review Panel found that "[e]xposure to pesticide drift is an important cause of documented pesticide-

[17] Draft Spray Drift PR Notice, Attachment 6.

[18] Id.

[19] Id.

[20] Id.

related illness in Washington."[21] The California Department of Pesticide Regulation ("CDPR") documented 3,997 reported pesticide drift incidents in California between 1992 and 2007.[22] These reports are admittedly only the tip of the iceberg due to the well-documented disincentives and obstacles to such reporting.[23] In addition, a growing number of epidemiological studies link pesticide drift to specific adverse health effects in humans, including autism spectrum disorders,[24] Parkinson's disease,[25] and childhood acute lymphoblastic leukemia.[26]

Pesticide monitoring and modeling studies further confirm that pesticide drift may pose significant health risks to children who live near fields.[27] For example:

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[21] Washington State Pesticide Incident Reporting and Tracking Review Panel, Annual Report: 2005, at 81 (May 2007), Attachment 2; see also Barbara Morrissey, Washington State Department of Health, Spray Drift and Human Health Incidents, Attachment 9.

[22] Cal. Dep't of Pesticide Regulation, California Pesticide Illness Query, Attachment 10.

[23] Pesticide incidents are notoriously underreported. See General Accounting Office, Pesticides: Improvements Needed to Ensure the Safety of Farmworkers and Their Children (Mar. 2000), Attachment 11. EPA has recognized that pesticide incident reporting is of limited usefulness and questionable reliability due to the lack of any consistent national system for collecting such reports, the failure of health professionals and exposed persons to associate symptoms with pesticide exposure, lack of health insurance or financial resources to seek medical attention, and failure to record pesticide poisoning incidents in the various incident databases. EPA, Regulatory Impact Analysis of Worker Protection Standard for Agricultural Pesticides, at V-11 to V-20 (Aug. 1992), Attachment 12.

[24] E.g., Roberts, E., et al., Maternal Residence Near Agricultural Pesticide Applications and Autism Spectrum Disorders Among Children in the California Central Valley, Envtl. Health Perspectives, Vol. 115, No. 10, at 1482 (Oct. 2007), Attachment 13.

[25] E.g., Costello, S., et al., Parkinson's Disease and Residential Exposure to Maneb and Paraquat From Agricultural Applications in the Central Valley of California, Am. Journal of Epidemiology, Vol. 169, No. 8, at 919 (Jan. 2009), Attachment 14.

[26] E.g., Rull, R., et al., Residential Proximity to Agricultural Pesticide Applications and Childhood Acute Lymphoblastic Leukemia, Envtl. Research, Vol. 109, at 891 (July 2009), Attachment 15.

[27] See generally Tupper, K., Written Testimony of Karl Tupper, Staff Scientist, Pesticide Action Network North America for the Illinois Senate Agriculture and Conservation Committee, at 1-6 (Sept. 2009), Attachment 16.

- In 2007, an air monitoring study conducted near the Southwoods Elementary School in Hastings, Florida, detected four pesticides—endosulfan, diazinon, trifluralin, and chlorothalonil. At least one pesticide was found in each of the 39 samples, with three or four of the pesticides detected in 74% of samples, sometimes at levels exceeding levels of concern based on end points selected by the EPA as appropriate for assessing inhalation risk.[28] Exposure to these four chemicals is associated with a wide range of adverse health effects—endosulfan interferes with hormones and was linked to autism in an epidemiological study, diazinon is neurotoxic, and trifluralin and chlorothalonil are rated by the EPA as "possible" and "probable" carcinogens, respectively.

- In 2006 and 2007, air monitoring at homes and an elementary school in rural Minnesota also detected chlorothalonil—a fungicide EPA has classified as a "probable" carcinogen—in 123 of the 186 samples analyzed.[29]

- In Spring 2006, air monitoring in the Yakima Valley of Washington State, an area known for apple and grape production, detected chlorpyrifos—an acutely toxic organophosphate insecticide associated with developmental harm to children—in communities in amounts exceeding levels of concern derived from EPA selected endpoints and including EPA's FQPA safety factor.[30]

- Air monitoring in Lindsay, California, found chlorpyrifos in the air at levels exceeding the level of concern for children by up to 7.9 times in 2004, and up to 6.6 times in 2005.[31]

- In 2004, a study by scientists from the University of Washington and Washington State University on the organophosphate methamidophos determined that pesticide volatilization drift "could be a potentially high percentage of inhalation exposure" that "has implications in agricultural communities, where children are allowed to play outside immediately after spraying. . . ."[32]

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[28] Pesticide Action Network North America, Air Monitoring in Hastings, Florida: October 1–December 6, 2007 (Sept. 2008), Attachment 17.

[29] Pesticide Action Network North America, Pesticides and Air Pollution in Minnesota: The Frequency of Detection of Chlorothalonil, a Fungicide Used on Potatoes, at 11 Sites in 2006-07, Attachment 18.

[30] Farm Worker Pesticide Project & Pesticide Action Network North America, Poisons on the Wind: Community Air Monitoring for Chlorpyrifos in the Yakima Valley (Dec. 2006), Attachment 19.

[31] Pesticide Action Network North America, Air Monitoring for Chlorpyrifos in Lindsay, California (July 2006), Attachment 20.

[32] Ramaprasad, J., et al., The Washington Aerial Spray Drift Study: Assessment of Off-Target Organophosphorus Insecticide Atmospheric Movement by Plant Surface Volatilization, Atmospheric Environment 38 at 5703-13 (2004), Attachment 21.

- In 2000, chlorpyrifos was detected in one-third of all ambient air samples collected in California's San Joaquin Valley at levels that sometimes exceeded the level of concern for young children.[33]

- In 1996, the California Air Resources Board ("CARB") found chlorpyrifos in 74 percent of air samples taken at elementary schools and other sites near orange fields in Tulare County, California.[34] CARB has also detected potentially unsafe levels of other pesticides, including methidathion and molinate, in studies conducted between 1986 and 2000.[35]

These data indicate that pesticide drift is a potentially significant route of exposure for children who live or go to school near agricultural fields. In light of the vulnerabilities of children to pesticides, EPA cannot ensure that children will be protected from harm unless it accounts fully for such exposures.

II. EPA IS VIOLATING FEDERAL LAW BY FAILING TO PROTECT CHILDREN FROM PESTICIDE DRIFT.

A. Federal Law Requires EPA to Protect Children From Pesticide Drift.

1. Federal Law Governing Pesticides and Food

The Federal Food, Drug and Cosmetic Act ("FFDCA") regulates food safety and requires EPA to set "tolerances" (*i.e.*, maximum allowable levels) for pesticide residues in food.[36] A pesticide may not be used on a particular food unless EPA has established a tolerance or exemption for that food.[37] If a food contains pesticide residues that exceed the levels permitted

[33] Environmental Working Group, Every Breath You Take: Airborne Pesticides in the San Joaquin Valley (Jan. 2001), Attachment 22.

[34] CARB, Final Report for the 1996 Chlorpyrifos Monitoring in Tulare County (Apr. 13, 1998), Attachment 23.

[35] Lee, S., et al., Community Exposures to Airborne Agricultural Pesticides in California: Ranking of Inhalation Risks, Environmental Health Perspectives, vol. 110, no. 12, at 1175 (Dec. 2002), Attachment 24.

[36] 21 U.S.C. §§ 346a(b), (c).

[37] 21 U.S.C. § 346a(a)(1).

under a tolerance, the food is characterized as "adulterated" and is unlawful under the FFDCA.[38]

In 1996, Congress unanimously adopted the FQPA, which amended the FFDCA to incorporate NAS's 1993 recommendations for EPA to ensure that children are protected from pesticide exposures.[39] Under the FQPA, before EPA can allow a pesticide residue on a food, the agency must "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure" to the pesticide.[40] The FQPA defines "aggregate exposure" to include "all anticipated dietary exposures and all other exposures for which there is reliable information."[41] The FQPA gave EPA 10 years to bring all uses of pesticides on food into compliance with the new standards.

To implement these statutory mandates, EPA has developed a "risk cup" approach that first quantifies the exposure level for a pesticide that would exceed the safety standard for specific population groups, including fetuses, infants, and children in different age ranges. EPA then adds up exposures from various sources, such as consumption of each food on which the pesticide is used, residues in drinking water, and exposure to the pesticide through residential uses. If aggregate exposures to the pesticide from non-occupational sources "overflow" the risk cup for a particular subpopulation, the pesticide does not meet the FQPA safety standard. EPA will then look for ways to reduce exposure by, for example, eliminating some uses to reduce total exposure to levels that meet the safety standard.

Not surprisingly, given the FQPA's mandate to protect children, exposures to infants and

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[38] 21 U.S.C. § 342.

[39] H.R. Rep. No. 104-669, Pt. 2, at 43.

[40] 21 U.S.C. §§ 346a(b)(2)(C)(ii)(I), (II).

[41] 21 U.S.C. § 346a(b)(2)(A)(ii) (emphasis added); see also 21 U.S.C. § 346a(b)(2)(C)(vi) (In setting tolerances, EPA "shall consider . . . available information concerning the aggregate exposure levels of consumers . . . to the pesticide chemical and to other related substances, including dietary exposure . . . and exposure from other non-occupational sources . . . .").

children have often been the driving force behind pesticide cancellations and use limitations under the FQPA. For example, in 1997, EPA began a phase-out of almost all food uses of vinclozolin after finding that the pesticide posed unacceptable risks of sexual deformities in male fetuses.[42] In 2000 and 2001, EPA began a phase-out of almost all home and garden uses of the organophosphates chlorpyrifos and diazinon after determining that residential uses of these pesticides cause the child risk cup for each of these pesticides to overflow.[43] Most recently, in May 2009, EPA revoked all tolerances for carbofuran after determining that "estimated exposures significantly exceeded EPA's level of concern for children."[44] However, EPA has left children who are exposed to many of these same chemicals that drift from agricultural sites unprotected.

2. Federal Law Governing Pesticide Usage

Congress enacted the Federal Insecticide, Fungicide, and Rodenticide Act ("FIFRA") in 1947 to protect farmers from adulterated and ineffective pesticides. FIFRA had no health or environmental standards until 1972, when Rachel Carson's book Silent Spring and the controversy over DDT prompted Congress to amend FIFRA to incorporate health and environmental standards.[45]

Under FIFRA, EPA must register a pesticide before it can be sold or used in the United States.[46] To register or reregister a pesticide use, EPA must ensure that the chemical will

[42] EPA, R.E.D. Facts: Vinclozolin (Oct. 2000), Attachment 25.

[43] EPA, Occupational/Residential Handler and Postapplication Residential Risk Assessment for Chlorpyrifos, at 6 (Oct. 1999); Attachment 26; EPA, Diazinon Revised Risk Assessment and Agreement with Registrants, at 2-3 (Jan. 2001), Attachment 27.

[44] 74 Fed. Reg. 23,046, 23,052 (May 15, 2009).

[45] See Pub. L. No. 92-516, 86 Stat. 996 (1972); H.R. Rep. No. 511, 92d Cong., 1st Sess. (1971).

[46] 7 U.S.C. § 136a.

perform its intended function without causing any "unreasonable adverse effects on the environment."[47] FIFRA defines this standard as "any unreasonable risk to man or the environment, taking into account the economic, social, and environmental costs and benefits of the use of any pesticide . . . ."[48] In applying this standard, EPA must undertake a comprehensive assessment of all risks from a pesticide encompassing "every relevant factor that the Administrator can conceive into account," [49] including pesticide drift.

The FQPA amended FIFRA's "unreasonable adverse effects" definition to include "a human dietary risk from residues that result from a use of a pesticide in or on any food inconsistent with the [FQPA] standard."[50] Accordingly, EPA can register a pesticide only if there is reasonable certainty of no harm from aggregate exposure to the pesticide under the FQPA standard.[51] EPA can impose use restrictions as necessary to meet this standard, which are included on the legally enforceable pesticide label. The August 2006 deadline for bringing food-use pesticides into compliance with the FQPA extends to both tolerances under the FFDCA and registrations under FIFRA.[52]

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[47] 7 U.S.C. § 136a(c)(5)(C).

[48] 7 U.S.C. § 136(bb)(1).

[49] See S. Rep. No. 838, 92d Cong. 2d Sess., reprinted in 1972 U.S.C.C.A.N. 3993, 4032-33; see also EPA, General Principles for Performing Aggregate Exposure and Risk Assessments, at 9 (Nov. 28, 2001), Attachment 28 ("The FQPA-amended FIFRA also speaks to the requirement that [EPA] evaluate risks on an aggregate basis.").

[50] 7 U.S.C. § 136(bb)(2).

[51] E.g., EPA, General Principles for Performing Aggregate Exposure and Risk Assessments, at 9 (Nov. 28, 2001), Attachment 28 ("[T]he [FIFRA] standard for making decisions whether to register or continue registration of a pesticide for food-use must satisfy the standards in the FFDCA.").

[52] Use restrictions are set out on the EPA-approved label affixed to the product. A pesticide may not be used in a manner inconsistent with the label. If EPA determines that a pesticide registration does not comply with FIFRA, it may commence administrative proceedings to

3. Executive Orders on Environmental Justice and Child Health

Two executive orders issued during the 1990s also require EPA to protect children from pesticide drift. First, the 1994 Environmental Justice Executive Order requires EPA to ensure that its actions do not have disproportionate impacts on low-income and/or minority populations.[53] Specifically, EPA and other executive agencies must, to the maximum extent practicable, "identify[] and address[] . . . disproportionately high and adverse human health or environmental effects of its programs, policies, and activities on minority populations and low-income populations."[54] In furtherance of this mandate, EPA is required to "collect, maintain, and analyze information assessing and comparing environmental and human health risks borne by populations identified by race, national origin, or income" and "use this information to determine whether their programs, policies, and activities have disproportionately high and adverse human health or environmental effects on minority populations and low-income populations . . . ."[55]

Second, the 1997 Executive Order on Children's Health[56] requires EPA to protect children from environmental health and safety risks. Specifically, EPA is required to "ensure that its policies, programs, activities, and standards address disproportionate risks to children that result from environmental health or safety risks . . . that are attributable to products or substances that the child is likely to come in contact with or ingest (such as the air we breath [sic], the food we eat, the water we drink or use for recreation, the soil we live on, and the products we use or

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cancel the pesticide's registration or amend the registration to require additional safeguards. 7 U.S.C. § 136d(b).

[53] Exec. Order No. 12,898, 59 Fed. Reg. 7,629 (Feb. 11, 1994).

[54] Id. at § 1-101.

[55] Id. at § 3-302(a).

[56] Exec. Order No. 13,045, 62 Fed. Reg. 19,885 (Apr. 23, 1997).

are exposed to)."[57] Viewed together, these two executive orders require EPA, in making pesticide registration and tolerance decisions, to assess pesticide drift exposures along with all other pesticide exposures to ensure that pesticide exposures do not disproportionately impact children, low income populations, and/or minority populations.

B. EPA Has Violated Its Legal Duties by Ignoring Children's Exposures to Harmful Pesticides Through Drift.

EPA's record of compliance with its mandate to protect children from all pesticide exposures, including drift, is dismal. In the vast majority of cases, EPA has not even examined pesticide drift exposures, let alone imposed protections necessary to prevent harmful exposures to children.

As illustrated by its actions on chlorpyrifos, EPA has essentially applied a double standard, protecting urban children, but not protecting rural kids or suburban and ex-urban children that live or go to school near agricultural areas. Chlorpyrifos is a nerve poison that has been used in the United States since 1965 to kill insects in homes, schools, parks, and farms.[58] Chlorpyrifos is also associated with birth defects and impacts on human reproduction.[59] In 2000, EPA prohibited most home and residential uses of chlorpyrifos,[60] which the agency heralded as "particularly good news for children, who are among the most vulnerable to the risks posed by pesticides."[61] However, children are also exposed to chlorpyrifos particles that drift from target sites during application, and vapors that drift from the fields for days or even weeks after

[57] Id. at §§ 1-101(b), 2-202(b).

[58] EPA, Interim Reregistration Eligibility Decision for Chlorpyrifos, at 9-10 (Sept. 2001), Attachment 29 (hereinafter "Chlorpyrifos IRED").

[59] EPA, Human Health Risk Assessment: Chlorpyrifos, at 15-16 (June 2000), Attachment 30.

[60] EPA, Human Health Risk Assessment: Chlorpyrifos, at 3-10 (June 2000), Attachment 30.

[61] EPA, Administrator Carol M. Browner, Dursban Announcement, Remarks Prepared for Delivery June 8, 2000, Attachment 31.

application.[62] Unfortunately, EPA ignored and continues to disregard the harm to kids in or near agricultural communities when chlorpyrifos drifts from farms and contaminates the air at nearby schools, homes, parks, and daycare centers. Even as recently as 2006, EPA re-authorized use of chlorpyrifos on apples, citrus, cotton, corn, and many other crops without any protections to reduce drift exposures, despite considerable evidence (discussed above) that chlorpyrifos drifts from farms into nearby communities at alarming levels.[63]

EPA's failure to protect children from pesticide drift is not limited to chlorpyrifos. For example:

- Endosulfan is an organochlorine pesticide included on EPA's endocrine disruptor screening list and designated as a suspected endocrine disruptor by the Illinois EPA and the European Union.[64] Due to its toxicity, its ability to travel far distances after application, and its ability to bioaccumulate, endosulfan has been banned in over 60 countries around the world.[65] California Department of Pesticide Regulation ("CDPR") air monitoring has detected endosulfan drift in the air adjacent to agricultural sites at levels that pose toxic risks to bystanders.[66] Additional air monitoring data has detected endosulfan in the ambient air near schools and other locations where bystanders can be exposed.[67] Despite this evidence, EPA reregistered endosulfan in 2002 for dozens of food uses without considering drift exposures and their contribution to risk to children.

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[62] E.g., Farm Worker Pesticide Project & Pesticide Action Network North America, Poisons on the Wind: Community Air Monitoring for Chlorpyrifos in the Yakima Valley (Dec. 2006), Attachment 19; Pesticide Action Network North America, Air Monitoring for Chlorpyrifos in Lindsay, California (July 2006), Attachment 20.

[63] EPA, Organophosphorus Cumulative Risk Assessment - 2006 Update (Aug. 2006), Attachment 32.

[64] Inst. for Env't & Health, Chemicals Purported to be Endocrine Disruptors: A Compilation of Published Lists (Mar. 2005), Attachment 33.

[65] See PIC Circular 29 at 399, Attachment 34; see also Endosulfan Draft Risk Profile, prepared by the Persistent Organic Pollutants Review Committee of the Stockholm Convention (April 2009), Attachment 35.

[66] Cal. Dep't of Pesticide Regulation, Endosulfan Risk Characterization Document Volume I (August 2008), Attachment 36.

[67] E.g., Pesticide Action Network North America, Air Monitoring in Hastings, Florida: October 1–December 6, 2007 (Sept. 2008), Attachment 17.

- Both EPA and California have determined that oxydemeton-methyl ("ODM") is a developmental and reproductive toxicant.[68] ODM is an organophosphate registered for use in agriculture, primarily on Brussels sprouts, broccoli, and cauliflower, and lettuce.[69] EPA has acknowledged that exposure to ODM may cause reproductive effects such as reduced fertility, viability, ovarian and testicular weights, and increased estrous cycles.[70] The California DPR drift incident database reports 155 drift incidents associated with agricultural use of ODM between 1992 and 2007 in California alone.[71] Again, EPA reregistered ODM in 2006 without assessing or mitigating the risks posed by drift.

- Ethoprop is an organophosphate pesticide that EPA has classified as a likely human carcinogen.[72] EPA has acknowledged incidents in which ethoprop drifted following application and caused poisoning of children and other bystanders.[73] A 1998 study by the California Air Resources Board found concentrations of ethoprop in the air at application sites and in the ambient air at an elementary school approximately one-quarter mile from the nearest agricultural fields.[74] EPA ignored these exposures and risks when it reregistered ethoprop in 2006.

- The California Department of Pesticide Regulation ("CDPR") determined in 1999 that the organophosphate methyl parathion was a toxic air contaminant ("TAC") because the pesticide "may cause or contribute to increases in serious illness or death, or . . . may pose a present or potential hazard to human health."[75] Based on air monitoring data, CDPR estimated that the risk to infants from application site exposure to methyl parathion far exceed what EPA typically considers acceptable.[76] Despite this evidence, EPA ignored potential child drift exposures near application sites when it made registration decisions for methyl parathion in 2003 and 2006.[77]

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[68] EPA, Interim Reregistration Eligibility Decision for ODM, at 10-12 (Aug. 2002), Attachment 37 (hereinafter "ODM IRED"); Cal. EPA, Chemicals Known to the State to Cause Cancer or Reproductive Toxicity, at 14 (Sept. 2009), Attachment 38.

[69] ODM IRED at 5, Attachment 37.

[70] ODM IRED at 10, 46, Attachment 37.

[71] CDPR, California Pesticide Illness Query, Attachment 10.

[72] EPA, Interim Reregistration Eligibility Decision for Ethoprop, at 14 (Sept. 2001), Attachment 39 (hereinafter "Ethoprop IRED").

[73] Ethoprop IRED at 35, Attachment 39.

[74] Cal. Air Resources Bd., Final Report for the 1998 Ethoprop Air Monitoring (Dec. 1998), Attachment 40.

[75] CDPR, Toxic Air Contaminant Program, Attachment 41.

[76] CDPR, Evaluation of Methyl Parathion as a Toxic Air Contaminant: Executive Summary, at ix, Attachment 42.

[77] See EPA, Reregistration Eligibility Decision for Methyl Parathion, at 23-24 (May 2003), Attachment 43.

EPA's systematic failure to protect children from drift affects all children and violates the FQPA, which requires EPA to ensure that aggregate exposure to pesticides is safe for children,[78] and the 1997 Executive Order on Children's Health, which mandates that EPA protect children from disproportionate harm from pesticides and other environmental poisons.[79]

In addition, pesticide drift has disproportionate impacts on children from low income households. Farmworker families tend to be poor—on average, a farmworker family earned an annual income ranging from $15,000 to $17,499 in 2003.[80] In the top five agricultural counties in Texas (the state with the most acres of agriculture), between 10 to 30 percent of children live below the poverty line.[81] Likewise, in California (the top agricultural state by revenue), between 24 to 32 percent of children under the age of 17 live in poverty in the top three agricultural counties (compared with the state average poverty rate of 12.4%).[82]

Pesticide drift also has disproportionate impacts on children in minority populations. The

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[78] In light of EPA's systemic failure to protect children from pesticide drift and other flaws in EPA's pesticide risk-benefit assessment process, a broad coalition of farmworker and public health groups have attempted to compel EPA to more adequately address pesticide risks through several lawsuits that are now pending in the federal courts. See UFW v. Adm'r., EPA, No. C04-0099C (W.D. Wash.) (filed Jan. 13, 2004) (challenging reregistrations for AZM and phosmet); UFW v. Adm'r, EPA, No. C07-3950 JF (N.D. Cal.) (filed July 30, 2007) (challenging reregistrations for chlorpyrifos); PANNA v. EPA, No. C08-1814 MHP (N.D. Cal.) (filed Apr. 4, 2008) (challenging reregistrations for methidathion, oxydemeton-methyl ("ODM"), methamidophos, and ethoprop); PANNA v. EPA, 08-3542 MPH (N.D. Cal.) (filed July 24, 2008) (challenging reregistrations for endosulfan); UFW v. EPA, No. C08-3595 MHP (N.D. Cal.) (filed July 28, 2008) (challenging reregistrations for diazinon).

[79] Exec. Order No. 13,045, 62 Fed. Reg. 19,885 (Apr. 23, 1997).

[80] National Center for Farmworker Health, Migrant and Seasonal Farmworker Demographics (2009), Attachment 44.

[81] United States Department of Agriculture, 2007 County-Level Poverty Rates for TX (Dec. 2008), Attachment 45.

[82] Alice Larson, Migrant and Seasonal Farmworker Enumeration Profiles Study: California (Sept. 2000), Attachment 46.

vast majority of U.S. farmworkers are of Latin American origin—approximately 83 percent of U.S. farmworkers are of Latin American ancestry.[83] A majority of these farmworkers have children,[84] and these children live and go to school near the agricultural sites where their parents work. For example, in California over 73 percent of children attending schools within 1.5 miles of sites where at least 10,000 pounds of pesticides were applied in 1998 were non-white.[85] Similarly, in 2008 approximately 53 percent of students in Washington State's top five agricultural counties were non-white (the statewide average was 31 percent).[86]

The Environmental Justice Executive Order requires EPA to address disproportionate impacts of pesticide use on minority and low income populations, and the Child Health Executive Order requires EPA to address risks to children from pesticides. Contrary to these obligations, EPA has ignored a pesticide exposure pathway that directly and pervasively affects low income minority children who live near the fields. Indeed, for certain pesticides, such as chlorpyrifos and diazinon, EPA maintains a double-standard by protecting children from urban and residential uses, but ignoring exposures to children who live, play, and go to school near fields. These failures not only violate EPA's statutory obligations, they also violate EPA's obligations to address disproportionate impacts to children, minority, and low-income populations when it authorizes pesticide uses.

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[83] United States Department of Labor, The National Agricultural Workers Survey (Oct. 2006), Attachment 47.

[84] Id.

[85] Environmental Working Group, Every Breath You Take: Airborne Pesticides in the San Joaquin Valley (Jan. 2001), Attachment 22.

[86] School Data Direct, District-by-District Query, available at http://www.schooldatadirect.org/ (select "District" in the brown search box at the top of the screen, enter the district "Name" and "State" in the respective boxes. then click on the hyperlink for the district) (last viewed September 24, 2009).

ACTIONS NEEDED TO CORRECT EPA'S LEGAL VIOLATIONS

By maintaining registrations for hundreds of pesticides without accounting for and protecting children from pesticide drift, EPA is in violation of federal pesticide laws and executive orders directing it to protect minority and low-income populations as well as children from adverse health effects. To remedy these ongoing violations of law, this petition asks EPA to: (1) expeditiously evaluate the exposure of children to pesticide drift and impose safeguards to ensure that children are protected from aggregate pesticide exposures, including drift; and (2) immediately adopt interim controls that prohibit use of toxic drift-prone pesticides near homes, schools, parks, daycare centers, and other locations where children congregate.

I. EPA MUST CONDUCT PESTICIDE-SPECIFIC ASSESSMENTS OF THE RISKS TO CHILDREN FROM PESTICIDE DRIFT AND MODIFY PESTICIDE REGISTRATIONS TO ELIMINATE EXCESSIVE RISKS TO CHILDREN.

EPA cannot register a pesticide unless it has ensured that the chemical will perform its intended function without causing any "unreasonable adverse effects on the environment," which is defined to include violations of the FQPA safety standard.[87] By registering or reregistering pesticides without accounting for drift risks to children, EPA has overlooked a potentially significant route of exposure and has failed to fulfill its ongoing legal duty to protect all children from unsafe aggregate exposures to food-use pesticides. When a registered pesticide use poses unreasonable adverse effects or violates the FQPA safety standard, EPA must amend the registration or cancel offending uses.

To bring its pesticide registrations into compliance with these legal obligations, EPA must take two steps for all FFDCA-regulated pesticides. First, EPA must fully assess the exposure of children to drift from registered pesticide uses and determine whether such

[87] 7 U.S.C. § 136a(c)(5)(C).

exposures pose excessive risks. Such assessments must encompass applications of pesticides by ground sprayers, broadcast equipment, and aerial equipment, all of which have the potential to drift from application sites during and immediately after application.[88] In addition, depending on certain variables, such as the physical characteristics of the pesticide and meteorological conditions, pesticides can volatize for hours or days after application and drift as a vapor thousands of feet from application sites.[89] To address all pathways through which children may be over-exposed to pesticides, EPA's assessments must evaluate inhalation, oral, and dermal exposures to both spray drift and volatilization drift.

Second, based on the results of the drift exposure assessments, EPA must limit pesticide uses as necessary to protect children from drift. Such limitations would likely take the form of amendments to pesticide registrations, but might also lead to cancellations of uses that pose particularly high risks to children.

EPA has long recognized the need to assess drift exposures and incorporate necessary safeguards into pesticide registrations. For example, EPA's 2001 spray drift proposal indicated that the agency would conduct pesticide-by-pesticide reviews to determine "whether one or more no-spray zones and their distance(s) are necessary for products using available information about the pesticide's uses and risk assessments."[90] Likewise, in many of EPA's pesticide reregistration eligibility decisions, EPA specifically identified drift risks as a data gap needing further

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[88] See Draft Spray Drift PR Notice, Attachment 6.

[89] See, e.g., Ramaprasad, J., et al., The Washington Aerial Spray Drift Study: Assessment of Off-Target Organophosphorus Insecticide Atmospheric Movement by Plant Surface Volatilization, Atmospheric Environment 38, at 5703-13 (2004), Attachment 21.

[90] Draft Spray Drift PR Notice, Attachment 6 ("EPA in its risks management decisions will determine whether one or more no-spray zones and their distance(s) are necessary for products using available information about the pesticide's uses and risk assessments.").

assessment.[91] When EPA was called on to protect rural children as an FQPA subpopulation and revoke tolerances for several specific pesticides, EPA recognized the potentially significant risks associated with pesticide drift and acknowledged the need to further analyze and incorporate that risk into EPA's aggregate exposure assessments for drift-prone pesticides.[92]

EPA may be inclined to conduct these required drift assessments as part of FIFRA's pesticide registration review program, under which EPA plans to review pesticides originally registered before October 2007 by 2022.[93] While the pesticide registration review process should and indeed must assess drift exposures, it is not an appropriate vehicle for correcting the legal violations highlighted in this petition for two reasons. First, while EPA currently plans to initiate registration reviews for approximately 70 pesticides per year over the next four years, the agency has set no dates for completing those reviews, except to say that it "expects a total of about 710 pesticide cases comprising 1,136 pesticide active ingredients to undergo registration review by 2022."[94] Thirteen years is far too long to allow children to be exposed to pesticide drift without any EPA assessment of the risks posed to kids. Second, the registration review process is designed to address *new evidence* of risks and exposures that emerges after 2007. In

[91] E.g., EPA, Interim Reregistration Eligibility Decision for Methidathion, at 19-20 (Apr. 2002), Attachment 48 ("The Agency recognizes that there are many issues related to the use of agricultural chemicals in the general population, i.e., spray drift exposures and exposures to farm worker children and farm residents. The Agency is in the process of developing guidance and procedures for characterizing these kinds of risks.").

[92] Imidacloprid; Order Denying Objections to Issuance of Tolerance, 69 FR 30042, 30050 and 30054-55 (May 26, 2004); Order Denying Objections to Issuance of Tolerances, 70 FR 46706, 46730 (August 10, 2005). In these decisions, EPA chose not to identify farm children as a sensitive subpopulation under the FFDCA. This petition does not seek such a designation, but rather asks EPA to protect all children from the dangers of pesticide drift. Indeed, even in EPA's response to the petition, the agency agreed that pesticide drift exposure is a problem that needs to be addressed on a pesticide-specific basis.

[93] 7 U.S.C. § 136a(g)(1)(iii)-(iv).

[94] 74 Fed. Reg. 10,576 (Mar. 11, 2009).

other words, Congress assumed that all pesticide uses would be brought into compliance with the FQPA's protection mandates for children through the reregistration decisions completed by 2007. This premise is faulty for drift exposures since EPA ignored such exposures entirely in making its reregistration decisions.

Given this backdrop, EPA must implement an accelerated schedule for completing drift assessments and modifying registrations that prioritizes assessments based on the suspected degree of risk to children posed by the pesticide. EPA could undertake such accelerated reviews either through modifications to the registration review program or by utilizing other authorities. See, e.g., 7 U.S.C. § 136w(a)(1) (general rulemaking authority); 40 C.F.R. § 154.7 (special review).

II. EPA SHOULD IMMEDIATELY ADOPT INTERIM NO-SPRAY BUFFERS AROUND HOMES, SCHOOLS, DAYCARE CENTERS, AND PARKS TO PROTECT CHILDREN FROM DRIFT.

EPA should also adopt immediate interim measures to ensure that children are not harmed by pesticide drift while EPA completes the pesticide-specific drift assessments. Specifically, the agency should impose interim no-spray buffers around locations where children congregate, such as schools, homes, daycare centers, and parks, to prevent unassessed pesticide drift exposures to children. These measures should apply to organophosphates, n-methyl carbamates, and all other pesticides that are (1) registered for application by ground sprayers, broadcast equipment, and/or aerial equipment; and (2) suspected of causing acute poisonings, cancer, endocrine disruption, developmental effects, and/or reproductive effects.

A. EPA Should Take Immediate Action to Ensure that Children Are Protected From Pesticide Drift.

EPA has already determined that children may be exposed to many pesticides at or near levels that EPA considers unsafe without assessing drift risks. Indeed, according to EPA's own

assessments, children are possibly already at risk of being exposed to unsafe levels of two classes of pesticides—organophosphates and n-methyl carbamates.[95] These classes of pesticides are acutely toxic nerve poisons that are associated with other serious adverse health effects, including endocrine disruption, cancer, and developmental and reproductive effects. In EPA's 2006 cumulative risk assessment for organophosphates, the agency determined that the cumulative risk cup for organophosphates was overflowing for children aged 3 to 5 nationally, and also for children aged 1 to 5 in southern Florida.[96] EPA similarly found in 2007 that the risk from n-methyl carbamates overflowed the cumulative risk cup for children aged 1 to 5 nationally.[97]

EPA allowed children to continue to be exposed to organophosphates and n-methyl carbamates at levels exceeding its regulatory thresholds by asserting that its cumulative risk assessments may have overstated the risks.[98] This justification is undercut by EPA's failure to account for drift exposures in either the organophosphate or the n-methyl carbamate cumulative risk assessment. By leaving out a potentially significant route of exposure from its cumulative

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[95] EPA made these findings pursuant to the FFDCA requirement that EPA assess risks of cumulative exposure to pesticides that share a "common mechanism of toxicity." See 21 U.S.C. § 346a(b)(2)(C)-(D). In these cumulative risk assessments, EPA considers pesticide "exposures from food, drinking water, and residential sources" to "approximate as closely as possible people's actual exposures and potential risks resulting from current uses of these pesticides in different parts of the country." EPA, Assessing Pesticide Cumulative Risk (June 2008), Attachment 49. EPA also makes risk cup findings for individual pesticides and, in some cases, has cancelled pesticide uses to ensure that exposure to those individual pesticides conforms with the FQPA safety standard and "fit" within their individual pesticide risk cups. E.g., EPA, Chlorpyrifos Facts (Feb. 2002), Attachment 50; EPA, Carbaryl IRED Facts (Oct. 2004), Attachment 51.

[96] EPA, Organophosphorus Cumulative Risk Assessment: 2006 Update, at 13 (Aug. 2006), Attachment 32 (hereinafter "OP Cumulative Risk Assessment").

[97] EPA, Revised *N*-Methyl Carbamate Cumulative Risk Assessment, at 225 (Sept. 2007), Attachment 52 (hereinafter "NMC Cumulative Risk Assessment").

[98] OP Cumulative Risk Assessment at 15, Attachment 32; NMC Cumulative Risk Assessment at 225 n.22, Attachment 52.

assessment, EPA has not ensured that cumulative exposures to organophosphates and n-methyl carbamates comply with the FQPA safety standard. The omission of drift exposures from these cumulative assessments is particularly troublesome because many organophosphate and n-methyl carbamate pesticides are prone to drift and have been implicated in reported drift poisoning incidents. Indeed, one of the most commonly used organophosphates, chlorpyrifos, has been detected in several air monitoring studies, sometimes at possibly unsafe levels.

By failing to account for drift exposures in its organophosphate and n-methyl carbamate cumulative risk assessments, EPA has potentially understated children's exposure to these pesticides. According to EPA's own analysis, the cumulative risks from organophosphates and n-methyl carbamates is at or is even in excess of regulatory thresholds for some groups of children without accounting for the drift exposures. There is therefore no room left in the risk cup for additional exposures to children from drift.

B. EPA Should Impose Interim No-Application Buffer Zones to Protect Children From Exposure to Dangerous Drift-Prone Pesticides.

Petitioners ask EPA to immediately impose no-application buffer zones (designated areas around critical sites where pesticide applications are prohibited) around schools, homes, daycare centers, and parks to prevent drift exposures of children to toxic pesticides. EPA has recognized that such buffer zones can effectively reduce risks associated with pesticide drift. For example, EPA's own spray drift modeling indicates that spray drift concentrations are highest "adjacent to the treated area" and that levels "decrease with increasing distance from the treated area."[99] Likewise, in its 2001 spray drift proposal, EPA found that no application buffer zones effectively reduce harmful drift exposures.[100]

[99] 69 Fed. Reg. at 30,055.

[100] Draft Spray Drift PR Notice, Attachment 6.

Buffer zones are a workable and efficient drift mitigation strategy.[101] Indeed, in the few limited instances in which the agency has considered human exposures to pesticide drift, EPA has required no-spray buffer zones were necessary to reduce drift exposures. For example, EPA recently reregistered several soil fumigants (chloropicrin, dazomet, metam sodium/potassium, methyl bromide, and iodomethane), which are easily transported in the wind because the pesticides have an extremely high vapor pressure and are applied as a gas to soil before planting or for structural pest control.[102] Due to their acute toxicity and propensity to drift and volatilize, soil fumigants "have a well-documented history of causing large-scale human exposure incidents up to several thousand feet from treated fields."[103] One such mass poisoning incident occurred in 2003, when the soil fumigant chloropicrin drifted from an application site into homes and a daycare center that were approximately one-quarter mile away, poisoning over 165 people (including 62 people under the age of 14).[104] To reduce exposure to these acutely toxic fumigants, EPA prescribed buffer zones around application sites ranging from 25 feet to one-half mile.[105]

Another example is azinphos-methyl ("AZM"), an acutely toxic organophosphate pesticide. In 2001, without considering drift exposures, EPA found that AZM is so dangerous and causes so many poisonings of workers that it was not eligible for reregistration under

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[101] Declaration of D. Ken Giles, Ph.D., at 10-11 (Nov. 22, 2002), Attachment 53.

[102] See 74 Fed. Reg. 26,690 (June 3, 2009); EPA, Extension of Conditional Registration of Iodomethane (Methyl Iodide), Attachment 54; EPA, Amended Reregistration Eligibility Decision for Methyl Bromide, at 18-19 (May 27, 2009), Attachment 55.

[103] EPA, Amended Reregistration Eligibility Decision for Methyl Bromide, at 18-19 (May 27, 2009), Attachment 55.

[104] Center for Disease Control, Brief Report: Illness Associated with Drift of Chloropicrin Soil Fumigant into a Residential Area (Aug. 20, 2004), Attachment 56.

[105] EPA, Buffer Zone Fact Sheet (May 27, 2009), Attachment 57.

FIFRA's unreasonable adverse effects standard.[106] EPA cancelled dozens of AZM uses and allowed other uses to continue for an additional four years to allow growers to shift to alternatives.[107] At the end of this transition period, EPA again found all AZM uses were ineligible for reregistration due to the nerve poisoning risks to workers. However, EPA allowed some AZM uses to continue during a six-year phase-out (ending in 2012) with additional mitigation required to reduce risks during the phase-out period.[108] In identifying appropriate mitigation, EPA used the AgDRIFT model to conduct a cursory examination of the efficacy of buffer zones and ultimately imposed 60-foot no-use buffers around houses and other occupied dwellings for all uses of AZM.[109] Although this assessment was limited to dermal and oral ingestion exposures (it did not consider inhalation exposures), and indicated that buffers larger than the 60-foot buffer ultimately imposed were required,[110] EPA's decision to impose the 60-foot buffer is an acknowledgment that no-spray buffers around sensitive sites can protect children and other bystanders from pesticide drift exposures.[111]

Other federal agencies have similarly found that no-application buffer zones are an effective method for minimizing pesticide drift. For example, the National Marine Fisheries Service ("NMFS") recently issued two biological opinions concluding that three organophosphate pesticides (chlorpyrifos, diazinon, and malathion), and three carbamate

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[106] EPA, Interim Reregistration Eligibility Decision for Azinphos-Methyl, at 55 (Oct. 2001), Attachment 58.

[107] Id.

[108] EPA, Final Decisions for the Remaining Uses of Azinphos-methyl, at 1 (Nov. 2006), Attachment 59.

[109] EPA, Final Decisions for the Remaining Uses of Azinphos-methyl, at 1 (Nov. 16, 2006), Attachment 59.

[110] EPA, Determination of Buffer Zones for AZM Applications, at 2 (Oct. 2006), Attachment 60.

[111] EPA, Final Decisions for the Remaining Uses of Azinphos-methyl, at 8 (Nov. 16, 2006), Attachment 59.

pesticides (carbaryl, carbofuran, and methomyl) are jeopardizing the survival and recovery of endangered and threatened salmon and steelhead populations in Washington, Oregon, and California.[112] To reduce the movement of these pesticides from application sites into salmon-bearing waters, NMFS prescribed no-application buffers ranging from 50 to 1,000 feet.[113] NMFS has also issued several biological opinions prescribing buffers from salmon and steelhead habitat for aerial, ground spraying, and broadcast spraying of herbicides in connection with Bureau of Land Management and Forest Service noxious weed control programs.[114]

In addition, pursuant to the Endangered Species Act, federal courts have ordered EPA to impose buffer zones around wildlife habitat while the agency develops and implements pesticide-specific mitigation measures.[115] The first such case involved threatened and endangered salmon and steelhead. Based on evidence demonstrating the efficacy of no-spray buffers to lessen the migration of pesticides into rivers and streams, and scientific evidence and past practice, a federal district court directed EPA to impose no-spray buffers of 60 feet for

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[112] NMFS, Biological Opinion: Environmental Protection Agency Registration of Pesticides Containing Chlorpyrifos, Diazinon, and Malathion (Nov. 2008), Attachment 61 (hereinafter "OP Biological Opinion"); NMFS, Biological Opinion: Environmental Protection Agency Registration of Pesticides Containing Carbaryl, Carbofuran, and Methomyl (Apr. 2009), Attachment 62 (hereinafter "NMC Biological Opinion").

[113] OP Biological Opinion at 393-96, Attachment 61; NMC Biological Opinion at 489-91, Attachment 62. EPA recently responded to the first of these decisions with a plan to implement buffers ranging from 100 to 1,000 feet from salmon habitat for chlorpyrifos, diazinon, and malathion. EPA, Response to NMFS November 18, 2008, Final Biological Opinion (Sept. 10, 2009), Attachment 63.

[114] E.g., NMFS, Nez Perce National Forest Noxious Weeds Programmatic Biological Opinion (Jun. 2009), Attachment 64; NMFS, Reinitiation of Bureau of Land Management's 2002 Noxious Weeds Programmatic Biological Opinion (Jul. 2007), Attachment 65; NMFS, Vale District Noxious Weed Control Program, FY2003-2013, Union, Wallowa, Grant, and Umatilla Counties Biological Opinion (May 2003), Attachment 66.

[115] See Wash. Toxics Coal. v. EPA, No. C01-0132 (W.D. Wash. Jan. 22, 2004) (order), Attachment 67; Ctr. for Biological Diversity v. Johnson, No. 02-1580-JSW (N.D. Cal. Oct. 20, 2006) (order), Attachment 68; see also Ctr. for Biological Diversity v. EPA, No. 07-2794-JCS (N.D. Cal. June 30, 2009) (Stipulated Injunction and Proposed Order), Attachment 69.

ground applications and 300 feet for aerial applications.[116] The Ninth Circuit upheld this injunction imposing interim protection while EPA brings its pesticide registrations into compliance with legal requirements.[117] Ultimately, these interim buffers proved to be too small to prevent harm to the salmon and steelhead and to comply with the law for the organophosphate and carbamate pesticides that have since undergone full review under the ESA.[118]

Some state and local jurisdictions have also adopted buffer zones to protect children and other populations from pesticides. For example, in North Carolina, pesticide applications are prohibited within 100 feet of residences, and aerial applications are prohibited within 300 feet of schools, hospitals, nursing homes, churches, and businesses.[119] New Jersey has also prohibited aerial application of pesticides within 100 feet of certain residences and 300 feet of schools, hospitals, nursing homes, churches, and other buildings.[120] And numerous counties in California have adopted no-spray buffers of various sizes around homes and schools.[121]

Petitioners ask EPA to impose similar no-spray buffer zones for toxic drift-prone pesticides around places where children congregate such as schools, homes, daycare centers, and parks. The interim buffer zone should be at least 60 feet from these sensitive sites for ground spraying (including spraying with ground boom and air-blast equipment).[122] For aerial

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[116] Wash. Toxics Coal. v. EPA, No. C01-0132 (W.D. Wash. Jan. 22, 2004) (order), Attachment 67.

[117] Wash. Toxics Coal. v. EPA, 413 F.3d 1024, 1035 (9th Cir. 2005).

[118] OP Biological Opinion at 393-96, Attachment 61; NMC Biological Opinion at 489-91, Attachment 62.

[119] N.C. Admin. Code tit. 2, r. 9L.1005(b), (e).

[120] N.J. Admin. Code tit. 7, § 30-10.6(q), (s).

[121] See PANNA, Secondhand Pesticides: Airborne Pesticide Drift in California, at 45-46 (2003), Attachment 70.

[122] See Declaration of D. Ken Giles, Ph.D., at 9 (Nov. 22, 2002), Attachment 53.

applications, EPA should impose a 300-foot horizontal no spray buffer around these sensitive sites.[123]

These 60-foot and 300-foot buffers will reduce spray drift risks considerably,[124] but they will likely be insufficient to fully protect children from drift. For example, in EPA assessing the required 60-foot buffer zones for AZM, the agency recognized that larger buffers may be needed to ensure that children were not exposed to ground applications of AZM at levels exceeding what EPA typically considers acceptable, particularly from air-blast applications (although EPA ultimately declined to implement the larger buffers).[125] Many drift-prone pesticides are more toxic than AZM and are applied at higher rates than those authorized for AZM, indicating that buffers even larger than those needed for AZM may be necessary. In addition, the interim buffers sought in this petition are designed to protect from spray drift only; they do not address the volatilization drift exposures. It is therefore critical that, in addition to imposing the 60- and 300-foot interim buffers, EPA expeditiously complete pesticide-specific drift evaluations that assess both spray and volatilization drift exposures and determine whether larger buffers, and other mitigation are necessary to protect children from pesticide drift.

EPA has various mechanisms at its disposal to instate no-spray buffers that reduce drift exposures to children. For example, EPA has long resorted to the issuance of notices (called "pesticide registration notices" or "PR notices") to inform registrants of label amendments that are necessary to ensure compliance with FIFRA and to avoid cancellation or misbranding proceedings. EPA described this process when it initiated the Label Improvement Program,

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[123] Id.

[124] Id.

[125] EPA, Determination of Buffer Zones for AZM Applications, at 1-3 (Oct. 2006), Attachment 60.

which was "designed to upgrade pesticide labels in certain areas that contribute to the protection of health and environmental safety" but "are not adequately addressed in present labeling, and cannot await the development of registration standards."[126] EPA has utilized this PR notice process to protect the public and the environment from pesticides. For example, EPA issued PR notices limiting pesticide uses that pose dangers to farmworkers,[127] and restricting rodenticide and termiticide uses and formulations that can harm children, pets, and wildlife.[128] EPA should exercise that same authority to ensure that children are protected from pesticide drift.

Alternatively, EPA could impose the no-spray buffers under its broad authority "to prescribe regulations to carry out the provisions of" the statute.[129] For example, under this authority, EPA adopted the "Worker Protection Standard" ("WPS"), a suite of generally applicable regulations designed to reduce illness and injury to workers and their families, including through measures to lessen drift.[130] The requirements imposed through the WPS regulations must be incorporated into FIFRA pesticide labels, which makes them enforceable under FIFRA.[131]

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[126] 45 Fed. Reg. 37,884, 37,884 (June 5, 1980).

[127] EPA, Pesticide Registration (PR) Notice 83-2: Pesticide Label Improvement Program for Farmworker Safety (Mar. 1983); see also EPA, Pesticide Registration (PR) Notice 95-5: Labeling Revisions Required by the Worker Protection Standard for Sale or Distribution of Certain Agricultural Pesticides After October 23, 1995 (Sept. 1995), Attachment 71.

[128] EPA, Pesticide Registration (PR) Notice 94-7: Label Improvement Program for the Revision of Use Directions for Commensal Rodenticides and Statement of the Agency's Policies on the Use of Rodenticide Bait Stations (Sept. 1994), Attachment 72; EPA, Pesticide Registration (PR) Notice 96-7: Termiticide Labeling (Oct. 1996), Attachment 73.

[129] See 7 U.S.C. § 136w(a)(1).

[130] 40 C.F.R. §§ 170.1-170.250.

[131] 40 C.F.R. § 156.206(b); see also 40 C.F.R. § 170.210(a) (Requiring pesticide handlers and handler employers to "assure that no pesticide is applied so as to contact, either directly or through drift, any worker or other person, other than an appropriately trained and equipped handler.").

CONCLUSION

For these reasons, Petitioners petition EPA to take the following actions:

1) Conduct pesticide-specific drift assessments for all pesticides with the potential to drift and impose measures necessary to protect children from harmful drift exposures; and

2) For toxic drift-prone pesticides, including organophosphates and n-methyl carbamates, immediately adopt interim no-spray buffer zones of at least 60 feet for ground applications and 300 feet for aerial applications around areas where children may congregate such as homes, schools, parks, playfields, and daycare centers.

Please do not hesitate to call us if you have questions or would like to discuss the contents of this petition.

Respectfully submitted this 13th day of October, 2009.

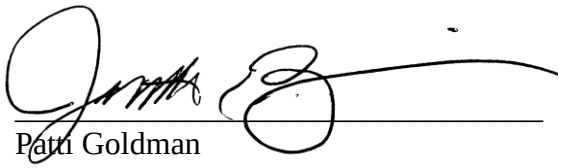

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EXHIBIT 6

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

MAR 3 1 2014

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

VIA EMAIL AND POST

To Addressees Listed in the Enclosure to this Letter

Subject: EPA Response to "Pesticides in the Air – Kids at Risk: Petition to EPA to Protect Children from Pesticide Drift"

Dear Petitioners:

Enclosed please find the Agency's response to your petition, "Pesticides in the Air – Kids at Risk: Petition to EPA to Protect Children from Pesticide Drift," submitted in October 2009 on behalf of a number of health and environmental organizations.[1] As you may recall, EPA posted the petition to the public docket (www.regulations.gov, docket ID EPA-HQ-OPP-2009-0825) in November 2009 and opened a comment period that ran 120 days in its entirety. In summary, and as related more specifically in our response, the petition asked EPA to account for children's exposures to pesticide drift and volatilization in its risk assessments and to take certain steps to reduce the risks from these exposures.

On July 24, 2013, you filed a writ of mandamus lawsuit against EPA alleging that EPA had unreasonably delayed responding to the petition. The parties agreed to stay the case as long as EPA promised to issue a final response to the Petition by March 31, 2014.[2] The enclosed response fulfills that promise.

During the several years that passed between the time the petition was submitted and the present, the Agency was actively developing drift and volatilization assessment methodologies, applying those methodologies to both fumigant and conventional pesticides, and finding ways to mitigate the risks to adults and children posed by pesticide drift and volatilization.

Recently the Agency posted to the docket and solicited comments on the methodologies it has developed for assessing the risks from pesticide drift and volatilization. The comment periods

[1] The organizations were Pesticide Action Network of North America (PANNA), United Farmworkers, Pineros y Campesinos Unidos Del Noroeste, MomsRising, Sea Mar Community Health Center, California Rural Legal Assistance Foundation, Farm Labor Organizing Committee, and Physicians for Social Responsibility.

[2] In re: Pesticide Action Network North America, et al. v. EPA, No. 13-72616 (9th Circuit)

for both methodologies are open at this time (at www.regulations.gov; the docket ID for the drift methodology is EPA-HQ-OPP-2013-0676; for the volatilization methodology, EPA-HQ-OPP-2014-0219).

The enclosed response discusses background on the petition, the pending lawsuit, the statutory and regulatory framework for EPA's pesticide programs, how these programs are implemented, and how EPA assesses and manages the risks associated with pesticide use, including risks from spray drift and volatilization. The response also directly addresses the three major changes to current practices that were requested by the petitioners, that the Agency 1) assess the potential risks posed by drift and volatilization for children in places where they live and play, 2) accelerate the assessment of these potential risks, and 3) adopt uniform buffers for pesticides of special concern in the interim while the risk assessments are being conducted.

EPA shares the concerns expressed by the petitioners, and agrees that the risks from drift and volatilization must be accounted for, for both children and adults, and that action must be taken to mitigate any such risks. The Agency believes that the registration review program already in place is a timely, efficient, and effective way to assess and take action on these risks, and does not believe that imposing requirements for uniform, interim buffers is scientifically supportable or defensible. Thus the Agency grants in part and denies in part the petitioners' requests.

We thank you for your interest and for your role in advancing awareness of the risks that pesticide drift and volatilization can pose to children. We hope that you will review and provide constructive comments on the newly published assessment methodologies for addressing those risks. Should you desire further information on the Agency's approach to assessing drift and volatilization, please contact Jill Bloom of my staff, at bloom.jill@epa.gov or (703) 308- 8019, and she will do her best to assist you.

Sincerely,

Martha Monell, Acting Director
Office of Pesticide Programs

Enclosures:
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Agency Response to "Pesticides in the Air – Kids at Risk: Petition to EPA to Protect Children from Pesticide Drift (2009)"

Table of Contents

I. Executive Summary

This document presents EPA's response to a petition that asked the agency to: 1) evaluate the risks to children exposed to pesticides through drift and volatilization, 2) establish a separate process or modify its pesticide re-evaluation process to expedite assessment and management of these risks, and 3) for certain types of pesticides, require the adoption of "one size fits all" buffer zones between treated areas and places where children congregate. EPA grants in part and denies in part this petition.

EPA agrees with Petitioners that individuals including children may be exposed to pesticides through drift and/or volatilization; that these exposures can occur in places where children live, go to school, play, or are otherwise present; and that, apart from occupational activities, children (depending on certain factors, including age) may experience higher levels of pesticide exposure relative to their size than do adults. Furthermore, the Agency agrees with the petitioners that it should conduct pesticide-specific assessments of the risks that include drift and volatilization exposures, as appropriate, and that if warranted, the Agency will take action to mitigate those risks. The steps the Agency has been taking to address these exposures are discussed later in this document.

Petitioners define the term drift to include "any airborne movement of pesticides away from a target site during and/or after application, including the airborne movement of pesticide droplets, pesticide powders, volatilized vapor-phase pesticides, and pesticide contaminated soil particles." In Sections VI through VIII of this response, EPA defines drift and volatilization as they relate to our risk assessments. The definitions differ mainly because the Agency draws a distinction between the off-site movement of spray droplets or pesticide particles during and shortly after the application process ("pesticide drift") and the movement of pesticide active ingredients as a vapor or gas that can occur for longer time after application is completed ("volatilization"). Both processes describe movement of pesticides through the air. The type of pesticide drift on which this response focuses is "spray" drift, the off-site movement of aerosols originating with pesticides applied as liquids, because spray drift is more likely to occur than the drift of pesticides in solid form, and when it does, it generally poses greater risk. EPA's efforts to assess and manage pesticide drift consequently are concentrated on spray drift.

EPA denies the Petitioners' request that EPA use a process outside of the ongoing pesticide re-evaluation process, as currently scheduled, to assess and manage spray drift and volatilization risks. The Petitioners suggest that the Agency should use alternative approaches that reprioritize pesticides for registration review or speed up risk assessments. The Agency believes that such adjustments to the registration review process are not needed and do not represent an efficient use of limited Agency resources.[1]

[1] If specific and significant concerns arise about an individual pesticide not in registration review at the time, the Agency can utilize other processes as appropriate to assess and mitigate risks (as needed). These processes are described later in this document and are not intended to replace the systematic and regular actions that constitute registration review. See IV.C.iii, which discusses regulatory options for cases in which the Agency has determined that a registered pesticide no longer satisfies the statutory standard.

EPA also denies the petition as it relates to Petitioners' request that EPA immediately adopt interim prohibitions on the use of certain pesticides that they allege are toxic and may be prone to drift or volatilization, near homes, schools, parks, and daycare centers or wherever children congregate. EPA instead believes that case-by-case, chemical-specific risk assessment is a sound, science-based approach, consistent with the Agency's mandate, that yields a more realistic representation of actual risks and facilitates the identification of what, if any, mitigation measures (potentially including no-spray buffers) are needed to protect potentially exposed individuals.

The response to the petition is organized in the following manner. After this executive summary, EPA follows with two sections (II and III) that discuss background on the petition, the pending lawsuit brought by the Petitioners, and the statutory and regulatory framework as it relates to EPA's implementation of its pesticide programs. The next two sections of the response (IV and V) outline the Agency's pesticide regulatory programs and how EPA assesses and manages the risks associated with pesticide use. The following sections (VI through VIII) revisit the Agency's terminology for describing the off-site movement of pesticides through the air via spray drift and volatilization and explain how the risks of each are assessed and managed. The next section (IX) contains EPA's response to the Petitioners' request for three changes to the Agency's current practices. The last section (X) provides EPA's conclusion.

II. Background

A. Petition History and Major Claims by Petitioners

In October 2009, a group of health and environmental organizations[2] ("Petitioners") jointly filed a petition entitled, "Pesticides in the Air- Kids at Risk: Petition to EPA to Protect Children from Pesticide Drift ("Petition"). The Petition alleged that EPA has failed to address children's exposures to and potential risks from pesticide drift and volatilization. More specifically the Petitioners ask EPA to:

1. fully and expeditiously evaluate the exposure of children to pesticide drift or vapors that originate from agricultural applications and travel to areas where children congregate, such as homes, parks, schools, and daycare centers. Furthermore, the Petitioners ask that the Agency act to ensure that children are protected from aggregate pesticide exposures, including exposures to drift;
2. implement an accelerated schedule (relative to the schedule for registration review) for completing drift assessments and modifying registrations that prioritizes assessments based on the suspected degree of risk posed by the pesticide drift and volatilization; and

[2] The organizations include Pesticide Action Network North America (PANNA), United Farm Workers, Pineros Y Campesinos Unidos Del Noroeste, Moms Rising, Sea Mar Community Health Center, California Rural Legal Assistance Foundation, Farm Labor Organizing Committee, and Physicians for Social Responsibility.

3. immediately adopt interim prohibitions on the use of toxic drift-prone pesticides such as organophosphates and n-methyl carbamates and certain other pesticides that are used near homes, schools, parks, and daycare centers or wherever children congregate.[3]

On November 4, 2009, EPA issued a Federal Register Notice requesting public comment on the assertions and requests made in the October 2009 petition. The comment period for the petition closed on January 4, 2010. EPA has reviewed the comments received.[4]

B. Lawsuit

On July 24, 2013, the Petitioners filed a writ of mandamus lawsuit against EPA alleging that EPA had unreasonably delayed responding to their 2009 Petition. EPA and the Petitioners agreed to stay the case. In EPA's unopposed motion to stay the case, the Agency promised to issue a final response to the Petition by March 31, 2014.[5] This response fulfills that promise.

III. Statutory and Regulatory Background/Framework

EPA regulates pesticides under the Federal Insecticide, Fungicide, and Rodenticide Act ("FIFRA"), 7 U.S.C. §§ 136-136y, and the Federal Food, Drug, and Cosmetic Act ("FFDCA"), 21 U.S.C. §346a. FIFRA sets forth a federal licensing scheme for the sale, distribution and use of pesticides; FFDCA establishes the mechanism and standards by which EPA must set tolerances (allowable levels) for pesticide residues in food. As a general matter, under FIFRA section 3, before a pesticide can be distributed or sold in the United States, it must be registered. Petitioners' administrative petition implicates both statutes.

A. The Federal Insecticide, Fungicide, and Rodenticide Act

The principal purpose of FIFRA is to regulate the sale, distribution and use of pesticides (through registrations) while protecting human health and the environment from unreasonable adverse effects associated with pesticides. *See generally* FIFRA section 3. Under FIFRA, EPA registers a pesticide only after conducting a scientific review of the risks, and when appropriate, benefits of that pesticide to determine whether the use of the pesticide causes "unreasonable adverse effects" to human health or the environment.[6] Registration and registration review decisions under section 3, reregistration decisions under section 4, and cancellation decisions under section 6 are governed by the same statutory standard, which generally is referred to as "risk-benefit" balancing, *i.e.*, a pesticide must not pose "any unreasonable risk to man or the

[3] The petitioners believe these interim measures also should apply to "all other pesticides that are (1) registered for application by ground sprayers, broadcast equipment, and/or aerial equipment; and (2) suspected of causing acute poisonings, cancer, endocrine disruption, developmental effects, and/or reproductive effects.

[4] The Petition docket is found at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2009-0825.

[5] In re: Pesticide Action Network North America, et al. v. EPA, No. 13-72616 (9th Circuit)

environment, taking into account the economic, social, and environmental costs and benefits of the use of any pesticide." FIFRA §§ 3(c)(5) & 2(bb); 7 U.S.C. §§ 136a(c)(5) & 136(bb). If this standard is not satisfied, EPA may not register the pesticide and existing pesticides are subject to modification or cancellation. *See* FIFRA §§ 3(c)(5) & 6(b); 7 U.S.C. §§ 136a(c)(5), & 136d(b).

In order to properly evaluate pesticide applications, FIFRA and its implementing regulations generally require applicants for registration to submit or cite to a significant body of toxicity and exposure data for the pesticides for which they are seeking registration. *See* 7 U.S.C. § 136a(c)(2)(A) (directing EPA to publish guidelines for submissions by applicants); 40 C.F.R. §§ 158.1 *et seq.*161.20 *et seq.* (setting forth information to be provided by applicants).

While EPA must consider a broad range of factors in determining whether a pesticide meets this standard, the balancing of the various risks and benefits of the pesticide, and consideration of inherent policy questions, is left largely to the discretion of EPA: "[W]ithin broad limits, the [A]dministrator has latitude not merely to find facts, but also to set policy in the public interest. Like most regulatory statutes,… FIFRA confers broad discretion on the [Administrator]." *Wellford v. Ruckelshaus*, 439 F.2d 598, 601 (D.C. Cir. 1971); *See also Envtl. Def. Fund v. EPA*, 465 F.2d 528, 538 (D.C. Cir. 1972) (FIFRA empowers EPA to "take account of benefits or their absence as affecting imminency of hazard").

As part of the process of EPA's approval of a pesticide registration, the agency must review and ultimately approve proposed labeling and directions for use for each pesticide. *See* FIFRA § 3(c)(5)(B). The approved pesticide label sets forth the lawful conditions of use for a pesticide, *i.e.*, those mandated by EPA in order to ensure that the pesticide will not cause unreasonable adverse effects to human health or the environment. *See Id.* § 3(d). Indeed, it is a violation of FIFRA for any person to use a pesticide in a manner inconsistent with the EPA-approved labeling. *See Id.* § 12(a)(2)(G).

B. The Federal Food, Drug, and Cosmetic Act

In 1996, the Food Quality Protection Act ("FQPA") was enacted, amending FFDCA and FIFRA to require all pesticides the use of which results in residues on food to meet new dietary risk standards. The FQPA amended the FIFRA risk-benefit standard ("any unreasonable risk to man or the environment") to add another element to the definition of that term: "a human dietary risk from residues that result from a use of a pesticide in or on any food inconsistent with the standard under [FFDCA section 408]." *See* FIFRA § 2(bb); FFDCA § 408(b)(1). In other words, the registration of a pesticide is contingent upon its meeting the food safety standard established under FFDCA section 408, if use of the pesticide results in residues on food. Section 408 also was amended by FQPA to add protections for infants and children and to establish the estrogenic substances screening program.[7]

[7] Public Law 104– 170, 110 Stat. 1489 (1996).

Section 408 of the FFDCA authorizes EPA to establish by regulation "tolerances" setting the maximum permissible levels of pesticide residues in foods.[8] FFDCA §§ 301(a), 408(a). Tolerance setting is discussed in more detail in Section IV.B.

EPA may only promulgate a pesticide tolerance, if the tolerance is "safe." FFDCA § 408(b)(2)(A)(i). "Safe" is defined by the FFDCA section 408 to mean that "there is a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue, including all anticipated dietary exposures and all other exposures for which there is reliable information." *Id.* § *408*(b)(2)(A)(ii) [emphasis added]. Section 408's reasonable certainty of no harm standard is a risk-only standard, which generally does not allow consideration of benefits.[9] Congress also amended the FFDCA to require that EPA re-assess, using the new safety standard, the existing tolerances and exemptions for all pesticide chemical residues that were in effect on August 3, 1996. *Id.* § 408(q)(1). Congress directed EPA to complete the reassessments by August 3, 2006.

Congress instructed EPA, when applying the new safety standard, to assess, among other things, the risks of pesticide chemicals based on available information concerning the special susceptibility of infants and children to pesticide chemical residues. FFDCA § *408*(b)(2)(C). To ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure to pesticide chemical residues, Congress mandated that EPA apply an additional ten-fold margin of safety (known as the FQPA safety factor) in setting tolerances unless reliable data show that a different margin of safety will be safe for infants and children. *Id*. The additional ten-fold margin of safety can be reduced or removed based on such a finding.

The FQPA amendments to the FFDCA also directed EPA to consider "aggregate exposure" in its decision-making. EPA has interpreted "aggregate exposure" to refer to the combined exposures-to a single chemical across multiple routes (oral, dermal, inhalation) and across multiple pathways (food, drinking water, residential). As amended by FQPA, section 408(b)(2)(ii) of FFDCA requires the Agency to make a finding for each tolerance or tolerance exemption "that there is a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue, including all anticipated dietary exposures and all other exposures for which there is reliable information"[emphasis added]. Section 408(b)(2)(C)(ii)(I) of FFDCA states that the Agency must find "there is a reasonable certainty that no harm will result to infants and children from aggregate exposure to the pesticide chemical residues." Finally, section 408(b)(2)(D)(vi) directs the Agency, when making tolerance decisions, to consider "aggregate exposure levels...to the pesticide chemical residue...including dietary exposure and exposure from other non-occupational sources."

EPA reaffirms its consistent interpretation of FFDCA section 408 as requiring consideration of all exposures to pesticide residues and other related substances other than those exposures

[8] Without such a tolerance or an exemption from the requirement of a tolerance, a food containing a pesticide residue is "adulterated" under section 402 of the FFDCA and may not be legally moved in interstate commerce.

[9] Benefits may only be considered under section 408 in one very narrow circumstance not applicable in this case.

occurring in the occupational setting.[10] Relevant exposures include pesticide residues in food and water and exposures to pesticides around the home or in public from sources other than food and water.

It is important to note that Congress has expressly provided that any issue that can be raised through the FFDCA review process can only be reviewed through that process.[11] Accordingly, to the extent a petition to revoke tolerances and cancel registrations raises issues relevant to the establishment or revocation of tolerances, EPA's response to those issues may be challenged only through the administrative and judicial review procedures provided in section 408 of the FFDCA and are not reviewable under FIFRA or any other provision of law.

C. Executive Orders Cited in Petition

EPA includes the following general discussion on two Executive Orders mentioned by Petitioners as support to their claims.[12]

i. Executive Order 12898: Federal Actions to Address Environmental Justice in Minority Populations and Low-Income Populations

On February 11, 1994, President Clinton issued Executive Order 12898 on Federal Actions to Address Environmental Justice in Minority Populations and Low-Income Populations.[13] This Order focuses federal attention on the environmental and human health conditions of minority and low-income communities and calls on agencies to make achieving environmental justice ("EJ") part of their mission. Since the issuance of that Executive Order, EPA has been actively working to ensure that EJ issues are considered in its decision-making processes. In September 2011, EPA issued its Plan EJ 2014 strategy.[14] The strategy is the Agency's roadmap for advancing environmental justice. Based on this strategy, the Agency seeks to:

- Protect the environment and health in overburdened communities.

[10] See Imidacloprid; Order Denying Objections to Issuance of Tolerance 69 Federal Register 30042, 30073 (May 26, 2004).

[11] FFDCA § 408(h)(5); NRDC v. Johnson, 461 F.3d 164, 176 (2d Cir. 2006).

[12] These Executive Orders do not create an independent right of action against the United States.

[13] http://www2.epa.gov/laws-regulations/summary-executive-order-12898-federal-actions-address-environmental-justice

[14] See http://www.epa.gov/compliance/ej/plan-ej/for more information. The plan is not a rule or regulation. It is a strategy to help integrate environmental justice into EPA's day-to-day activities. Plan EJ 2014 has three major sections: Cross-Agency Focus Areas, Tools Development Areas, and Program Initiatives. Within these areas, EPA plans to more effectively protect human health and the environment for overburdened populations by developing and implementing guidance on incorporating environmental justice into EPA's rulemaking process. EPA also plans to enable overburdened communities to have full and meaningful access to the permitting process and to develop permits that address environmental justice issues to the greatest extent practicable under existing environmental laws. Under the two statutes at issue here, FIFRA and the FFDCA, EPA has already begun to incorporate these considerations into its licensing program.

- Empower communities to take action to improve their health and environment.
- Establish partnerships with local, state, tribal, and federal governments and organizations to achieve healthy and sustainable communities.[15]

EPA is committed to addressing risks to population groups with unique exposure pathways, such as children, farm and migrant workers, urban poor populations, rural or isolated populations, and Native Americans and Alaskan Natives [emphasis added].[16] EPA's Office of Pesticide Programs has developed an internal training program for staff that provides an overview of environmental justice issues to be considered in risk assessment. Guidance materials and the templates for risk assessment documents direct risk assessors to address environmental justice concerns specifically as a basic element of pesticide risk assessment and the Agency risk management decision-making process includes consideration of any such concerns identified for the subject pesticide.

ii. Executive Order 13045: Protection of Children from Environmental Health Risks and Safety Risks

On April 21, 1997, President Clinton signed Executive Order 13045 on the Protection of Children from Environmental Health Risks and Safety Risks.[17] This Executive Order requires all federal agencies to assign a high priority to addressing health and safety risks to children, coordinate research priorities on children's health, and ensure that their standards take into account special risks to children. While not directly cited in the Petition, it should also be noted that on October 20, 1995, EPA adopted the Policy on Evaluating Health Risks to Children (predating the Executive Order). This policy requires the EPA to consider the risks to infants and children consistently and explicitly as part of risk assessments generated during its decision making process, including the setting of standards to protect public health and the environment.[18]

In response to Executive Order 13045, EPA established the Office of Children's Health Protection ("OCHP") in May 1997. The focus of this office is to make the protection of children's health a fundamental goal of public health and environmental protection in the United States. The Agency's Office of Pesticide Programs and OCHP are committed to ensuring that the Agency's risk assessments for pesticides are protective of children's health.[19]

IV. EPA's Pesticide Regulatory Programs

[15] Petitioners claim that EPA's pesticide assessments do not adequately address environmental justice issues as directed by the 1994 EJ Executive Order (Exec. Order No. 12,898, 59 Fed. Reg. 7,629 (Feb. 11, 1994)). EPA disagrees. EPA's commitment to EJ issues is clear from its recent pronouncements on these issues.

[16] http://ajph.aphapublications.org/doi/full/10.2105/AJPH.2011.300121

[17] http://yosemite.epa.gov/ochp/ochpweb.nsf/content/whatwe_executiv.htm

[18] http://yosemite.epa.gov/ochp/ochpweb.nsf/content/policy-eval_risks_children.htm

[19] http://gao.gov/products/GAO-13-254

The following sections discuss how EPA implements its statutory obligations under FIFRA and the FFDCA.

A. EPA's Review of Pesticide Registration Applications

EPA provides an application kit[20] to assist in preparation of an application to register a pesticide. The applicant is responsible for submitting or citing all of the information and data that are required to support the registration, including proposed product labeling, and scientific data that meet the data requirements related to the specific product the applicant intends to register. In addition, the applicant provides a Confidential Statement of Formula that details the composition of the pesticide product, i.e., 1) all active ingredients, 2) all inert ingredients, 3) all impurities of toxicological significance associated with the active ingredient, and 4) all impurities found to be present at a level equal to or greater than 0.1 percent by weight of the technical grade active ingredient.[21]

The applicant will also provide EPA with draft labeling. FIFRA section 2(p)(1) defines "label" as "the written, printed or graphic material on, or attached to, the pesticide or device or any of its containers or wrappers." The term "labeling" is defined as "all labels and all other written, printed, or graphic matter –

(A) accompanying the pesticide or device at any time; or
(B) to which reference is made on the label or in literature accompanying the pesticide or device." *See* FIFRA § 2(p)(2)

Labeling includes detailed information such as the ingredients statement, warnings and precautionary statements, and directions for use. It is unlawful to sell or distribute a pesticide if any claims made for it differ from claims made on labeling required for registration. *See* FIFRA § 12(a)(1)(B). Therefore, advertising claims for a pesticide product must not contradict claims made in the product's labeling. Labeling requirements are codified in 40 CFR Part 156. EPA has developed a Label Review Manual[22] as guidance to its staff on reviewing labels.

Applicants are responsible for citing or generating all data to meet data requirements. The purpose of these data requirements is to demonstrate that the product will not cause unreasonable adverse effects on the environment. In general, these data are used to evaluate whether a pesticide has the potential to cause adverse effects on humans, non-target wildlife, and plants, as well as possible contamination of surface water or groundwater from leaching, run-off, and spray drift. For pesticides that will need a tolerance or tolerance exemption to demonstrate a reasonable certainty of no harm, additional data are needed.

Requirements may include, as applicable, data on:

- spray drift,
- residue chemistry,

[20] http://www.epa.gov/pesticides/registrationkit/

[21] http://www.gpo.gov/fdsys/pkg/CFR-2011-title40-vol24/pdf/CFR-2011-title40-vol24-sec158-320.pdf

[22] http://www.epa.gov/oppfead1/labeling/lrm/

- environmental fate,
- toxicology,
- applicator exposures
- reentry protection,
- wildlife and aquatic organisms,
- plant protection,
- nontarget insects,
- product performance, and
- product chemistry.

Data requirements in support of applications for registration of a pesticide product are specified in 40 CFR Part 158.[23]

EPA's review of the application includes the assessment of the risks to human health and the environment that may be posed by the pesticide.[24]

B. Tolerance Setting

i. Process overview

A tolerance is the maximum allowable concentration of a pesticide on a particular food item. The tolerance is the residue level that triggers enforcement actions. That is, if residues are found above that level, the commodity will be subject to seizure by the government. EPA must consider a number of factors when it establishes, modifies, leaves in effect, or revokes a tolerance for a pesticide chemical residue. FFDCA § 408(b)(2)(D). The process for the establishment, modification, or revocation of tolerances, is described directly below.

A tolerance action may be initiated by EPA of its own accord[25] or in response to an administrative petition. *Id.* § 408(d)(1), (e)(1). "Any person may file with [EPA] a petition proposing the issuance of a regulation . . . establishing, modifying, or revoking a tolerance for a pesticide chemical residue in or on a food." *Id.* § 408(d)(1)(A). If EPA determines that an administrative petition meets the statutory and regulatory requirements governing petition contents, EPA publishes a notice of the administrative petition's filing. *Id.* § 408(d)(3). (The Agency will also publish a notice when the action is Agency-initiated.)

After the publication of the notice, EPA must give "due consideration" to the petition and then: i) issue a final regulation establishing, modifying, or revoking a tolerance; ii) issue a proposed regulation under the separate provisions of the FFDCA § 408(e), and thereafter issue a

[23] Data requirements are described at http://www.gpo.gov/fdsys/pkg/CFR-2011-title40-vol24/pdf/CFR-2011-title40-vol24-part158.pdf; links to guidelines for the conduct of required studies are located at http://www.epa.gov/pesticides/science/guidelines.htm

[24] For further details on the pesticide registration see http://epa.gov/pesticides/factsheets/registration.htm.

[25] When, for example, the tolerance action follows a cancellation action.

final regulation after additional public notice and comment; or iii) issue an order denying the petition. FFDCA § 408(d)(4)(A). *See NRDC v. Johnson*, 461 F.3d at 173-74.

After EPA issues a regulation or order establishing, modifying, or revoking a tolerance for a pesticide chemical residue on food, any person may file objections with EPA and request an evidentiary hearing on those objections. FFDCA § 408(g)(2). After consideration of any such objections, EPA must issue a final order separately stating the action taken on each objection and whether any hearing is appropriate. *Id*. § 408(g)(2)(C). Then the Agency can conclude its deliberations and grant, modify, or deny the tolerance, as appropriate.

ii. The Tolerance Petition and Required Documentation

As discussed above, a determination on the proposed tolerance relies on the risk-only standard from FFDCA section 408. EPA must ensure that the use associated with the tolerance will not pose unreasonable risks to human health. Except in certain instances,[26] a tolerance petition request is usually accompanied by an application for registration, an application to amend the registration of a currently registered product, or an experimental use permit for the uses proposed in the petition. As risk assessments are a component of the standards for evaluating both tolerance proposals and registration actions, EPA determines whether any meaningful risks exist for the proposed uses, based on an evaluation of the applicant's petition for a tolerance. The Agency bases its tolerance decision on the aggregate exposures and risks associated with the pesticide and the use to which the petition applies. Aggregate exposure is the combined exposures to a single chemical across multiple routes (oral, dermal, inhalation) and across multiple pathways (food, drinking water, residential).[27]

iii. How the Proposed Tolerance Action is Assessed

The risks of concern that are considered in the setting of tolerances are the human health risks from aggregate exposures, which are the sum of exposures from each relevant pathway--food, drinking water, and/or residential. The assessment of human health risks is described in more detail in Section V. Aggregate risks are calculated based on varying durations of exposure. When no residential uses exist, aggregate risks are based on exposure contributions from food and drinking water for the acute and chronic durations. For residential-use pesticides, residential exposures are combined with food and drinking water exposures for each applicable duration of exposure. As discussed above, a determination on the proposed tolerance will be based on the risk-only standard from FFDCA section 408.

[26] A request for an import tolerance generally would not require an accompanying application for registration.

[27] Residential exposures include exposures associated with homes, home lawns, yards, gardens, apartments and grounds around apartment buildings, schools, schoolyards, daycare facilities, playgrounds, athletic fields, and parks and other public spaces.

In 1997, EPA issued "HED SOP 97.2 Interim Guidance for Conducting Aggregate Exposure and Risk Assessments (11/26/97)."[28] The Agency continued to work on more sophisticated methods for estimating aggregate exposures to pesticides, and in 2001, released "General Principles for Performing Aggregate Exposure and Risk Assessments,"[29] to augment the guidance document.

The aggregate risk assessment process relies on available data, assumptions designed to be protective of public health, standard analytical methods and Agency SOPs to estimate exposures to a pesticide for each potential pathway and route of exposure. EPA combines these separate exposure estimates to calculate potential aggregate exposure and risk; aggregate exposures estimated in this way reflect upper-bound or high-end of exposures for each route/pathway. The most highly exposed group, which generally also has the highest associated risk, is used as the basis for decision-making purposes. Aggregate risk assessments conducted in this manner typically can be refined by the use of additional exposure data and data specifically designed to address the uncertainties within an individual aggregate analysis, as well as more sophisticated analysis techniques.

The assumption implicit in this approach is that individuals can encounter the high-end exposures from the different pathways all at one time. In actuality, co-occurrence of high-end food, drinking water, and residential exposure scenarios is very unlikely. Thus, in using this approach, EPA is confident that aggregate exposure estimates will overstate, sometimes significantly, the amount of a pesticide to which people actually are exposed. The primary advantage to relying on these highly conservative assessments is that they require relatively fewer data and analytical resources and less time to conduct. In addition, an aggregate risk assessment of this type may be enough to indicate that a particular pesticide use satisfies the appropriate regulatory standards.

C. Pesticide Re-evaluation Processes

i. Reregistration

FIFRA requires the periodic re-evaluation of currently registered pesticides. In 1988, Congress amended FIFRA section 4 to include a specific process for the "reregistration" of pesticides containing active ingredients first registered before November 1, 1984. Pub. L. 100-532, title I, § 102(a), 102 Stat. 2655, 2683(1988). To make a Reregistration Eligibility Decision (RED), [30] EPA reviewed all the studies that were submitted by the pesticide registrants for a

[28] U.S. Environmental Protection Agency. 1997e. Memorandum from Margaret Stasikowski, Health Effects Division to Health Effects Division Staff. "HED SOP 97.2 Interim Guidance for Conducting Aggregate Exposure and Risk Assessments (11/26/97);" November 26, 1997. Office of Pesticide Programs, Office of Prevention, Pesticides, and Toxic Substances, Washington, D.C. Available upon request.

[29] http://www.epa.gov/pesticides/trac/science/aggregate.pdf

[30] In particular cases, the Agency issued a variation on the RED, i.e., a Tolerance Reassessment Decision, or, for individual active ingredients identified as belonging to a group of pesticides with a common mechanism of action, an Interim Reregistration Decision may have been issued before the RED.

pesticide active ingredient, as well as other relevant information, developed the appropriate risk assessments, and decided whether or not the pesticide active ingredient satisfied the risk-benefit standard of FIFRA section 3(c)(5). After determining eligibility of the active ingredient, EPA determined whether to reregister products containing the active ingredient.[31] *See* FIFRA § 4(g)(2)(B). In conjunction with most REDs, the Agency "called-in" active ingredient- or product-specific data considered necessary to reduce uncertainty in the RED risk assessments.

Reregistration was an open and transparent process. Before finalizing its decision on the eligibility for reregistration of a pesticide, EPA made the supporting risk assessments available for public comment, although it was not required to do so. Comments were solicited particularly on the factual basis of the risk assessments and also on options for mitigating the risks posed by the subject pesticide. These comments were considered in the Agency's decision-making.

Most of the pesticides specifically named in the Petition went through the reregistration process. Information on the reregistration status and links to the reregistration dockets for any pesticide can be accessed via the Agency's Chemsearch database.[32] And, as explained more fully below, they have also been scheduled for review early in the current re-evaluation process, known as Registration Review. *See* FIFRA § 3(g).

ii. Registration Review

Once the reregistration decisions for all the subject active ingredients were completed, the Agency began the next re-evaluation process under FIFRA, which requires EPA to regularly review pesticides to ensure that they continue to satisfy the statutory standard for registration. The ongoing re-evaluation process is called registration review. Section 3(g) of FIFRA requires EPA to complete its initial registration review cycle by October 1, 2022, for all pesticides registered prior to October 1, 2007, and by 15 years after the date of initial registration for pesticides registered after that date. *Id.*[33] Following the initial review of the pesticide, EPA must conduct subsequent reviews of each registered pesticide every 15 years thereafter.

The registration review program[34] makes sure that, as the ability to assess risk evolves and as policies and practices change, all registered pesticides continue to meet the statutory standard of no unreasonable adverse effects. Through registration review, the Agency is ensuring that registered pesticides do not cause unreasonable risks to human health or the environment when used as directed on product labeling. Changes in science, public policy, and pesticide use practices will occur over time, and the cyclical nature of registration review will enable the

[31] While the Agency has completed its statutory requirement to make eligibility determinations on the subject active ingredients, product reregistration is still ongoing for some reregistration cases.

[32] http://iaspub.epa.gov/apex/pesticides/f?p=chemicalsearch:1; search by active ingredient name, PC Code, or CAS number.

[33] See also 40 CFR Part 155 for the implementing regulations. An overview of the process is found at http://www.epa.gov/oppsrrd1/registration_review/highlights.htm.

[34] Unlike earlier re-evaluation programs, registration review operates continuously, and provides for the review of all registered pesticides.

Agency to consider updated information every time a pesticide comes up for registration review.[35]

In conducting the registration review program, EPA generally is reviewing pesticides in chronological order according to their baseline dates;[36] that is, older cases are being reviewed first. In addition, many pesticides that received priority scheduling for reregistration have been scheduled early in registration review. Thus, food-use chemicals[37] that were identified as or suspected of having risk concerns at the outset of reregistration generally are scheduled early in the registration review process. Additionally, within this structure, EPA plans to review certain related pesticides at the same time, particularly pesticides in the same family, with the same general structure and mode of action.

In reregistration, EPA gained experience and benefited from efficiencies in the concurrent review of pesticides in families like the organophosphates (OPs), N-methyl carbamates (NMCs), triazines, and chloroacetanilides. The rodenticides and soil fumigants were also reviewed concurrently. EPA plans to continue the practice of grouping related pesticides during registration review.[38] Potential efficiencies from this practice include:

- Technical and regulatory issues may be resolved more easily looking across an entire chemical class or group;
- Resources can be optimized within EPA, among stakeholders, and within other federal agencies; and
- In developing decisions, a "level playing field" among chemicals in the group may be assured so that EPA's actions do not inadvertently cause increased risks.

EPA completed cumulative risk assessments and reregistration risk management decisions for the OP pesticides in August 2006 and the NMC pesticides in September 2007. In recent years, EPA and stakeholders have invested significant resources in gaining a better understanding of these classes of pesticides. The registration review of the OPs began in 2008, and the N-methyl carbamate review began in 2010. The Petitioners are requesting expedited reviews for both classes of chemicals. These classes of pesticides were among the first pesticides to enter the registration review program, so the assessment of risks associated with their use (including risks to children, and risks from spray drift), and the management of those risks, should be accomplished early in the current registration review cycle, and in subsequent cycles as well. Volatilization will also be addressed based on the results of the screening analysis and the subsequent availability of pertinent data, should they be required for individual pesticides. The scheduling of these classes of pesticides in registration review reflects an understanding of the importance of addressing the toxicity, dietary and aggregate risks, and the

[35] The Agency uses the best, most recent information in any risk assessment, but the cyclical nature of registration review assures that assessments for any one pesticide will be updated at least every 15 years.

[36] The baseline date is the date when a RED was completed, or for a pesticide not subject to reregistration, the date when the pesticide was first registered.

[37] Pesticides used on food crops were given high priority because they have potential to affect the population at-large via dietary exposure.

[38] The overview at http://www.epa.gov/oppsrrd1/registration_review/highlights.htm provides links to information on the pesticide family groupings.

volume of use for these pesticides. *See* Appendix to this response for the registration review schedules for the OPs and NMCs.

EPA initiates a registration review by establishing a docket for a pesticide registration review case and opening the docket for public review and comment. The Agency publishes a Federal Register notice that announces the availability of a Preliminary Work Plan (PWP) and provides a comment period of at least 60 days. Anyone may submit data or information in response. EPA will consider information received during the comment period in conducting a pesticide's registration review. The PWP:

- explains what EPA knows about the pesticide from previous risk assessments including hazard and exposure information related to children, when available, and application of uncertainty factors including the FQPA safety factor;
- tentatively identifies what kind of risk assessments are needed;
- tentatively identifies what data will be needed to conduct the assessments;
- addresses the uncertainties in the database that will impact the risk assessments (particularly missing or unclear use parameters);
- provides basic use and usage information and other background information on the pesticide; and
- provides a proposed schedule that lays out milestones up until the registration review decision is made.

Information from registration review dockets that have opened is readily accessible via Chemical Search on a chemical-by-chemical basis.

Registration review, as set forth in the regulations, is a transparent process in which stakeholders of all types are invited to provide relevant data and comments. At the beginning of registration review, the public is asked to comment on the PWP and its supporting documents; by soliciting these comments, EPA aims to gather additional information that can enhance registration review planning and decision-making. Other registration review documents also are subject to a public comment period, such as when the preliminary risk assessments are posted to the docket later in the process. Similar approaches to public notification and comment were used during reregistration and can be used in decision-making about new active ingredients.

EPA may also, as needed, consult with registrants, the U.S. Department of Agriculture. and other stakeholders to resolve any uncertainties about how the product is used, particularly if use parameters are not clear from product labeling, or if the registrant may have data not already submitted to the Agency that could inform the process. For example, some labels are not specific about retreatment intervals or seasonal maximum application rates. Without actual use parameters, the Agency is forced to make conservative assumptions that can result in overly conservative risk assessment results. The Agency may solicit such information in a "Focus Meeting." These meetings can be held whenever in the registration process they might be helpful. To ensure transparency, materials associated with Focus Meetings are available in the pesticide-specific registration review dockets. If a focus meeting is held prior to the docket

opening, these materials are posted to separate docket.[39] Once the docket opens, a copy of the focus meeting notes will be posted to the case-specific docket.

After the close of the initial comment period, EPA revises its work plan as needed based on public input and any additional information that has become available in the interim. The Final Work Plan is then posted to the docket, and EPA prepares to call-in any data needed for the risk assessments. Once the registrants submit the required data, work on the risk assessments begins. As noted above, EPA makes the draft risk assessments available for public review and comment, and subsequent to review of public comments, the Agency posts the revised assessments. If the revised assessments indicate that there are risks of concern, EPA may invite the public to submit suggestions for mitigating those risks. These suggestions are used in the development of a proposed registration review decision.

EPA will announce the availability of a proposed registration review decision on the docket and will provide a public comment period of at least 60 days. The process culminates with a final registration review decision—EPA's determination on whether the pesticide in question meets or does not meet the standards for registration. The final registration review decision discusses any changes that are needed to the pesticide's registration or labels to address the risks of concern. If a registrant fails to take action to implement these changes, EPA may take appropriate action under FIFRA.

To meet its statutory obligations for registration review, EPA is opening 70 or more dockets annually continuing through 2017, and almost all of the pesticides registered at the start of the program will have dockets opened by 2017. The Agency is directed to complete the first round of registration reviews by October 1, 2022; during that time the Agency will complete the registration reviews of at least 744 pesticide cases comprising 1,165 active ingredients. Pesticides registered after 2007 will be folded in each year.[40]

iii. Regulatory Responses to Unacceptable Risks

If EPA determines that a pesticide product does not meet the statutory standard, the Agency may take "appropriate regulatory action." Such regulatory actions can include, but are not limited to, restricting pesticide uses or canceling the pesticide's registration. *See* FIFRA §§ 3(d) & 6(b). If EPA chooses to cancel a pesticide registration, EPA must first issue a Notice of Intent to Cancel and hold a formal administrative hearing, if one is requested by a person adversely affected by the notice. *See* FIFRA § 6(b).

If the Agency determines that an "imminent hazard" exists from the use of a pesticide (including a pesticide that was not eligible for reregistration), EPA may commence proceedings

[39] General information about focus meetings is at http://www.epa.gov/oppsrrd1/registration_review/focus-meetings.html; the docket itself is at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2012-0778.

[40] The status of all completed and ongoing registration reviews can be found at http://www.epa.gov/oppsrrd1/registration_review/reg_review_status.htm.

to suspend the registration of a pesticide during the time needed to complete cancellation proceedings. *See* FIFRA § 6(c). Section 2(l) of FIFRA defines imminent hazard as:

> [a] situation which exists when the continued use of a pesticide during the time required for cancellation proceeding would be likely to result in unreasonable adverse effects on the environment or will involve unreasonable hazard to the survival of a species declared endangered or threatened by the Secretary pursuant to the Endangered Species Act….

If the EPA determines that an emergency exists such that the imminent hazard will occur during the period necessary to complete normal suspension proceedings, the EPA may issue an immediately-effective emergency suspension order in advance of completing suspension proceedings. *Id*. § 6(c)(3).

Courts addressing the suspension provisions have held that an imminent hazard exists if there is "a substantial likelihood that serious harm will be experienced during the year or two required in any realistic projection of the administrative process." *Love v. Thomas*, 858 F.2d 1347, 1350 (9th Cir. 1988) (quoting *Environmental Defense Fund v. EPA*, 465 F.2d 528, 540 (D.C. Cir. 1972). In the case of an emergency suspension, one court has found by analogy that suspension is appropriate if there is a "substantial likelihood that serious harm will be experienced during the three or four months required in any realistic projection of the administrative suspension process." *Dow v. Blum*, 469 F.Supp. 892, 901 (E.D. Mich. 1979). Thus, courts interpreting the FIFRA suspension standard have made clear that an imminent hazard finding requires a greater degree of likelihood, immediacy, and severity of harm than is otherwise required to take a cancellation action under FIFRA. And in evaluating the nature and extent of information before EPA, the courts have instructed the Agency to consider (1) the seriousness of the threatened harm, (2) the immediacy of the threatened harm, (3) the probability that the threatened harm will occur, and (4) the benefits to the public of the continued use of the pesticide *Id*. at 902.

EPA's review and re-evaluation processes for pesticides have been developed to account for advancements in science so that risks can be identified and managed before a pesticide is registered and at regular intervals thereafter. Through these processes, the Agency can anticipate and correct problems with the use of pesticides as time goes on, and, ideally, reduce the chances that a pesticide would pose risks of an immediacy and magnitude like those associated with an imminent harm finding.

V. How Pesticide Risks Are Assessed

The type of assessment pertinent to this Petition is the human health risk assessment[41] (ecological risk assessments are not discussed in the Petition). EPA uses a science-based risk assessment approach. Risk is a function of both the hazard associated with a pesticide and how much exposure occurs to that pesticide. Hazard is the innate ability of a pesticide or other stressor to cause an adverse effect (its toxicity). At EPA, hazards are typically identified from the results of testing on several animal species and typically the most sensitive effect is used as

[41] See http://www.epa.gov/pesticides/about/overview_risk_assess.htm

the basis for risk assessment. Exposure is the amount or concentration of a stressor with which an affected individual or group interacts. Risk is a function of both hazard and exposure and represents the likelihood that an individual or group will be adversely affected by that stressor. Both hazard and exposure can differ according to a person's age, thus EPA uses age-appropriate behaviors to determine exposures and also looks at any special sensitivity to pesticides associated with the age of the exposed parties. EPA uses the National Research Council's four-step process[42] for its human health risk assessments. The four steps include: 1) hazard identification, 2) dose-response assessment, 3) exposure assessment, and 4) risk characterization. Each of these steps is summarized below.

It is important to note that risk assessment and risk management are separate activities. Risk management relates to the ways in which the risks characterized in the assessment may be reduced or eliminated. Risk management measures can include tolerance revocation or the cancellation of registrations, termination of some uses of a pesticide, changes to a pesticide's use parameters, and risk reduction training for people who are occupationally exposed, such as pesticide applicators.

A. Hazard identification

The first step in the risk assessment is to identify potential health effects that may occur from different types of pesticide exposure. EPA considers the full spectrum of a pesticide's potential health effects.

Typically, a pesticide active ingredient is subjected to many toxicity studies, and the data from these studies (if determined to be acceptable) are used in risk assessment. Requirements for the relevant data are typically imposed on the pesticide applicant or registrant, and the studies are typically conducted by independent laboratories, with strict standards for data surety.[43] The data are evaluated for acceptability by EPA scientists. The toxicity studies primarily are performed on laboratory animals or *in vitro*, although some of the required studies are conducted in the field. The Agency will also review human toxicity studies, with qualifications, as discussed below, but does not require them. EPA evaluates pesticides for a wide range of potential toxic effects including eye and skin irritation, neurological effects, cancer, and birth defects. In addition to reviewing the required studies, EPA also consults the public literature or other sources of supporting information.

The required tests are used in the assessment of potential health effects in infants, children, and adults. They are conducted for exposures of different durations, as appropriate to represent the durations of exposure anticipated for various lifestages and behaviors. Exposures may be of single day or longer durations, up to and including durations spanning a lifetime. Additionally, EPA considers the route by which these exposures may occur—orally, e.g., through the diet or via children's mouthing behaviors; through the skin; or by inhalation.

[42] The National Research Council produces reports for the National Academy of Sciences. The process is explained at http://epa.gov/riskassessment/health-risk.htm.

[43] Required laboratory practices for studies used to support pesticide registration are detailed at http://www.gpo.gov/fdsys/pkg/CFR-2011-title40-vol24/xml/CFR-2011-title40-vol24-part160.xml

To address risks to infants, children, and adults, EPA typically requires animal testing at multiple life stages, including during gestation and shortly after. The effects that are observed in this type of testing are fetal development (including birth defects) and reproductive success. The results of these studies are particularly applicable to the assessment of risks to the fetus and young children and, when compared to studies with adult animals, provide a basis for evaluating the relative sensitivity of the young to adults.

In order to develop a risk assessment that is protective of human health, the Agency will select the "most sensitive" endpoints and the corresponding point of departure (POD) from among these different studies for the relevant populations, taking into account the durations and routes of exposure. The endpoint can be described as the toxic effect itself. The POD is typically the dose level below the lowest dose at which the adverse effect is manifested.

As an example, consider an endpoint that was selected from a shorter-term animal study in which the animals were exposed via the dermal route, for a pesticide that is registered for use on lawns. In this example, the POD based on this endpoint would be used to estimate risks to adults and to children aged 1 to 2 years old. Children in this age group typically are the most highly exposed relative to children of all ages for pesticide residues on turf by weight and because of their behavior on lawns, as they are exposed to residues through contact with their skin when they play in their yards or in parks and playgrounds, and are also exposed orally, predominantly via hand-to-mouth behaviors.

As noted previously, the Agency also will consider relevant data from sources other than required studies, including data from prospective and retrospective epidemiologic studies, incident reports,[44] and studies in which human subjects have been exposed intentionally to a pesticide (although the latter must undergo an specialized review to ensure that the Agency does not rely on data from studies that violate established ethical standards).[45] EPA uses its draft "Framework for Incorporating Human Epidemiologic & Incident Data in Risk Assessments" when considering those types of data.[46]

B. Dose-response

The second step in the risk assessment process is to consider the dose levels at which adverse effects were observed in test animals and to extrapolate those dose levels to an equivalent dose in humans. For animal studies, the uncertainty around the extrapolation from test animals to humans and the variability of sensitivity in the human population are accounted for by uncertainty factors. The default factors assume there could be up to a ten-fold difference between animals and humans and up to a ten-fold difference between the average person and the most sensitive people in the population. That is, humans generally are assumed to be 10 times more sensitive than animals, and the most sensitive individuals generally are assumed to be 10 times more sensitive than that. These uncertainty factors create a margin of safety for protecting

[44] For example, per analysis of pesticide acute illnesses based on 1998-2006 NIOSH SENSOR data, see http://ehp03.niehs.nih.gov/article/info%3Adoi%2F10.1289%2Fehp.1002843, referenced in the Petition.

[45] http://www.epa.gov/oppfead1/guidance/human-test.htm

[46] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0851-0004

people who may be exposed to the pesticides. The FFDCA requires EPA to use an extra 10-fold safety factor to protect infants and children from effects of the pesticide, unless reliable data show that a different (larger or smaller) factor would protect the safety of infants and children.[47]

C. Exposure

The third step in the risk assessment process is to address how long, and at what level people are exposed to a pesticide, a critical consideration when selecting endpoints and also for calculating risks. For spray drift, exposures are assessed for contact with previously contaminated surfaces such as lawns adjacent to treatment areas. More specifically, the Agency assesses exposures from dermal contact (e.g., children playing on lawns) and also non-dietary ingestion from mouthing behaviors of young children. Adults also are expected to have exposures from dermal contact. For volatilization exposure, the primary exposures result from inhalation since exposure is to the gas or vapor form of a pesticide.

In general, pesticide residues deposited on surfaces (such as grass or the leaves of treated crop plants) remain available for exposure to people entering treated areas, or areas in which spray drift residues have settled, for more than a single day, so subchronic studies are used to derive endpoints and PODs. Single day (acute toxicity) studies also may be considered in order to evaluate risks on the day of application (when the greatest exposures are likely). Impacts on fetuses due to the exposure of pregnant women are included in the risk assessments when information on reproductive and developmental toxicities is available. If information on reproductive and developmental toxicities is not available, an uncertainty factor may be used to account for the possibility of the special sensitivity of children not apparent from available data.

More details are provided in Sections VII and VIII below on how exposures to spray drift and volatilization are determined.

D. Risk characterization

The last step in the risk assessment process is to combine the hazard and exposure assessments to describe the overall risk from a pesticide. Risk characterization explains the assumptions used in assessing exposure as well as the uncertainties[48] that are built into the dose-response assessment, and whether or not the assumptions and uncertainties used are likely to overstate or understate potential risks. The strength of the overall database is considered, and broad conclusions are made.

In summary, the risks to human health from a pesticide depend on both the toxicity of the pesticide and the likelihood of people coming into contact with it. At least *some* exposure and *some* toxicity are required to result in a risk. For example, if the pesticide is very toxic, but no people are exposed, there is no risk. Likewise, if there is ample exposure but the chemical is non-toxic, there is no risk.

[47] EPA may modify these uncertainty factors on a case-by-case basis when supported by chemical-specific behavior.

[48] These are uncertainties not covered by FQPA or other uncertainty factors, e.g. deriving from an incomplete database.

E. Children and pesticides

EPA has developed methods for estimating pesticide exposures to children through the diet and via non-dietary sources such as residential exposures. These methods rely on the best available scientific sources, such as EPA's "Exposure Factor's Handbook."[49] Dietary exposures are based on consumption data for children, and residential exposures are based on methods outlined in the "Standard Operating Procedures (or SOPs) for Residential Exposure Assessment."[50] The SOPs address exposures for various lifestages, and allow for the identification of the most highly exposed lifestage. The SOPs, including the concept of lifestage, have been discussed extensively by the FIFRA Scientific Advisory Panel (SAP).[51] The 2012 revision of the SOPs reflect the input of the SAP.[52]

Young children may have unique exposures that adults do not have because of age-specific behaviors, for example, picking things up from the ground and mouthing them, or putting their hands (potentially contaminated with pesticide residues) in their mouths. They may also come into contact with pesticides when crawling or at play on treated or contaminated surfaces. Children up to adolescence have a higher surface to weight ratio than adults, so they may also be proportionately more highly exposed via the dermal route. Children can also be more highly exposed via the dietary route because they consume more food and water in proportion to body size than adults, and the types of food they eat a lot of tend to contain more pesticide residues.

Available data pertinent to children's health risks are evaluated along with data on adults and the most sensitive, appropriate POD is defined (e.g., NOAEL or no observed adverse effect level) for the most sensitive critical effect(s) based on consideration of all health effects. By doing this, protection of the health of children will be considered along with that of other potentially sensitive populations. In most cases, it is appropriate to evaluate the potential hazard to children separately from the assessment for the general population or other population subgroups.

The approach used by EPA to account for pesticide exposures in children is consistent with EPA's general risk assessment methods[53] and follows the Agency's age grouping guidance.[54] The approach is also consistent with recommendations from the National Academy of Science

[49] http://cfpub.epa.gov/ncea/risk/recordisplay.cfm?deid=236252.

[50] http://www.epa.gov/pesticides/science/residential-exposure-sop.html

[51] The SAP is a federal advisory committee consisting of independent, external scientific experts that advises the Agency's Office of Pesticide Programs (OPP) on technical issues.

[52] The SAP meeting report is at http://www.epa.gov/scipoly/sap/meetings/2009/100609meeting.html.

[53] EPA's risk assessment methods can be found at http://www.epa.gov/risk/guidance.htm; An overview of EPA's approach to assessing and managing these risks is provided in the 2010 report "Protecting Children's Health," found at http://www.epa.gov/pesticides/health/protecting-children.pdf.

[54] An overview of EPA's approach to assessing and managing these risks is provided in the 2010 report "Protecting Children's Health," found at http://www.epa.gov/pesticides/health/protecting-children.pdf: the 2005 paper "Guidance on Selecting Age Groups for Monitoring and Assessing Childhood Exposures to Environmental Contaminants, at http://www.epa.gov/raf/publications/pdfs/AGEGROUPS.PDF, advises risk assessors on selecting appropriate age ranges for use in implementing the Agency's initiatives on pr

1993 report, **"**Pesticides in the Diets of Infants and Children,"[55] cited extensively by the Petitioners. EPA has adopted many of the recommendations from that report into its current risk assessment procedures.

VI. Nomenclature Associated with Spray Drift and Volatilization

EPA uses an informative nomenclature that allows for a clear delineation between the possible forms and/or sources of pesticide movement through the air and away from treated fields. This approach provides for a means of avoiding confusion when describing the unique processes and factors that can contribute to pesticide movement. Whether or not pesticide drift occurs during or after application is an important factor, as is whether or not pesticides are applied in liquid form or as solid material.

As indicated previously, for this response, the Agency focuses on "spray" drift, the off-site movement of aerosols originating with pesticides applied as liquids, rather than dust drift, because spray drift is more likely to occur and generally poses greater risk. Also as noted previously, the Petitioners do not appear to differentiate between drift and volatilization.

Although there are similarities in the mode of transport (through the air) associated with volatilization and spray drift, EPA assesses spray drift and volatilization separately, for several reasons:

- They are two distinctly different processes. Spray drift is dependent on how applications are made and on the form in which the pesticide is applied, while volatilization is driven by the physical and chemical characteristics of the pesticide active ingredient (especially its vapor pressure).
- Spray drift occurs at the time of application and shortly thereafter, for as long as droplets remain aloft. Volatilization, on the other hand, can occur during the application process and also over longer periods of time depending upon the physical and chemical characteristics of the pesticide and how it was applied.
- The route of exposure that the Agency assesses for volatilization is inhalation. The Agency's assessment of spray drift focuses on dermal exposures and exposures from non-dietary oral ingestion (predominantly from hand-to-mouth behaviors in young children). Inhalation exposures are not included in the Agency's spray drift assessments for the following reasons:
    - Most, but not all, of the droplets in spray drift are too large to be respirable.
    - For agricultural pesticides, the Worker Protection Standard (WPS) prohibits the application of a pesticide such that it contacts, either directly or through drift, any worker or other person.[56] This WPS prohibition mitigates the potential for bystander exposure to active drift, but not the residues of drift that are deposited on surfaces to which bystanders may be exposed.

  The Agency at present lacks an assessment methodology for drift inhalation exposures.
- The ways that the risk from volatilization and spray drift can be mitigated are different.

[55] http://www.nap.edu/openbook.php?record_id=2126&page=1.
[56] http://www.epa.gov/pesticides/health/worker.htm

The operating definitions of "spray drift" and "volatilization" are discussed in detail in Sections VII and VIII, respectively.

VII. Assessing and Managing Risks from Spray Drift

EPA has been working with a broad range of public and private stakeholders to address concerns related to spray drift and the potential for adverse effects related to drift exposure.[57] EPA's goal is to assess, and if necessary, to mitigate spray drift via a science-based approach relying on case-specific information.

Spray drift is influenced primarily by environmental conditions (such as wind speed) and application parameters (such as formulation type, application method, application rate, droplet/particle size, application release height). Some degree of pesticide drift is an inevitable result of nearly all types of pesticide application. Even under the best of circumstances, a minute amount of pesticide can move out of the treatment area for a short distance. When the amount of drift is such that it poses risks of concern, the Agency will take action to mitigate those risks.[58]

Quantifying the potential risks of spray drift is a complex process that involves predicting the amount of drift associated with various types of application equipment, estimating potential exposures, and considering the potential health effects from such exposures. Managing the risks associated with spray drift can be complex as well and there are a variety of potential approaches that can be used, as discussed more thoroughly below.

A. Estimating Spray Drift and Potential Exposures to Bystanders[59]

Since the early 1980's, EPA has been working to better understand spray drift. Information key to this effort was developed by a group of pesticide registrants working collaboratively to create a database that addressed spray drift data requirements under 40 CFR 158.440.[60] This group is referred to as the Spray Drift Task Force (SDTF).[61] Since its formation in 1990, the SDTF has generated standardized data on spray drift levels associated with a variety of application methods under varying field and meteorological conditions. The database was reviewed by EPA internally, through external peer review workshops, and by the SAP.[62] Using the database, the SDTF began working with EPA's Office of Research and Development and the USDA's Agricultural Research Service and Forest Service in 2000 to develop and validate a

[57] The Pesticide Program Committee (PPDC) is a federal advisory committee that includes representatives from a broad variety of stakeholders interested in pesticide regulation. See http://www.epa.gov/oppfead1/cb/ppdc/.The PPDC's Spray Drift Workgroup has provided valuable input on the Agency's approach to assessing and mitigating spray drift. Membership of the Workgroup includes environmental advocacy groups, grower associations, registrants, and state officials. See http://www.epa.gov/oppfead1/cb/ppdc/spraydrift/members.htm.

[58] http://www.epa.gov/pesticides/factsheets/spraydrift.htm.

[59] For purposes of this response, bystanders are defined as people who, live, play, or work in areas at proximity to pesticide-treated areas.

[60] See http://www.epa.gov/ocspp/pubs/frs/publications/Test_Guidelines/series840.htm

[61] The Task Force's website is at http://www.agdrift.com/

[62] See http://www.epa.gov/scipoly/sap/meetings/1997/december/spraydrift.htm.

model for predicting the magnitude of off-target movement of pesticides via spray drift, called the "AgDrift" model.

The AgDrift model was developed to assess spray drift under a variety of different conditions for aerial, ground, and orchard air-blast applications. Input features provide the capability to alter over 30 parameters related to the aerial application method including types and numbers of nozzles, weather conditions, and terrain features. AgDRIFT also can provide empirically based predictions for ground and airblast applications made under various conditions, and can accommodate differences in use patterns that relate to crop-specific pest management needs. In addition, users can run the AgDrift model to estimate the amount of spray drift at specified distances from the application site.

Spray drift associated with aerial application has been evaluated extensively by the U.S. Army and the USDA Forest Service, so the drift database is more extensive for aerial applications compared to ground applications. For orchard airblast and groundboom sprayer estimates, AgDrift is more limited; unlike estimates for aerial applications, there is no mechanistic option for these ground applications. Rather, an empirical approach based on the available data is used, and users are more limited in the number of factors that can be considered (e.g., orchard canopy type for airblast sprayers). Even so, AgDrift is a powerful tool for quantifying spray drift for these application methods.

For aerial applications, EPA uses AgDrift to predict conservative estimates of the amount of spray drift given the conditions specified on pesticide labels (when such conditions are not specified on the proposed label, the Agency uses conservative assumptions). AgDRIFT has more limited capabilities to reflect label specifications for ground applications. AgDrift can be used to estimate the risk reduction attributable to buffer zones of specified widths (essentially by estimating the differences in the amount of spray drift at different distances from the application site). In addition to its use in assessing bystander risks, AgDrift can be used in environmental assessments to estimate the potential spray drift exposures to non-target animals and plants. It also is used to estimate the potential contribution of spray drift to pesticide residues in drinking water.

Results from the use of AgDrift represent a range of possible outcomes that are reflective of cultural practices tied to how the target crops are produced and the nature of the pesticide being applied. For example, a contact insecticide application to dense-canopy field crops may be most efficacious when the spray is composed of finer aerosols, while a systemic herbicide applied to a field crop canopy of lesser density, where complete spray coverage is not needed to achieve the desired degree of efficacy, can contain droplets of a larger size (as an aside, larger size or coarse droplets tend to drift less, all other factors being equal).

EPA acknowledges that there is some potential for drift exposures to occur indoors, but believes that the amount of drift entering enclosed structures is very small relative to the amount present out-of-doors. The Agency's efforts to understand the human health risks posed by drift are focused on outdoor exposures.

B. Assessment of Risk to Bystanders from Spray Drift

EPA has been working to refine and standardize the way it assesses the risks to bystanders from spray drift. On December 5, 2013, EPA presented its approach to assessing spray drift to the PPDC. EPA made this new methodology for assessing spray drift available for public comment on January 29, 2014. The announcement and directions on how to submit comments on the proposal can be accessed on the public docket.[63] EPA will continue to conduct spray drift risk assessments under its current process while it reviews and analyzes comments received during the public comment period, which originally was scheduled to end on March 31, 2014, but is being extended by 30 days. After reviewing public comments, EPA plans to finalize its methodology and consider it in cases that warrant spray drift risk assessment.

The methodology for assessing spray drift exposures is based in part on the methodology for assessing residential exposures to pesticides on turf, as explained below,[64] coupled with estimates of the amount of spray drift reaching the area in question, which are derived as described in Section A., above. EPA has developed methods for estimating risks for residential scenarios in which people may be exposed through their use of a pesticide or because they live, work, or play in places where pesticide use occurs. As noted previously, EPA uses its SOPs for residential exposure assessment as the basis for estimating exposures in these situations. These SOPs have undergone extensive external peer review by the SAP.[65] SOPs exist for a wide range of possible exposure scenarios.

The Agency assesses bystander risks from spray drift based on the residential turf scenario, in which people (including children) are exposed to pesticide residues on lawns.[66] If an agricultural pesticide is also registered for use on residential turf, EPA has determined that an additional drift assessment is not necessary beyond that of the residential turf. For an agricultural pesticide not also registered for use on turf, the Agency can use the screening methodology that is included in the new drift methodology, and may be able to conclude, qualitatively, that drift does not pose risks of concern for bystanders, so a quantitative assessment is not needed. When a quantitative assessment is needed, the methodology calls for the use of the AgDrift model to estimate the amount of pesticide residue available on turf for the exposures of adult and child bystanders. The estimated amount of residue and the exposure factors for adults' and childrens' time and activities on lawns are used to calculate exposures through the skin and from the mouthing behaviors (predominantly hand-to-mouth) of children of appropriate developmental age. These exposures are compared to the appropriate endpoint and POD, as discussed in V.B. "Hazard Identification" above.

The development of the SOPs for evaluating lawn pesticides considered a number of factors related to how residues should be quantified, the appropriate behavioral considerations for adults

[63] http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2013-0676

[64] http://www.epa.gov/pesticides/science/residential-exposure-sop.html

[65] http://www.epa.gov/scipoly/sap/tools/atozindex/residentexp.htm and http://www.epa.gov/scipoly/sap/meetings/2009/100609meeting.html

[66] See particularly the Lawns/Turf SOP at http://www.epa.gov/pesticides/science/residential-exposure-sop.html. The SOP identifies default values for exposure parameters e.g., time that a child spends on the lawn, how often children will put their hands in their mouths, etc. These values have been selected so that exposure estimates overall will be conservative.

and children, development of appropriate exposure metrics, how exposures should be combined, and what age groups should be considered as the basis for risk management. EPA considers adults involved in heavy yardwork and children ages 1 to 2 (based on both body weight and play behavior) as the two groups most highly exposed to turf–applied pesticides. EPA has extensively considered children of varying ages in order to ascertain which lifestage (referred to as index lifestage in the SOPs) has the highest relative exposure given behaviors that occur at various stages of development. Children between 1 and 2 years old routinely and very actively engage in outdoor play, and they exhibit mouthing behaviors (predominantly hand-to-mouth) which contribute to the overall exposure levels.

EPA relies on a number of assumptions when using the SOPs in the calculation of risks from spray drift. Risks are based on residue levels present on the day of application when they are at their highest levels because they have not had a chance to dissipate. Risks are also estimated based on a standardized lawn width of 50 feet. The standardized lawn was derived from U.S. Census information for single- and multi-unit dwellings—it is the mean lot width for multi-unit housing and also is a reasonable representation for single-unit housing with smaller lots. A low-end lot size is used because the concentration of spray drift residues is assumed to be inversely correlated to lot size due to the effects of residue dilution in larger lot sizes.[67] Use of "day of application" residues and the standardized lawn size contribute to a data-informed conservative estimate of risk.

C. How EPA Mitigates Potential Risks Associated with Spray Drift

Unacceptable risks associated with spray drift can be mitigated in a number of ways, including changes to application parameters, use of drift reduction technologies,[68] changes to formulations, and no-spray buffer zones, either in combination or by themselves.[69] While the use of buffer zones is one of the key issues raised by the petitioners, other measures also can be used to manage potential risks associated with spray drift. Changes to application parameters and pesticide labels that may mitigate drift risks include reduced application rates; prohibition of certain application methods; soil-incorporation of pesticides at the time of application; and prescriptive, product-specific labeling that requires a particular spray quality (i.e., droplet sizes) or climatic conditions. The Agency may also undertake cancellation of specific uses, in those rare cases where spray drift causes unreasonable risks that cannot otherwise be mitigated.

There is a significant level of effort within the agricultural engineering community to develop both drift reduction technologies and best management practices to reduce spray drift. Useful drift reduction technologies include different forms of spray nozzles and other sophisticated application equipment (e.g., sensors for canopy identification that turn off nozzles

[67] More information on the standardized lawn is found in the proposed methodology at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2013-0676.

[68] http://www.epa.gov/etop/etc_at_psdt.html and http://www.epa.gov/etop/etc_at_proppsdt.html . An instructive presentation on the Drift Reduction Technology Program is located at http://www.epa.gov/oppfead1/cb/ppdc/2012/may/session-7-drift-reduction.pdf

[69] When appropriate, EPA considers the potential consequences of mitigation measures in light of the impact on producers and on the potential for undesirable risk trade-offs.

at ends of rows), and the use of adjuvants.[70] EPA, working with academia and industry, has developed a program to rate drift reduction technologies, and plans to identify on its website tested technologies and the risk reduction potential attributable to them.[71] Other potential means of reducing exposures to spray drift are already commonly accepted,[72] for example, the adjustment of release height in ground applications. Some formulation types are less prone to drift than others; for example, switching to a dry (soil-incorporated) formulation from one that is applied as a liquid may reduce drift potential. A series of possible drift reduction measures are already included in the proposed EPA method for calculating risks from spray drift as a starting point for considering mitigation options should they be required. This element of the method will facilitate consistency in the Agency's decision-making process for managing spray drift risks.

The Agency has used the proposed assessment approach to support pesticide drift risk mitigation in past decision-making. The Agency completed its Preliminary Human Health Risk Assessment for chlorpyrifos (an organophosphate pesticide) under the registration review process in July 2011.[73] That assessment identified risk concerns for bystanders from exposure to spray drift from agricultural applications of chlorpyrifos and provided estimates of the potential risk reductions associated with various drift mitigation options.

In the Spray Drift Mitigation Decision for Chlorpyrifos (July 2012),[74] EPA announced an agreement with the chlorpyrifos registrants for implementing use restrictions intended to reduce spray drift risks to bystanders. In accordance with the agreement, risk mitigation was accomplished through amendments to chlorpyrifos product labels, which were put in place by the end of the same year. Mitigation measures were: buffer zones for groundboom, airblast, and aerial applications of chlorpyrifos around sites such as homes, sidewalks, and recreational areas; and a reduced application rate for aerial applications of chlorpyrifos (from 6 to 2 pounds active ingredient per acre).

VIII. Assessing and Managing the Risks Due to Pesticide Volatilization

Pesticide volatilization can be defined as the change of a pesticide in solid or liquid form to a gas or vapor after application has occurred.[75] Volatilized pesticides can move off-site resulting in the potential for exposure outside the treatment area. The volatilization process is complex and depends on many factors that include the innate physical and chemical properties of the pesticide, the innate characteristics of the site where it is applied, and the atmospheric conditions at the time of application. Other factors that impact volatilization, particularly those associated with application parameters, directions for use, and best management practices are under the

[70] An adjuvant is broadly defined as any non-pesticide material added to a pesticide product or pesticide spray mixture to enhance the pesticide's performance and/or the physical properties of the spray mixture.

[71] http://www.epa.gov/pesticides/factsheets/spraydrift.htm#other

[72] For example, http://pesticidestewardship.org/drift/Pages/default.aspx provides a general overview of existing technologies.

[73] See http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0025.

[74] See http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0103

[75] See http://www.epa.gov/opp00001/about/intheworks/volatilization.htm

control of the user and can be manipulated to reduce off-site movement.[76] For example, soil incorporation of the pesticide, compaction of the soil, and (particularly in the case of fumigant pesticides) tarping and tenting can reduce volatilization from soil- applied pesticides. The use of certain adjuvants may reduce volatilization from pesticides applied to foliage.

Fumigant pesticides are highly volatile.[77] Once applied, they will change into a gaseous form that works by filling the application space or by permeating the soil to kill a wide array of pests. Efficacy is achieved by ensuring that the appropriate air concentration is maintained for the necessary time in order to control the pest of concern.[78] Practices that are used to reduce exposure to (while also improving the efficacy of) fumigant applications include the use of tarps, field conditions management (e.g., soil moisture levels), and the use of specialized application implements (e.g., specially designed shanks for closing up and compacting the soil disturbances to retain the fumigant in soil).

Conventional pesticides (pesticides other than fumigants) tend to be much less volatile than fumigants because of differences in their physical-chemical properties, although some conventional pesticides volatilize under some circumstances. Conventional pesticides also are designed to achieve efficacy via different mechanisms so volatility is not a key required characteristic. When conventional pesticides do volatilize in the field, they too can move outside of the treatment area.

A. Quantifying Volatilization For Conventional Pesticides

EPA has developed a good understanding of the volatilization of fumigant pesticides, as noted above and discussed by the SAP,[79] including an understanding of how use site conditions can impact volatilization. The volatilization of conventional pesticides has not been studied to the same extent. A number of entities are now focusing on the volatilization of conventional pesticides. Research to enhance our understanding of the volatilization of conventional pesticides could include work on the impact of a crop canopy or leaf type on the volatilization process. Air monitoring data are also important in efforts to characterize how much of a pesticide will volatilize and travel out of the treatment area.

The approach used in the past fumigant risk assessments and EPA's proposed approach for conducting volatilization exposure assessments for conventional pesticides consider both single application events and the contamination of ambient air within a local community, region, or the

[76] As in controlling drift, practices that limit the off-site movement of volatilized pesticides can improve the efficacy of an application by keeping more pesticide where it is needed to control the target pests.

[77] Background and status of the Agency's re-evaluation of the fumigants is found at http://www.epa.gov/pesticides/reregistration/soil_fumigants/soil-fum-reg-backgrnd.html

[78] The interaction of concentration and time to achieve efficacy is commonly referred to as the required concentration x time schedule (CxT).

[79] Supporting documents and the final report from the December 1-4, 2009 SAP meeting on "Scientific Issues Associated with Field Volatilization of Conventional Pesticides are located in the docket at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2009-0687.

airshed (the air supply for a defined geographical region) from multiple applications within the same area.

Potential impacts associated with individual application events can be managed more directly through pesticide labels than potential exposures to ambient air. Also, more information can be collected to quantify the volatilization associated with a single pesticide application event than the concentrations expected in ambient air. "Flux" is the term used to describe how much volatilization (or emission) of a pesticide can occur across a given area for specific period of time. Field data can be collected for empirical use in risk assessment and also as the basis for empirically based dispersion modeling. There are a number of recognized flux methods in the peer-reviewed literature.[80] Along with air concentration measurements each method requires that detailed meteorological data be collected and that the conditions of the application are also well- documented.

A large number of flux studies have been completed for fumigants, so their behavior under various field conditions is relatively well understood. Flux data have been also generated for a limited number of conventional pesticides including chlorpyrifos,[81] but in general, they are not available for conventional pesticides. This lack of flux data for conventional pesticides was a primary focus of the 2009 SAP review. EPA presented a number of options for predicting flux in lieu of data which would allow EPA to screen conventional pesticides for potential risk concerns. Building on advice from the SAP,[82] the Agency developed a screening methodology using a preferred approach known as the Woodrow equation.[83]

EPA has recently announced the availability of this screening methodology[84] and is soliciting public input on the new methodology and the proposed approach for assessing volatilization for conventional pesticides. EPA will finalize the approach after considering public comment.

Along with distinct application events, EPA also considers potential exposures to ambient air that may represent those within a community, region, or widespread airshed depending upon where, when, and how such monitoring data were collected. Such data have been used empirically and EPA characterizes such data to the extent possible given available resources. The 2009 EPA SAP analysis provides an example of how an air monitoring result could be characterized by risk assessors based on use information and knowledge of the application site relative to where monitors are placed. To date, EPA has not attempted a modeling approach for

[80] Relevant citations can be found in the bibliography to "Scientific Issues Associated with Field Volatilization of Conventional Pesticides Presented Jointly to the FIFRA Scientific Advisory Panel, USEPA, 2009" at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0687-0006. They include: USEPA, 2008; Majewski and Capel, 1995; Lenoir *et al.*, 1999; Majewski and Baston, 2002; McConnell *et al.*, 1998; Zamora *et al.*, 2003; Glotfelty *et al.*, 1990; Schomburg *et al.*, 1991.

[81] The preliminary volatilization evaluation for chlorpyrifos is found at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0114; supporting documents posted in the same docket discuss the modeling of flux rates.

[82] The final report of the SAP is found at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0687-0037.

[83] See Woodrow *et al.*, 1997 and 2001. Citations found at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0687-0006.

[84] http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2014-0219

predicting ambient exposures to pesticides for these types of circumstances, but plans on exploring such approaches in the future.

Air monitoring data for conventional pesticides are limited and the quality is generally lacking compared to current protocols. The data that do exist mostly come from California; data collection under authority of California's Toxic Air Contaminant statute[85] began in the mid-1980s and continues in the present time. The Washington Department of Health has collected air monitoring data with a focus on organophosphates involved in agricultural production.[86] PANNA (one of the Petitioners) has also collected air monitoring data for a number of pesticides in various locations throughout the United States.[87] The Agency does consider these "Drift Catcher" data for risk characterization, despite certain limitations, and has concluded that the data thus far are not suggestive of concentrations of pesticides in the air that pose significant risks to human health. Other sources of air monitoring data are found in the scientific literature.

B. Estimation of Risks Associated with Pesticide Volatilization

At this time, EPA has conducted at least one volatilization assessment for a conventional pesticide using the fumigant methodology approach.[88] Volatilization risk assessments for the fumigants and conventional pesticides consider distinct, individual pesticide applications as well as ambient air contamination from multiple applications of the same pesticide in the same general area. While single application event risk estimates are based on modeled values, ambient air analyses are based on empirical values; for ambient risk estimation the most representative exposure statistic is selected and compared to the appropriate toxicological value. Additionally, risk assessments for both ambient air and single application events are informed by incident information. Looking for commonalities in the incident information and the predictive risk estimates is a critical consideration for regulatory decision-making.

The Agency's approach to assessing risks associated with single application events is multi-faceted. It includes the use of information on how the pesticide is used; flux data, which are needed for use of the Woodrow equation[89] (or, if flux data are not available, information on physical and chemical properties of the pesticide); and information on the toxicology of the pesticide from the inhalation route of administration (if inhalation toxicity data are not available, other toxicity data may be used). An air dispersion model is used to estimate air concentrations

[85] http://www.cdpr.ca.gov/docs/emon/pubs/tacmenu.htm, and http://www.cdpr.ca.gov/docs/emon/pubs/tac/tacstdys.htm

[86] http://www.doh.wa.gov/ehp/Pest/drift.htm

[87] See http://www.panna.org/science/drift; the Agency considers Drift Catcher data when available and uses them in risk characterization. Unfortunately, the raw data that EPA prefers are not always available and the timing intervals for air samples under the PANNA protocol tend not to permit association with particular applications of the pesticides detected.

[88] Refer to the fumigant risk assessment documents for a detailed overview of the risk assessment process available: http://www.epa.gov/pesticides/reregistration/soil_fumigants/soil-fum-reg-backgrnd.html#information

[89] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2009-0687-0006

of volatilized pesticides surrounding treated fields.[90] Model inputs include standard assumptions about field sizes and the surrounding terrain, and weather data for representative locations. Dispersion modeling is a two-step process. First, flux information is used to characterize how much applied pesticide will volatilize from a treatment area, and then the dispersion of the volatilized pesticide around the treatment area is characterized. Changing weather patterns over time are considered. The mathematical approach to compute how volatilized residues will dissipate is based on a construct known as a Gaussian plume.[91]

EPA has data requirements for information needed to support registrations of pesticides that may result in inhalation exposures.[92] Such data are strongly preferred for use in volatilization risk assessments, but if necessary oral toxicity data can be used. Issues related to the use of oral data for inhalation exposures were discussed by the SAP in 2009.

As noted previously, EPA has recently released a screening methodology for characterizing the potential risks associated with volatilization of conventional pesticides. The Agency has used the methodology to conduct a screening level assessment of all currently-registered conventional pesticides, and found that only about 20 percent of conventional pesticides need to be evaluated further so that the Agency can better understand the potential risks associated with their volatilization and off-site movement. The results of this screening exercise will be released along with the methodology. The Agency may need additional information to perform more detailed assessments, such as data that are needed to model flux, inhalation toxicity data, and information on the pesticide's use parameters. Comments submitted by the public on the screening analysis and its conclusions will be considered as the Agency determines how to proceed. Also as noted previously, the Agency has already conducted at least one volatilization risk assessment for a conventional pesticide, and it can be viewed on the public docket.[93]

C. How EPA Mitigates Potential Risks Associated with Volatilization

After chemical risk assessments are completed, EPA must determine whether there are risks to be mitigated, and if so, how that should be accomplished. EPA has used this process to address risks posed by the volatilization of a pesticide and its off-site movement. Options that can be effective include: buffer zones, reduced application rates, low volatility formulations or adjuvants, tarping and tenting of treated fields, and crop management practices.

The fumigants assessment and mitigation measures provide a framework for considering how to manage potential volatility risks associated with conventional pesticides. For the fumigants, EPA required a suite of complementary mitigation measures to protect handlers, workers, and bystanders from risks resulting from exposure to the soil fumigant pesticides. The measures were designed to work together to address the full range of risks, but were focused on the risks of volatilization to people (workers and bystanders), and included restricted-use status,

[90] http://www.epa.gov/scipoly/sap/meetings/2004/082404_mtg.htm and http://www.exponent.com/perfum/.

[91] Distribution based on the standard bell-shaped curve.

[92] See 40 CFR §158.500.

[93] Chlorpyrifos: http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0130

use site limitations, application rate reductions, and buffer zones with chemical-specific widths.[94] Other risk management measures implemented for the fumigants are not relevant to conventional pesticides, such as the use of tarps over treated fields, which is employed in conjunction with fumigant applications to reduce the off-gassing from soil applications. Risk mitigation measures differ among pesticides because the individual risk assessments were based on chemical-specific information.

IX. Responses to Requests Made by Petitioners

EPA reads the petition to make three specific requests for programmatic changes: 1) that EPA evaluate the risks to children exposed to pesticide drift and volatilization for all pesticides, 2) that EPA modify its pesticide re-evaluation process to expedite assessment of these risks, and 3) that, for certain types of pesticides, the Agency require the adoption of generic buffer zones between treated areas and places where children could congregate.[95] The Agency's responses to each of these elements follow.

Although this petition addresses how EPA assesses risk under the FFDCA, it does not specifically request to cancel registrations or modify or revoke specific tolerances. The Petitioners also are requesting that EPA require interim buffers for all "drift-prone" pesticides during the time EPA makes the programmatic changes they have requested. Because the Petitioners are suggesting specific changes to use practices but not requesting cancellation of registrations, we also have interpreted the petition to request that EPA attempt to procure voluntary label amendments from the registrants. However if the registrants did not agree, Agency-initiated cancellation actions would likely be needed to achieve the requested relief.

A. EPA Will Evaluate the Risks to Children Associated with Spray Drift and Volatilization Exposures.

While it is true that EPA has not always assessed the risks to children from spray drift and volatilization, the need for consistent and refined methods have led to the development of appropriate methodologies for doing so. The development of these methods is described in detail in Sections VII and VIII of this response.

The Petitioners are requesting that pesticide drift and volatilization risk assessments be conducted for all pesticides. We agree. The first step in EPA's new spray drift assessment methodology is a screening process to facilitate the identification of conventional pesticides that could pose risks of concern to bystanders through spray drift. As previously noted, the Agency released the draft spray drift assessment methodology earlier this year. Elements of the "needs

[94] http://www.epa.gov/pesticides/reregistration/soil_fumigants/implementing-new-safety-measures.html#risk

[95] As noted earlier, the Agency considers residential exposures to include exposures associated with homes, home lawns, yards, gardens, apartments and grounds around apartment buildings, schools, schoolyards, daycare facilities, playgrounds, athletic fields, and parks and other public spaces.

screening" process are detailed in materials posted to the docket on "Consideration of Spray Drift in Pesticide Risk Assessment."[96] The new volatilization methodology is designed to serve as a stand-alone screening methodology for any and all conventional pesticides. As noted above, the Agency has already used the volatilization methodology to screen all currently-registered pesticides. The screening processes include consideration of factors such as methods of application and use patterns of the subject pesticide. Using the screening procedures for both spray drift and volatilization, EPA considers the potential for exposure in a qualitative way. In the case of spray drift, pesticides for which the screen indicates there are potential risk concerns will undergo a quantitative assessment. Thus, we grant the Petitioners' request to conduct spray drift and volatilization assessments for all pesticides, while noting that some of these assessments will not provide quantitative results.

In the last several years, EPA has conducted a number of spray drift and volatilization risk assessments, even while the proposed assessment methodologies were being developed and refined. EPA conducted volatilization assessments for the fumigant pesticides during reregistration.[97] Chlorpyrifos also recently underwent a spray drift evaluation,[98] while the recently released proposed drift assessment methodology[99] was still in development (see Section VII B). The preliminary human health risk assessment for chlorpyrifos was completed in July 2011[100] and attendant risk reduction measures for spray drift exposures were implemented in July 2012.[101] EPA also recently published a draft volatilization risk assessment for chlorpyrifos[102] and is working to finalize the assessment even as work continues on the volatilization methodology. The methodologies set out standardized procedures so that the way spray drift and volatilization are assessed will be consistent between pesticides in general, but they are not substantively different from the approaches that were used to assess spray drift and volatilization for pesticides in the recent past.

Further evidence of EPA's commitment to reducing the risks to children exposed to pesticides is demonstrated by actions taken during pesticide reregistration process to terminate residential uses, as was done with many organophosphate insecticides. EPA now has better-developed tools for determining when spray drift poses risks to bystanders, and is committed to taking action on risks to bystanders, including children, during registration review.

The Petitioners also are requesting that EPA include the drift and volatilization exposures of children in its aggregate assessments. Including spray drift and volatilization in EPA's aggregate risk assessments would involve the following steps. First, EPA would look at exposures from drift and volatilization under the new policies to determine whether any non-negligible exposure

[96] Process described at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2013-0676-0003.

[97] Based on the findings from these assessments, EPA required the implementation of risk mitigation measures, including measures intended to protect bystanders. The risk mitigation measures are detailed at http://www.epa.gov/pesticides/reregistration/soil_fumigants/implementing-new-safety-measures.html.

[98] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0105

[99] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2013-0676-0001

[100] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0025

[101] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0103

[102] http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0114

due to drift or volatilization could occur. If non-negligible exposure could occur, EPA would quantitatively assess that exposure under the new policies and then aggregate that exposure with other sources of exposure consistent with existing policies on aggregate exposure. However, aggregation of drift and volatilization exposure with other sources of exposure is not specifically addressed in existing aggregate exposure policies, and thus EPA's approach may need to be modified to account for the factors involved in drift and volatilization exposure. Finally, if initial aggregate exposure estimates using conservative methodologies indicate there could be a risk of concern, EPA would need to develop efficient approaches to refining those assessments.

EPA fully agrees with the Petitioners that exposures to spray drift and volatilized pesticides should be considered in our risk assessments, and that the risks to children, with their unique biology and behaviors, must be considered separately from risks to adults. The Agency has developed the methodologies for assessing drift and volatilization and is committed to considering the comments of the public on them so that we may employ the best possible science in assessing these risks and taking action to mitigate risks as needed.

B. Expediting Assessments of Spray Drift and Volatilization Outside of the Registration Review Schedule is not Necessary

Petitioners state that EPA should accelerate its schedule for completing drift and volatilization risk assessments and prioritize pesticide reviews based on the suspected degree of risks posed to children. Petitioners suggest that this acceleration should be accomplished through modifications to the registration review process or by utilizing other authorities. Petitioners believe that registration review is too slow to be protective of drift and volatilization risks to children.[103] EPA's response to this request covers aspects of public health policy and the Agency's obligations under FIFRA and the FFDCA, and issues of efficiency.

The Agency is denying the Petitioners' request to the extent that they ask EPA to perform bystander risk assessments of the chemicals highlighted in the Petition before considering other exposures and risks. The registration review program is an appropriate and risk-protective approach for evaluating and managing the risks associated with spray drift and volatilization (and other types of risk as well). Utilizing a process outside of registration review to assess these risks to children separately from other types of risk would bypass and defer the comprehensive evaluations that allow the Agency to rely on the best and newest developments in science and to address the full complement of potential risks. The consideration of all potential risks at one time for a single active ingredient can lead to the adoption of a package of risk mitigation measures that works for all the risks of concern, or at the very least, that do not work in opposition to each other.

[103] The Petitioners also assert that because the registration review process is designed to update the reregistration risk assessments, and since the Agency did not address the risks to bystanders from drift and volatilization during reregistration, registration review will not include such assessments. In reality, the Agency updates risk assessments on the basis of available data and scientific developments, so drift and volatilization will be included in registration review, even if they were not considered during reregistration.

To set the schedule for registration review, EPA relied primarily on the baseline date for each pesticide case (usually its RED date or the date the first product containing the active ingredient was registered). Additionally, some registration review cases were grouped for program efficiency. The OPs and NMCs were among those cases.[104] For the most part, food- use pesticides subject to reregistration were given priority scheduling in reregistration, and thus, they have the earliest baseline dates. The OPs and NMCs, on which the Petitioners focus, have been scheduled at the front-end of registration review and many individual pesticides from those families are currently undergoing risk assessment. EPA believes that, insofar as the Petition requests EPA to give high priority to certain chemicals in its registration review program, the petition asks for an action that has already occurred.

The OPs and NMCs account for more than 40 *individual* active ingredients. All of these pesticides have entered registration review (or were canceled prior to entering registration review).The preliminary risk assessments for 12 of the pesticides in these two families are scheduled to be completed before October 2014.The schedule for OP and NMC registration reviews[105] is summarized in the Appendix to this response.[106] The Appendix also identifies the pesticides in these groups that were cancelled subsequent to tolerance reassessment or during the beginning stages of registration review.

With respect to conducting separate bystander exposure assessments of individual pesticides, if the Agency granted Petitioners' proposal, it would significantly reduce the efficiency of the overall registration review program. Separating the bystander drift risk assessment for children from the ongoing comprehensive evaluations for these same chemicals would require Agency resources to be redirected to the evaluation of one type of potential risk, and management of the full complement of potential risks associated with a pesticide would be deferred. In addition, the overall demand for resources and the time needed to assess first spray drift and then all other potential risks for a given pesticide would be greater in total than the time and resources needed to conduct a comprehensive assessment of that pesticide, and thus would slow Agency action on risk management. Registration review, as currently planned, is the most efficient way to achieve the Petitioners' and EPA's common goal of protecting human health, including the health of children, and the environment. Adopting the approach proposed by the Petitioners also would significantly reduce EPA's ability to meet its statutory obligation to complete registration review by 2022 or the date that is 15 years after the date on which the first pesticide containing a new active ingredient is registered. *See* FIFRA § 3(g)(1)(A)(iii).

The same logic applies to the idea of assessing chemical-specific volatilization risks separately from the comprehensive registration review assessment. Significantly, preliminary application of the volatilization screening methodology currently under development led the Agency to conclude that only 20% or so of all pesticide active ingredients have characteristics that suggest that they potentially could pose any meaningful level of volatilization risk. Thus, including the volatilization risk assessment in the registration review process as a matter of

[104] Described in a Federal Register Notice: http://www.gpo.gov/fdsys/pkg/FR-2006-10-11/html/E6-16483.htm.

[105] http://www.epa.gov/pesticides/cumulative/ identifies the organophosphate and n-methyl carbamate pesticides by name.

[106] The full schedule is at http://www.epa.gov/oppsrrd1/registration_review/schedule.htm.

course will not appreciably affect the resources needed or timing for the vast majority of registration review decisions.

Petitioners suggest that EPA could accelerate the reviews of drift risks for children by utilizing other authorities, such as rulemaking[107] or the special review process.[108] Rulemaking is a long, resource-intensive process that can take many years to complete, and EPA believes its limited resources are better spent assessing and developing risk mitigation measures for pesticides individually than developing a regulation that could take many years to finalize. As an example, the Petitioners suggest that the WPS Rule is an appropriate model for implementing broad changes for a large number of pesticides at once. Although the WPS is an important and effective tool for reducing worker risk, it took approximately eight years to develop and promulgate the initial 1992 rule and as many years to develop a set of proposed amendments to the 1992 rule.[109]

And while Special Review served its purpose in the past, individual Special Reviews typically took many years to conclude and used Agency resources to address a narrow set of risk concerns at a time when there was no systematic re-evaluation process for pesticide registrations. Indeed, in 2009, the Agency announced that "[t]he pesticide program is moving toward closing out both the Special Review and the reregistration programs" in favor of new re-evaluations of previously registered pesticides to be conducted under registration review.[110] That is not to say that potential risk concerns that rise to the highest levels, including "emergency" or "imminent hazard" status, must wait to be addressed in registration review—the processes for such situations include those described in Section IV of this response.

Based on these considerations, for existing registrations, EPA has concluded that registration review, as currently planned, is the most appropriate, timely, and efficient way to achieve the Petitioners' and EPA's common goal of protecting human health, including the health of children, and the environment.

C. Immediate Adoption of Interim No-Spray Buffers Around Homes, Schools, Daycare Centers, and Parks to Protect Children from Drift Is Not Appropriate

Petitioners request that EPA impose interim no-spray buffers around locations where children congregate and that these measures should apply to OPs, NMCs, and all other pesticides that are: "(1) registered for application by ground sprayers, broadcast equipment, and/or aerial equipment; and (2) suspected of causing acute poisonings, cancer, endocrine disruption,

[107] Although Petitioners mention EPA's general rulemaking authority under FIFRA, EPA is not treating this petition as a specific request for rulemaking. Instead EPA understands Petitioners' statements being made as actions that could be taken by EPA. The Agency's general rulemaking authority is provided at 7 U.S.C. § 136w(a)(1).

[108] 40 C.F.R. § 154.7

[109] The revisions to the WPS were just released for public comment on February 20, 2014--http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2011-0184-0002.

[110] http://epa.gov/oppfead1/cb/csb_page/updates/2009/namechange-prd.html

developmental effects, and/or reproductive effects."[111] Petitioners further state that the interim buffers should be at least 60 feet in width for ground applications and 300 feet in width for aerial applications regardless of the pesticide being applied, and that these buffer requirements should remain in place at least until case-specific drift risk assessments can be undertaken. To accomplish this request, Petitioners further suggest that the Agency could use administrative procedures rather than chemical-specific risk assessment and management to effect the generic buffers i.e., rulemaking or a Pesticide Registration (PR) Notice. The Agency's response to this request covers the usefulness of the alternate approaches, the scientific basis for pesticide decision-making, and issues of efficiency.

The Agency denies the Petitioners' request to impose a requirement for interim buffers of either 60 or 300 feet on these pesticides before EPA completes the registration reviews for these pesticides. EPA contends that drift and volatilization are not posing risks of concern for all pesticides, and that interim buffers, as suggested by the Petitioners, are not the most efficient or scientifically appropriate way to mitigate such risks for any particular pesticide or group of pesticides.[112] It is the Agency's practice to assess pesticide risks based on chemical-specific data. The Agency acknowledges that the OPs and NMCs are generally among the more acutely toxic pesticides, but risk is a function of both toxicity and exposure, so toxicity alone is not sufficient to characterize the risks these pesticides may cause to bystanders via drift or volatilization, or to determine if risk mitigation is needed. Additionally, the OPs and the NMCs were reevaluated both in reregistration and in the tolerance reassessment process and, at that time found to meet the applicable statutory safety standards. These pesticides will be reassessed again in the next few years under registration review. Because pesticides vary in environmental fate characteristics, and use sites and parameters, potential exposures also differ, not just between different active ingredients, but also between different uses of the same active ingredient.

The pesticides other than the OPs and NMCs that the Petitioners believe warrant the use of 60 and 300 foot buffers are a very large and diverse group. Without considering pesticide-specific data, it is impossible to know the risks posed by each. Again, the manner in which the Petition proposes to manage the drift and volatilization risks associated with these pesticides ignores the interaction of exposure and hazard. When these pesticides underwent reregistration, the Agency found that (with certain conditions) they met the statutory standards of FIFRA and FFDCA. In order to make the same type of determinations when spray drift and volatilization exposures are appropriately considered for each individual case, the Agency must satisfy the same standards.

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[111] While an exact count of pesticides that fit in this second category cannot be made, the effects listed are a large subset of the complete set of adverse effects the Agency takes into consideration. Furthermore, EPA notes that the Petition describes the referenced pesticides and groups of pesticides as "drift prone." The Agency rejects this notion. Because drift is influenced by factors other than the characteristics of the pesticide active ingredient, primarily the physical form of the product as applied, the application method, climate, and wind, no one active ingredient or pesticide family can be considered to be drift-prone. There are particular formulations and application methods that may make certain applications of a pesticide product prone to drift. Volatilization, on the other hand, is a direct function of the physical and chemical properties of an active ingredient, and the Agency is able to identify pesticides that tend to volatilize.

[112] EPA does not believe the Petitioners have presented adequate justification for interim, across-the-board buffers for all the pesticides they have identified as being of special concern.

Even if buffer restrictions may be appropriate to mitigate the risks to children from spray drift and volatilization for some pesticides, the EPA contends that the same buffer width will not be appropriate for each of them. Despite the Petitioners' request for across-the-board, interim buffers, the Petition itself makes the point that case-specific assessments have shown that buffers of varying widths are needed to mitigate risks associated with different pesticides. They cite examples of the different buffer widths that EPA determined were necessary for products contain different active ingredients—e.g., for the fumigants, widths ranging from 25 ft to one-half mile. EPA believes that the more scientifically defensible approach involves determining whether no-spray buffers are necessary to protect children on a pesticide-by-pesticide basis. This is the approach the Agency has taken and intends to include in consideration of bystander exposure in future risk assessments.

Finally, the Agency believes that the requests made by the Petitioners with regard to interim buffers would divert limited Agency resources from important risk assessment and risk management activities and would diminish the overall level of protection EPA is able to achieve in its pesticide re-evaluations. The alternate means that the Petitioners suggest to implement the buffer requirement are also resource-intensive, time-consuming, and not likely to result in the broad protections the Petitioners desire. The resource and time requirements of rulemaking are discussed above.

The Petitioners suggest that EPA could use a Pesticide Registration (PR) Notice[113] to inform registrants that label amendments are necessary to address drift and volatilization, and, that failure to make these changes could result in cancellation or the finding that their products are misbranded. Although EPA agrees that a PR Notice is a useful tool to communicate new policies to pesticide registrants, compliance with a PR Notice is voluntary, and the Agency believes that registrants are not likely to implement changes that lack a risk-based rationale. If EPA took action to require the amendment of pesticide registrations to mitigate spray drift via buffers of uniform width, pesticide applicants and registrants could challenge the validity and applicability of the science behind the Agency-prescribed regulatory actions. Additionally, the resources needed to pursue a cancellation proceeding are extensive, and in the absence pesticide-specific assessments, would not be an effective use of EPA's limited resources.

X. Conclusion

The Agency appreciates the concern the petitioners express for bystanders, particularly children, who may be harmed by exposure to spray drift and the off-site movement of volatilized pesticides. We share this concern. The Agency has assessed spray drift and volatilization for particular pesticides in the past and is now taking steps to formalize the assessment methodologies for future assessments. These methodologies include screening-level assessment processes for use in determining if there is a need to take a more in-depth look at any given pesticide. Thus, all pesticides will be assessed either qualitatively or quantitatively. The Agency also believes that we are addressing pesticide risks on a schedule that gives the most potentially

[113] PR Notices are issued by EPA's Office of Pesticide Programs to inform pesticide registrants and other interested persons about important policies, procedures and regulatory decisions. The Agency's PR Notice webpage is at http://www.epa.gov/PR_Notices/.

risky pesticides precedence. We will continue to use approved approaches to account for the differences between children and adults in their exposures and sensitivity to pesticides.

The Agency believes that its current program of registration review is the most comprehensive and effective way to assess and mitigate pesticide risks and to take advantage of new and emerging science. The Agency believes that looking at a particular pesticide and all the potential risks associated with its use in a comprehensive fashion provides the best opportunity to effectuate necessary protections for human health and the environment. Other means of managing the risks posed by spray drift and volatilization as suggested by the Petitioners, such as conducting drift and volatilization-only assessments or re-ordering the pesticides in registration review, are either not needed, not likely to be successful, require more resources than are available, and/or take more time than registration review.

While we understand the thinking behind the proposal to mandate generic, uniform buffer requirements on pesticides of particular concern, we do not agree that a "one size fits all" solution is appropriate or scientifically defensible. Without case-specific assessments and risk mitigation, we believe it is unlikely that registrants will voluntarily adopt generic buffers. Pesticide registrants are by law afforded specific rights and opportunities to oppose decisions by the Agency that affect their registrations. Because the evidence needed to support the cancellation or amendment of registrations within this context must be scientifically defensible and specific to the subject pesticide, we believe that Agency resources are better spent in registration review.

APPENDIX

Registration Review Timelines for Pesticide Families Specifically Cited in the Petition

Table 1. Anticipated Organophosphate Registration Review Milestones[114]

Active Ingredient	Docket Opening	Draft Human Health Risk Assessment	Final Decision
Acephate	3/18/09	2014	2015
Azinphos-methyl	All registrations cancelled 2012		
Bensulide	6/25/08	2015	2015
Chlorethoxyfos	12/17/08	2014	2015
Chlorpyrifos	3/18/09	July 6, 2011	2016
Chlorpyrifos-methyl	3/13/10	2015	2016
Coumaphos	6/25/08	2014	2015
Diazinon	6/25/08	2014	2016
Dichlorvos	6/24/09	2015	2016
Dicrotophos	6/25/08	2014	2015
Dimethoate	3/18/09	2014	2015
Disulfoton	3/18/09	All registrations subsequently cancelled	
Ethoprop	12/17/08	2014	2015
Fenamiphos	All registrations cancelled 2007		
Fenitrothion	3/18/09	2015	2015
Fenthion	All registrations cancelled 2004		
Fosthiazate	6/24/09	2013	2014
Malathion	6/24/09	2014	2016
Methamidophos	12/17/08	All registrations subsequently cancelled	
Methidathion	3/18/09	All registrations subsequently cancelled	
Methyl parathion	6/24/09	All registrations subsequently cancelled	
Naled	3/18/09	2015	2016
Oxydemeton-methyl	6/25/08	All registrations subsequently cancelled	
Phorate	3/18/09	2015	2015
Phosmet	6/24/09	2015	2015
Phostebupirim	6/24/09	2016	2017
Pirimiphos-methyl	3/18/09	2015	2016
Profenofos	6/25/08	2015	2016
Propetamphos	6/25/08	All registrations subsequently cancelled	
Phosalone	2/19/08	All registrations cancelled effective 12/30/15	
Temephos	6/25/08	All registrations subsequently cancelled	
Terbufos	6/25/08	2014	2015
Tetrachlorvinphos	6/25/08	2014	2015
Tribufos	3/18/09	2015	2016
Trichlorfon	3/18/09	2015	2016

[114] Dates in the future should be considered tentative.

Table 2. Anticipated N-Methyl Carbamates Registration Review Milestones[115]

Active Ingredient	Docket Opening	Draft Human Health Risk Assessment	Final Decision
Aldicarb	6/20/12	2015	2017
Carbaryl	9/22/10	2016	2018
Carbofuran	All registrations cancelled 2009		
Formetanate HCl	12/22/10	2016	2017
Methiocarb	6/22/10	2015	2016
Methomyl	9/22/10	2015	2016
Oxamyl	9/22/10	Early 2015	2016
Pirimicarb	All registrations cancelled 2010		
Propoxur	12/16/09	2014	2016
Thiodicarb	12/16/09	2015	2016

[115] Dates in the future should be considered tentative.

EXHIBIT 7

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

DEC 18 2012

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Re: Chlorpyrifos petition dated September 12, 2007

Dear Mr. Colangelo and Dr. Reeves:

I am writing to provide you with an update on the U.S. Environmental Protection Agency's (EPA) plans for further responding to the petition dated September 12, 2007 (Petition), submitted jointly by the Natural Resources Defense Council (NRDC) and Pesticide Action Network North America (PANNA). The petition specifically requested that EPA revoke all tolerances and cancel all registrations for the insecticide chlorpyrifos.

As you are aware, in a letter dated July 16th of this year, EPA provided you with a partial response to six of the 10 claims raised in the petition and outlined its intended approach for completing work on the remaining four claims. At the same time, EPA partially granted your petition with its response to one part of your inhalation exposure risk claim, announcing that EPA was taking action to address risks from primary spray drift by limiting application rates and imposing buffer zones around sensitive sites adjacent to agricultural applications. I am pleased to announce that registrants have submitted draft amended labels for all agricultural use products to implement these additional use limitations. EPA anticipates its approval of these 41 amended labels by December 31, 2012.

In the partial response, we also informed you that EPA intended to provide its written response to the remaining issues by December 2012. We noted that it was our intention that the response be informed by the recommendations of the July 2012 FIFRA Scientific Advisory

Panel (SAP or Panel) report[1] that addressed issues relevant to three of petitioners' remaining four claims -- that EPA failed to quantitatively incorporate data exhibiting long-lasting effects from early life exposure to chlorpyrifos in children; that EPA disregarded data demonstrating that there is no evidence of a safe level of exposure during pre-birth and early life stages; and that EPA failed to cite or quantitatively incorporate studies and clinical reports suggesting potential adverse effects below 10% cholinesterase inhibition. Further, we also noted in the partial response that our work on the volatilization component of the fourth remaining claim (inhalation exposure) was also ongoing and would be impacted by the results of the SAP report. However, because EPA had just received the SAP report prior to the release of the partial response, EPA had not completed its review of the SAP's recommendations at that time. Thus, the extent and nature of the work necessary to address those recommendations was an uncertainty.

The recent SAP report contained several recommendations which require additional analyses by EPA to address the toxicology issues raised in your petition. Specifically, the SAP recommended that EPA conduct a dose reconstruction analysis of potential exposures to women and children studied in the Columbia University-sponsored epidemiology study[2] as an approach to aid in assessing the degree to which individuals in the cohort may or may not have experienced acetylcholinesterase inhibition. In addition, the Panel recommended that additional analyses of the epidemiological data be conducted, particularly in the areas of biological marker of exposure data and multi-chemical exposures.

EPA has made progress on the dose reconstruction analysis. However, the analysis of the biomarker of exposure data and evaluation of the multi-chemical exposures suggested by the Panel necessitate that EPA obtain the raw data from the epidemiology study. At this time, EPA only has access to summary information provided by the publications, but has been working to obtain the original data from the study authors to conduct the needed analyses.

Two additional considerations have necessitated further analysis of the toxicology issues raised in your petition. First, members of the Panel expressed concern during the oral deliberations that scientific experts in diagnostic and analytic tools, like those used to assess neuro- and motor development of the children in the epidemiology studies, were not included on the Panel. Second, after the SAP was held, a new epidemiology study from the Columbia University researchers describing the results of magnetic resonance imaging (MRI) on a subset of children in the cohort was published. Between August 2012 and October 2012, EPA solicited comments from scientists within the federal government who have expertise in these two scientific areas and is currently evaluating this input.

With respect to the volatility of chlorpyrifos, EPA has reviewed a new field volatility study recently submitted by Dow AgroSciences in response to the data call-in requirements for the chlorpyrifos registration review. EPA is currently working to complete its assessment of the

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[1] Available at http://www.epa.gov/scipoly/sap/meetings/2012/april/041012minutes.pdf

[2] Rauh, V., Arunajadai, S., Horton, M., Perera, F., Hoepner, L., Barr, D. B., & Whyatt, R. (2011). Seven-year neurodevelopmental scores and prenatal exposure to chlorpyrifos, a common agricultural pesticide. Environ Health Perspect, 119(8), 1196-1201. doi: 10.1289/ehp.1003160; Rauh, V. A., Garfinkel, R., Perera, F. P., Andrews, H. F., Hoepner, L., Barr, D. B., Whyatt, R. W. (2006). Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics, 118(6), e1845-1859. doi: 10.1542/peds.2006-0338.

potential risks associated with volatilization from chlorpyrifos applications. The scope and content of this on-going assessment is informed by recent risk assessments of field volatility of fumigant pesticides[3], Dow AgroSciences' recently submitted chlorpyrifos field volatility study coupled with existing volatility data found in the open literature, EPA modeling tools, and the report and recommendations from the 2009 SAP meeting[4] on pesticide volatilization where chlorpyrifos was one of the case studies presented.

While EPA has been working diligently, as outlined in the preceding paragraphs, and has made significant progress in addressing the recommendations of the SAP and completing our response to all four remaining claims in your petition, EPA will not be in a position to provide a complete response to the petition this month, as we previously believed. EPA currently intends to provide a further response to the petition by the end of January 2013 that will address some, but likely not all, of the four remaining claims. To the extent certain issues remain unaddressed, the January 2013 response will explain the additional work we will be doing and will set forth our anticipated timeline for completing the response.

Steven P. Bradbury, Ph.D.
Director, Office of Pesticide Programs

[3] The assessments can be found in the dockets for each fumigant. Four of which are provided here chloropicrin - EPA-HQ-OPP-2007-0350; dazomet - EPA-HQ-OPP-2005-0128; metam sodium/potassium - EPA-HQ-OPP-2005-0125; and methyl bromide - EPA-HQ-OPP-2005-0123.
[4] Available at http://www.epa.gov/scipoly/sap/meetings/2009/december/120309meetingminutes.pdf.

EXHIBIT 8

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

JAN 25 2013

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Re: Chlorpyrifos petition dated September 12, 2007; January 2013 Response

Dear Mr. Colangelo and Dr. Reeves:

I am writing to further update you on the U.S. Environmental Protection Agency's (EPA) efforts to respond to the Natural Resources Defense Council (NRDC) and Pesticide Action Network North America (PANNA) jointly submitted September 12, 2007[1], petition and our related efforts to complete the registration review of chlorpyrifos. In my letter to you of December 18, 2012[2], I provided you with an update on our efforts to implement label changes to put in place additional limitations to reduce primary spray drift from chlorpyrifos. I can report that EPA has now approved those changes for all 41 chlorpyrifos agricultural products subject to these use limitations.

As we also noted in December, while we have made significant progress in completing work on the four petition issues that EPA did not address in its July 16, 2012[3], partial response to your petition, we were not able to provide you with a complete response in December, as we previously believed we could. However, we committed to providing you with a response this month that further addresses the petition and outlined the approach we are taking for completing our response. Accordingly, this response will address what EPA has done and will do to address each of the following four outstanding claims that: (1) EPA failed to incorporate inhalation routes of exposure from pesticide volatilization; (2) EPA failed to incorporate into its risk

[1] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0005
[2] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0096
[3] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0095.

assessment, in a quantitative manner, data indicating that long-lasting effects result from early life exposure to chlorpyrifos in children; (3) EPA disregarded data demonstrating that there is no evidence of a safe level of exposure during pre-birth and early life stages; and (4) EPA failed to cite or quantitatively incorporate studies and clinical reports suggesting potential adverse effects below 10% cholinesterase inhibition.

As I indicated in the December response, EPA has been working to complete an assessment that will evaluate the potential risks of volatilization from chlorpyrifos applications. In early February 2013, we will publish a notice in the Federal Register announcing the availability of this preliminary assessment for public comment. This assessment represents a significant advancement in the evaluation of pesticide risks, as it will be the first probabilistic assessment of the risks posed by the post-application volatilization of a semi-volatile pesticide. Our approach builds upon the methodology we previously employed for volatile pesticides in the recent fumigant pesticide risk assessments[4] to assess bystander inhalation exposure from volatilization. In addition, it is consistent with the recommendations from the December 2009 Federal Insecticide, Fungicide, and Rodenticide Act Scientific Advisory Panel (SAP)[5] meeting on the scientific issues associated with field volatilization of conventional (semi-volatile) pesticides. The content of the preliminary volatilization assessment is further informed by Dow AgroSciences' recently submitted chlorpyrifos field volatility study[6] coupled with existing volatility data found in the open literature, and EPA modeling tools.

This assessment will supplement the July 2011 Preliminary Human Health Risk Assessment[7] (HHRA) and evaluates bystander exposure from chlorpyrifos and chlorpyrifos-oxon emitted from treated fields. Although the volatilization of chlorpyrifos was addressed in the preliminary HHRA, that analysis involved only a deterministic assessment based on limited monitoring data that did not attempt to evaluate a range of field conditions and, therefore, had correspondingly limited utility in a regulatory setting. Given the groundbreaking nature of the new assessment and its potential for use in guiding additional risk mitigation, EPA believes it is critical to involve the public in the development of this assessment before it is finalized. Further, EPA is examining other semi-volatile pesticides to determine if bystander volatilization assessments are needed. Any comments received on this assessment will serve to inform those assessments as well. Accordingly, EPA will begin taking public comment on the draft version of the assessment in February 2013, after publication of the Federal Register notice announcing its availability in docket number EPA-HQ-OPP-2008-0850.

Following completion of the public comment period and EPA's subsequent evaluation of the comments, EPA will determine whether additional regulatory action is necessary to address

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[4] The assessments can be found in the dockets for each fumigant. Four of which are provided here chloropicrin - EPA-HQ-OPP-2007-0350; dazomet - EPA-HQ-OPP-2005-0128; metam sodium/potassium - EPA-HQ-OPP-2005-0125; and methyl bromide - EPA-HQ-OPP-2005-0123

[5] U.S. EPA 2009. FIFRA Science Advisory Panel Meeting Minutes - Scientific Issues Associated with Field Volatilization of Conventional Pesticides. Available at http://www.epa.gov/scipoly/sap/meetings/2009/december/120309meetingminutes.pdf

[6] Rotondaro, A. and Havens, P. (2012). Direct Flux Measurement of Chlorpyrifos and Chlorpyrifos-Oxon Emissions Following Applications of Lorsban Advanced Insecticide to Alfalfa; Sponsor: Dow AgroSciences LLC, 9330 Zionsville Road Indianapolis, IN 46268-1054. EPA MRID 48883201.

[7] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0025.

these risks and, if so, whether the nature of that risk supports the need to take action in advance of our completion of the final broader HHRA, currently scheduled for December 2013.

Regarding the remaining three petition issues addressing chlorpyrifos toxicity identified above, as we have indicated previously, the analysis is complicated and multifaceted because it involves many lines of scientific evidence, including many recently conducted studies and peer review evaluations and recommendations. That work includes consideration of: *in vivo* and *in vitro* experimental toxicology studies that evaluate neurodevelopmental effects in laboratory animals, adverse outcome pathway framework analyses, exposure, the results of multiple human epidemiology studies, and biomonitoring data. Notwithstanding the complexity of this analysis, it was our hope to provide you with a written response last December that included our scientific conclusions on these issues. As you know, we convened a FIFRA SAP meeting in April 2012[8] to inform our work in generating a weight-of-evidence evaluation integrating the epidemiologic data with the experimental toxicology studies for the neurodevelopmental outcomes and acetylcholinesterase (AChE) inhibition. At the time EPA provided its partial petition response to you in July 2012, EPA had just received the written SAP report from the April meeting. EPA therefore had not had time to begin pursuing the SAP's recommendation when EPA provided its response to you and to the 9th Circuit in our ongoing litigation over this matter.

Thus far, EPA has not encountered epidemiological data of sufficient quality to support quantitative risk assessment of conventional pesticide chemicals. Before EPA decides how to use the epidemiological data on chlorpyrifos, we believe it is critical to attempt to resolve questions about these studies regarding the extent of the cohort members' exposures to chlorpyrifos, as well as the impact of exposure to other compounds capable of causing or contributing to the observed neurological outcomes. We acknowledge the lengthy conduct of our assessment, including multiple SAP reviews, but we believe the deliberate and considered approach we are taking is the most scientifically defensible method for re-evaluating our current approach to assessing risks from chlorpyrifos and other organophosphorous pesticides generally, and, specifically, for evaluating the strengths and weaknesses of the epidemiological data.

The July 2012 SAP report is in accord with EPA's assessment that the Agency should attempt to resolve certain key questions about the epidemiological data. Specifically, the SAP recommended that EPA pursue a number of possible approaches for attempting to resolve whether the neurological outcomes observed in the studies occurred in the absence of AChE inhibition – the effect EPA's current regulatory approach is designed to preclude. Further, given that the women and children studied in the Columbia University-sponsored epidemiology study[9] were exposed to multiple chemicals (including other pesticides, polycyclic aromatic hydrocarbons and lead), the SAP cautioned the agency about attributing the outcomes to a single chemical based on the current analysis conducted by Columbia University researchers. These statements by the SAP lead the agency to believe that we need to further explore the extent to

[8] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2012-0040-0029

[9] Rauh, V., Arunajadai, S., Horton, M., Perera, F., Hoepner, L., Barr, D. B., & Whyatt, R. (2011). Seven-year neurodevelopmental scores and prenatal exposure to chlorpyrifos, a common agricultural pesticide. Environ Health Perspect, 119(8), 1196-1201. doi: 10.1289/ehp.1003160; Rauh, V. A., Garfinkel, R., Perera, F. P., Andrews, H. F., Hoepner, L., Barr, D. B., Whyatt, R. W. (2006). Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics, 118(6), e1845-1859. doi: 10.1542/peds.2006-0338.

which the observed neurological outcomes were influenced by exposure to these other chemicals.

Following receipt of the report EPA began conducting a number of analyses to address these recommendations. As I indicated in our December response, we are making progress in conducting a dose-reconstruction analysis of potential exposures to the women and children studied in the Columbia University-sponsored epidemiology study[10] in order to assess the degree to which the individuals in the cohort may or may not have been exposed to chlorpyrifos levels high enough to cause AChE inhibition. In addition to this assessment, to address the SAP recommendations EPA also intends in the coming months to complete an evaluation of cohort exposures to other chemicals. In order to complete both the dose reconstruction and analyses on other chemical exposures, however, we will need to analyze the original data ("raw data") from the Columbia University study to better understand the exposure to chlorpyrifos and other chemicals. To date, the study authors have declined our request to provide that information to us, but we are continuing to discuss our need for evaluating these data with the study authors and we are hopeful that a resolution can be reached.

In addition to further analysis of the exposures in the Columbia study, EPA has also followed up on a recommendation that was brought up in the SAP's oral deliberations regarding the administration and interpretation of diagnostic and analytic tools used to assess neuro and motor development in children like those used in the Columbia study. The SAP noted that it lacked expertise in evaluating these aspects of the data. Because this expertise is relevant in assessing the potential for effects from exposures to other chemicals, between August and October 2012, we obtained additional peer review from scientists within the federal government who have expertise in this field. EPA will include consideration of the results of this peer review when it completes its assessment, as further discussed below.

Finally, as our previous response indicated, last fall, the Columbia University researchers published a new epidemiology study[11] describing the results of magnetic resonance imaging on a subset of children in the cohort. We solicited comments between August 2012 and October 2012, from scientists within the federal government who have expertise in this scientific area and are currently evaluating this input to determine the extent to which this information informs the earlier Columbia University study results.

In light of our ongoing work described above, we are not in a position to provide you with our conclusions on the three remaining toxicology issues in the petition at this time, and it is difficult to provide a precise time frame for the completion of that assessment. It is our hope that we can maintain our current schedule to complete the full chlorpyrifos HHRA by the end of 2013

[10] Rauh, V., Arunajadai, S., Horton, M., Perera, F., Hoepner, L., Barr, D. B., & Whyatt, R. (2011). Seven-year neurodevelopmental scores and prenatal exposure to chlorpyrifos, a common agricultural pesticide. Environ Health Perspect, 119(8), 1196-1201. doi: 10.1289/ehp.1003160; Rauh, V. A., Garfinkel, R., Perera, F. P., Andrews, H. F., Hoepner, L., Barr, D. B., Whyatt, R. W. (2006). Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics, 118(6), e1845-1859. doi: 10.1542/peds.2006-0338.

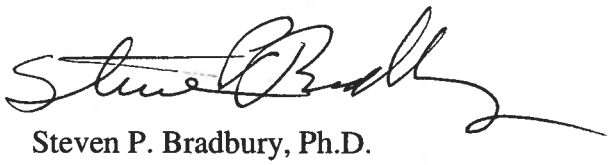
[11] Rauh VA, Perera FP, Horton MK, Whyatt RM, Bansal R, Hao X, Liu J, Barr DB, Slotkin TA, Peterson BS. Brain anomalies in children exposed prenatally to a common organophosphate pesticide. Proc Natl Acad Sci U S A. 2012 May 15;109(20):7871-6. doi: 10.1073/pnas.1203396109. Epub 2012 Apr 30. PubMed PMID: 22547821; PubMed Central PMCID: PMC3356641.

and respond to the remaining claims in your petition on the same time frame. As we previously explained to you, that schedule would result in our initiating any necessary regulatory action in early 2014. Given the complexity of the assessment, and in particular, the complications we are having in obtaining potentially important research data from the Columbia University study authors, I do have some concern about our ability to meet that time frame, but we will continue to work to meet that goal and will update you if our plans must change.

With that said, we have made significant progress in addressing the volatilization portion of your inhalation claim as will be evident with the release of the preliminary chlorpyrifos volatilization assessment in February. As noted, if, following review of the public comments, EPA determines that the risk posed from chlorpyrifos volatilization merits regulatory action in advance of the completion of the HHRA, we will initiate that action without first completing the entire HHRA.

Finally, I wish to reiterate that for efficiency purposes EPA does not intend to proceed with issuing a denial order of the six petition issues (the spray drift portion of your inhalation claim was granted) that we rejected in July 2012 until after we complete our review of all remaining issues. It has been our understanding that this approach is preferable to you as well. However, as previously indicated, if you wish to begin the objections process for the six denied claims and notify EPA in writing, we will publish a formal denial order for those claims, triggering your right to file objections under FFDCA section 408(g)(2).

Sincerely



Steven P. Bradbury, Ph.D.
Director, Office of Pesticide Programs

EXHIBIT 9

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF CHEMICAL SAFETY
AND POLLUTION PREVENTION

JUL 15 2014

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Re: Chlorpyrifos petition dated September 12, 2007; July 2014 Response

Dear Mr. Colangelo and Dr. Reeves:

I am writing to further respond and update you on the U.S. Environmental Protection Agency's (EPA) continued efforts to address the Natural Resources Defense Council (NRDC) and Pesticide Action Network North America (PANNA) jointly submitted September 12, 2007[1] petition, and our related efforts to complete the registration review of chlorpyrifos. The petition specifically requested that EPA revoke all tolerances and cancel all registrations for the insecticide chlorpyrifos.

We have made significant progress in completing our work on the remaining four petition issues that we did not address in our July 16, 2012[2] partial response to your petition. Today we are responding to your claim that EPA failed to incorporate inhalation routes of exposure from chlorpyrifos volatilization. This will complete our response to your broader claim that EPA failed to incorporate inhalation routes of exposure since we have already addressed the primary spray drift portion of this claim in our July 16, 2012 response. At that time, as you may recall, in conjunction with our partial petition response, we released our *Evaluation of the Potential Risks From Spray Drift and the Impacts of Potential Risk Reduction Measures*[3] along with spray drift mitigation that the chlorpyrifos registrants agreed to implement. The label mitigation (in the form of rate reductions and spray drift buffers) reduces risks to bystanders, particularly children, from

[1] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0005.
[2] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0095.
[3] Available at http://www.regulations.gov/#!docketDetail;D=EPA-HQ-OPP-2008-0850.

spray drift. We approved those changes for all 41 chlorpyrifos agricultural products subject to these use limitations by the end of 2012.

At this time, work still remains to be completed in order to respond to the other three remaining claims in your petition: (1) EPA failed to incorporate into its risk assessment, in a quantitative manner, data indicating that long-lasting effects in children result from early life exposure to chlorpyrifos; (2) EPA disregarded data demonstrating that there is no evidence of a safe level of exposure during pre-birth and early life stages; and (3) EPA failed to cite or quantitatively incorporate studies and clinical reports suggesting potential adverse effects below 10% cholinesterase inhibition. In addition to addressing your claim regarding chlorpyrifos volatilization, our response today will outline the approach we are taking for completing our work on those remaining petition issues and our anticipated schedule for releasing a revised human health risk assessment (RHHRA) for public comment, which informs our petition response on the remaining three issues. The RHHRA will also address risks from human dietary exposure resulting from drinking water. Although drinking water exposures were not a part of your petition, we believe that for purposes of efficiency and completeness, our work on the RHHRA, which is currently scheduled to be issued for comment in December 2014 and our response to your petition (i.e., either a proposed revocation rule or a proposed order denying your petition) will be on a similar schedule.

Inhalation Exposure from Volatilization

As you are aware, in February 2013 we released our *Chlorpyrifos: Preliminary Evaluation of the Potential Risks from Volatilization*[4] for chlorpyrifos for public comment. The assessment evaluated the potential risks to bystanders, those who live and/or work in proximity to treated fields, from inhalation exposure to vapor phase chlorpyrifos and chlorpyrifos-oxon emitted from fields following application of chlorpyrifos. The assessment was significant because it was the first probabilistic assessment of the risks posed by the post-application volatilization of a semi-volatile pesticide. The methodology used reflected the approach we employed for the fumigant pesticides where we assessed bystander inhalation risks from volatilization, the key exposure pathway for fumigants.[5] In addition, it was consistent with the recommendations from the December 2009 Federal Insecticide, Fungicide, and Rodenticide Act Scientific Advisory Panel (SAP)[6] meeting on the scientific issues associated with field volatilization of conventional (semi-volatile) pesticides. It also is similar to the methodology that was used to develop our recent Draft Human Health Bystander Screening Level Analysis: Volatilization of Conventional Pesticides that was released for public comment on March 28, 2014.[7]

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[4] R. Bohaty, C. Peck, A. Lowit, W. Britton, N. Mallampalli, and A. Grube. Chlorpyrifos: Preliminary Evaluation of the Potential Risks from Volatilization. 1/31/2013. U.S. EPA Office of Chemical Safety and Pollution Prevention. Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0114.

[5] The assessments can be found in the dockets for each fumigant. Four of which are provided here chloropicrin - EPA-HQ-OPP-2007-0350; dazomet - EPA-HQ-OPP-2005-0128; metam sodium/potassium - EPA-HQ-OPP-2005-0125; and methyl bromide - EPA-HQ-OPP-2005-0123.

[6] U.S. EPA 2009. FIFRA Science Advisory Panel Meeting Minutes - Scientific Issues Associated with Field Volatilization of Conventional Pesticides. Available at http://www.epa.gov/scipoly/sap/meetings/2009/december/120309meetingminutes.pdf.

[7] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2014-0219-0002.

The results of the preliminary assessment released in February 2013 indicated that offsite concentrations of volatilizing chlorpyrifos and chlorpyrifos-oxon may exceed the target concentration based on the toxicological endpoints used. It is important to understand, however, that the chlorpyrifos air concentration used in that assessment was derived from a study[8] that measured the effects of aerosolized chlorpyrifos, which is the form chlorpyrifos takes when applied as a spray – not the form it takes (vapor) when it volatilizes after application. As a result, we noted in the preliminary assessment that there was a significant uncertainty about the relevance of an aerosol toxicity study to human exposures to volatilized chlorpyrifos. Although it is well accepted that bystander exposures will be to vapor rather than aerosol and, as such, that vapor is the most relevant form for evaluating potential bystander risks from chlorpyrifos volatilization, we lacked toxicity data based on vapor administration to test animals for chlorpyrifos and the chlorpyrifos-oxon at the time of the 2013 preliminary volatilization assessment.

Since the release of the preliminary volatilization assessment, Dow AgroSciences LLC conducted two high quality nose-only rat vapor phase inhalation studies for both chlorpyrifos and chlorpyrifos-oxon[9] to address this uncertainty. These studies, as indicated in the attached memorandum *Chlorpyrifos: Reevaluation of the Potential Risks from Volatilization in Consideration of Chlorpyrifos Parent and Oxon Vapor Inhalation Toxicity Studies*[10], have significantly altered our analysis of the hazards resulting from chlorpyrifos and chlorpyrifos-oxon vapor. Both studies evaluated the effects of vapor forms of chlorpyrifos and its oxon at 100% air saturation – in other words, the maximum amount that the air can hold and therefore the maximum possible exposure level. Even at the point of air saturation, however, these studies revealed that concentrations of the vapor forms are much lower than the levels seen in the earlier aerosol study. Indeed, no cholinesterase inhibition was observed in either volatility study. What is clear from these data is that the air cannot hold levels of volatilized chlorpyrifos or its oxon that are capable of causing adverse effects from cholinesterase inhibition – the end point of concern for chlorpyrifos. Accordingly, based on the new data, we no longer anticipate that there are human health risks of concern from exposure to the volatilization of either chlorpyrifos or chlorpyrifos-oxon. As such, EPA therefore intends to deny the volatilization component of your inhalation exposure claim when we publish our complete response to the petition in the Federal Register.

For efficiency purposes we do not intend to proceed with issuing a denial order of the six petition issues that we responded to in July 2012 and the volatilization portion of your inhalation exposure issue (the spray drift portion of your inhalation claim was granted) addressed today until after we complete our review of all remaining issues. Our understanding is that this approach is preferable to you as well. However, if you wish to begin the objections process for

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[8] EPA MRID 48139303: Acute Inhalation Exposure of Adult Crl:CD(SD) rates to particulate chlorpyrifos aerosols: Kinetics of Concentration-Dependent Cholinesterase (CHE) Inhibition in Red Blood Cells, Plasma, Brain and Lung; Authors: J. A. Hotchkiss, S. M. Krieger, K. A. Brzak, and D. L. Rick; Sponsor: Dow AgroSciences LLC.

[9] W. Irwin. *Review of Parent Vapor Study Title. Date. U.S. EPA Office of Chemical Safety and Pollution Prevention. EPA. 6/25/14. D411959.*

W. Irwin. *Review of Oxon Vapor Study Title. Date. U.S. EPA Office of Chemical Safety and Pollution Prevention. EPA. 6/25/14. D415447.*

[10] Also available at www.regulations.gov in docket EPA-HQ-OPP-2008-0850.

the denied claims and notify EPA in writing, we will publish a formal denial order for those claims, triggering your right to file objections under FFDCA section 408(g)(2).

Remaining Work

In our January 2013 letter[11] updating you on the status of our efforts to respond to your remaining petition issues, we indicated that we were pursuing a number of avenues that would eventually inform our response. One such effort was to solicit comments, which occurred late in the summer of 2012, from scientists within the federal government who have expertise in the areas of (1) the assessment of neurodevelopmental outcomes and (2) the use of magnetic resonance imaging. This peer review, *Federal Letter-Review of Chlorpyrifos Epidemiology Studies*[12], which contains a list of the Federal reviewers; the questions we asked the reviewers to consider during their review of the Rauh et al. study; our brief synopsis of Rauh et al. 2012 and our evaluation of the epidemiological aspects of the MRI study; the letters sent to each reviewer soliciting their input; and their review can be found in the chlorpyrifos registration review docket[13]. As we indicated previously, the results of the peer review will be considered when completing our RHHRA.

Coinciding with all of our work addressing your petition issues is our work on the RHHRA. I wanted to inform you of recent developments on that front. This past winter the EPA received a multi-route physiologically based pharmacokinetic and pharmacodynamic (PBPK/PD) model for chlorpyrifos and chlorpyrifos-oxon from Dow AgroSciences LLC. We have held several Federal Insecticide, Fungicide, and Rodenticide Act Scientific Advisory Panels[14] on the relevance and usefulness that a PBPK/PD model can provide in assessing a chemical's risks and two SAPs specifically on PBPK/PD and chlorpyrifos[15]. This PBPK/PD model was a decade in the making and can assess oral, dermal, and inhalation routes of exposure in both rats and humans. We have conducted a quality assurance (QA) assessment of the model. The QA assessment focused on ensuring that the model's structure and parameter values were accurately reflected and implemented in the computer code, as well as determining whether human data can be reasonably simulated by the model. Based upon our assessment this is a robust model. Instead of the use of standard defaults, the PBPK/PD model refines the extrapolation from animals to humans and across the human population. Additionally, the model will be useful for informing

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[11] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2007-1005-0097.
[12] Available at http://www.regulations.gov/#!documentDetail;D=EPA-HQ-OPP-2008-0850-0170.
[13] EPA-HQ-OPP-2008-0850 available at www.regulations.gov.
[14] U.S. EPA, December 11 - 12, 2003: Physiologically-Based Pharmacokinetic/Pharmacodynamic Modeling: Preliminary Evaluation and Case Study for the N-Methyl Carbamate Pesticides: A Consultation, available at http://www.epa.gov/scipoly/sap/meetings/2003/121103_mtg.htm; U.S. EPA, December 2, 2004: Use of Pharmacokinetic Data to Refine Carbaryl Risk Estimates from Oral and Dermal Exposure, available at http://www.epa.gov/scipoly/sap/meetings/2004/120204_mtg.htm; U.S. EPA, December 3, 2004: The N-methyl Carbamate Cumulative Risk Assessment: Strategies and Methodologies for Exposure Assessment, available at http://www.epa.gov/scipoly/sap/meetings/2004/120304_mtg.htm; and U.S. EPA, August 16 - 17, 2007: Assessing Approaches for the Development of PBPK Models of Pyrethroid Pesticides, available at http://www.epa.gov/scipoly/sap/meetings/2007/081607_mtg.htm.
[15] U.S. EPA, February 15-18, 2011: Chlorpyrifos Physiologically-Based Pharmacokinetic/Pharmacodynamic (PBPK/PD) Modeling linked to the Cumulative and Aggregate Risk Evaluation System (CARES) available at http://www.epa.gov/scipoly/sap/meetings/2011/021511meeting.html.

our dose reconstruction work with respect to the epidemiological data, which is one of the items we have been working to address as it relates to your remaining petition issues.

In conjunction with our work on the RHHRA, we have worked over the last several months and continue to work with the technical and end use registrants to clarify application rates and uses that currently appear on labels. While this effort is designed primarily to ensure that labels reflect actual use, this effort should also help to preclude the overuse of chlorpyrifos in situations where the labeling may not clearly indicate application rates and uses on each crop. This effort will also help reduce uncertainties that could occur in the risk assessment resulting from unclear label language.

Finally, while we continue to make progress on your remaining three petition issues that deal with chlorpyrifos toxicity, for reasons noted above, we believe that a final response to your petition should correspond with completion of the RHHRA, which also addresses risks from human dietary exposure resulting from drinking water. We anticipate responding to your remaining three petition issues in connection with taking public comment on the RHHRA for chlorpyrifos by issuing either a proposed denial order or a proposed rule revoking tolerances, depending on the nature of the response. We anticipate releasing the RHHRA for public comment in December 2014 and would expect to issue a proposed denial order or a proposed revocation rule in the same time frame, accounting for any additional time that may be necessary to address procedural obligations that accompany Federal Register publication. As we have discussed with your counsel, the issuance of a proposed revocation rule constitutes a response to your petition. If the Agency determines that the petition should be denied following comments on the RHHRA and any proposed denial order, EPA would expect to publish a final denial order by the summer of 2015.

Sincerely

Jack E. Housenger
Director, Office of Pesticide Programs

No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

DECLARATION OF JENNIFER SASS, Ph.D., IN SUPPORT OF SECOND PETITION FOR A WRIT OF MANDAMUS

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I, Jennifer Sass, declare and state as follows:

1. I am a Senior Scientist for Petitioner Natural Resources Defense Council ("NRDC").

2. On April 11, 2012, I submitted a declaration in support of the first petition for writ of mandamus challenging the U.S. Environmental Protection Agency's ("EPA") unreasonable delay in responding to the petition of NRDC and Pesticide Action Network North America ("PANNA"), which requested that EPA ban the use of chlorpyrifos and revoke all tolerances for chlorpyrifos. That mandamus action was *In re Pesticide Action Network North America and Natural Resources Defense Council*, No. 12-71125, and I have attached my declaration in that case to this declaration as Attachment 1.

3. I have reviewed my previous statements to the Court in that matter and reaffirm them here. All information in my prior declaration remains true and accurate, and I incorporate all of that declaration as if set forth fully here. My prior statements apply with equal force to this renewed petition for writ of mandamus.

4. Since filing that declaration, I have remained in my capacity as Senior Scientist for NRDC and have closely followed EPA's work in responding to the 2007 Petition, and ongoing research in the study of chlorpyrifos and its harms in the greater scientific community.

5. As I discussed in my previous declaration, in 2001, EPA completed the chlorpyrifos aggregate assessment, known as the Interim Reregistration Eligibility Decision ("IRED"), which revised, but retained, many of the pre-existing food tolerances (allowable residue limits on food). NRDC submitted comments challenging the scientific limitations of the chlorpyrifos IRED and identifying evidence of harm. I have attached NRDC's comments on the chlorpyrifos IRED as Attachment 2. When EPA completed its cumulative risk assessment for all organophosphates, including chlorpyrifos, it reaffirmed the chlorpyrifos IRED without change, without addressing the significant published studies that had emerged showing risk of harm, and without responding to NRDC's comments.

6. I have included as Attachments 3-4 the comments NRDC prepared on EPA's action to date in response to the 2007 Petition. Notably, EPA has acknowledged the need to account for bystander exposures from drift and volatilization and found concentrations exceeded levels of concern for many applications. EPA also acknowledged the credibility of the new scientific studies of families showing long-lasting effects (possibly permanent and irreversible) on behavior and cognition in children resulting from exposures to chlorpyrifos when they were still in the womb. In addition, EPA estimated drinking water exposures

above safe levels for all infant scenarios. All of these findings underscore EPA's recognition of the danger chlorpyrifos poses and the urgency to act.

7. Since that time, EPA has failed to make a final decision on the 2007 Petition, offering instead responses to some of the rationales and evidence presented in the 2007 Petition. EPA has offered to make its partial responses final as to the rationales addressed, which could allow NRDC and PANNA to file objections and start the administrative review process. However, the 2007 Petition sought a ban on chlorpyrifos, not legal responses to arguments put forward to support such a ban.

8. Even as EPA was developing its approach to determining a way to measure the toxicity of chlorpyrifos and the other organophosphate ("OP") pesticides, it was being alerted in comments by NRDC, other NGO commenters, and its own Scientific Advisory Panel[1] that EPA should not overlook the long-lasting effects from even low exposures, particularly where they occur during critically sensitive life stages.

9. Although EPA eventually settled on the inhibition of brain cholinesterase enzyme in the female adult rat as a way to measure toxicity of chlorpyrifos and the other OP pesticides, a Scientific Advisory Panel in a

[1] The Report of EPA's FIFRA Scientific Advisory Panel Meeting (Sept. 5-6, 2001) ("SAP Report") is available at http://www.epa.gov/scipoly/sap/meetings/2001/september/finalreport.htm.

September 2001 report had previously noted that, "[s]tudies are needed on the sensitivity, specificity, and predictive validity of blood cell AChE [cholinesterase inhibition] in relation to more central health-associated measures, such as brain AChE levels and eventually neurobehavioral competence." SAP Report (emphasis added). The Scientific Advisory Panel was warning EPA that alterations in behavior and cognitive performance (learning, memory, IQ, etc.) may be impaired by chlorpyrifos and other OP pesticides, and it was unclear whether these impacts occurred at or below the exposure levels determined to be "acceptable" based on cholinesterase inhibition in adult rat brains that EPA used to determine a bright line indicator of toxicity.

10. Now those warnings have come true, and in the worst way – with the evidence collected from the children of farmworkers and urban families. When EPA ignores the evidence of harm from laboratory rodent studies, it places the burdens on the backs of those most at risk— those most highly exposed to pesticides such as through work, habits, or geography, and those most vulnerable by age, health status, diet or other circumstances. Now, we have evidence of the harm from chlorpyrifos exposure from the measurable damage it has already caused in people— EPA has already failed its duty to regulate pesticides so as to prevent harm to people, before exposure and harm happen.

11. EPA must evaluate all the scientific evidence, and not parse it into pieces small enough to be dismissed. The epidemiologic studies provide evidence of low-dose chlorpyrifos exposures leading to long-lasting measurable impairments in the behavior and cognitive functioning of children exposed prenatally. These scientific studies must be evaluated along with those measuring cholinesterase inhibition so that EPA may make a scientifically-defensible determination as to whether or not a 10% inhibition of the cholinesterase enzyme is an appropriately protective regulatory endpoint to fully protect vulnerable populations.

12. Because of EPA's now-longer failure to act on the pending petition, NRDC members and their children are still being exposed to unsafe levels of chlorpyrifos and will continue to be as long as the chlorpyrifos registrations and food tolerances challenged in the petition remain in effect.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct to the best of my knowledge. Executed on this 3rd day of September, 2014, at Washington, DC.

Jennifer Sass

JENNIFER SASS

ATTACHMENT 1

No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners.

DECLARATION OF JENNIFER SASS, PH.D, IN SUPPORT OF PETITION FOR A WRIT OF MANDAMUS

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I, Jennifer Sass, declare as follows:

1. I am a Senior Scientist for petitioner Natural Resources Defense Council (NRDC). I have advanced degrees in Anatomy and Cell Biology, with specific expertise in developmental toxicology, neurotoxicology, and molecular biology. In my position with NRDC I am responsible for reviewing the science underlying many of the federal regulations of industrial chemicals and pesticides. I have published over forty articles in peer-reviewed scientific journals, including many pertaining to pesticide hazards and regulations. I have provided testimony to the Environmental Protection Agency (EPA), both written and oral, on the registration of dozens of pesticides during the course of their registration process. I have represented NRDC since 2001 as an active member of the EPA/USDA Pesticide Program Dialogue Committee (PPDC), a stakeholder committee that provides feedback to the EPA Office of Pesticide Programs on various issues related to pesticide regulatory, policy, and program implementation issues. Through my work on the PPDC I have also served on a number of issue-specific PPDC workgroups to provide more in-depth perspectives and advice on pesticide issues. I was also a member of the EPA/USDA Committee to Advise on Reassessment and Transition (CARAT) from 2001 until the committee disbanded in 2003. The purpose of CARAT was to provide advice on strategic approaches for pest management planning, transition, and tolerance reassessment for pesticides

as required by the Food Quality Protection Act. My *Curriculum Vitae* is attached to this declaration as Exhibit A.

2. In this declaration, I provide background about the pesticide chlorpyrifos and the significant human health risks that it poses, particularly to children. I also describe the petition that NRDC and Pesticide Action Network North America (PANNA) submitted to EPA in September of 2007, which requested that EPA ban the use of chlorpyrifos and revoke all tolerances (allowable residue limits on food) for chlorpyrifos.

Background on Chlorpyrifos

3. Chlorpyrifos is one of the most widely used insecticides in the United States. It is used on various food and feed crops, on golf courses, as a non-structural wood treatment, and as an adult mosquitocide. According to the EPA fact sheet that was available in 2007, agriculturally, approximately 10 million pounds are applied annually, with use on corn comprising the largest market (using approximately 5.5 million pounds active ingredient).

4. Chlorpyrifos is an organophosphate pesticide. Organophosphates (also referred to as organophosphorous pesticides or OPs) are a class of chemicals originally developed many decades ago; some were used during World War II as nerve agents. They kill insects by shutting down their nervous system. Unfortunately, for the same reason that they are effective pesticides, OPs can exert

strong adverse effects on the human nervous system. One key nervous system effect of OPs is known as "cholinesterase inhibition," in which the OP interferes with the function of one of the body's natural proteins, an enzyme called cholinesterase. Cholinesterase is necessary to degrade one of the nervous system's key messenger, acetylcholine, in a timely manner. When OPs are in the system, the cholinesterase enzyme cannot do its job to degrade acetylcholine (ACh). Acetylcholine is a neurotransmitter protein that carries messages from the brain and spinal cord out to muscle cells and other cell receptors where it activates skeletal muscles, inhibits heart muscle, and aids in memory formation, learning, attentiveness, and other critical nervous system functions. OP poisoning leads to prolonged over-activation of ACh receptor cells. The result of OP poisoning can be a variety of effects in people depending on the dose and differences in ages, health status, and other factors. Effects include headaches, nausea, dizziness, anxiety, restlessness, muscle twitching, weakness, tremor, poor coordination, confusion, difficulty breathing, vomiting, and diarrhea. At very high exposures, more serious effects such as convulsions, respiratory paralysis, and death have been reported. Repeated exposure at levels too low to cause visible signs of poisoning may cause the same effects, including impaired learning, memory, and concentration. Poisoning can occur through any route of exposure, including inhalation, ingestion, eye contact, and absorption through the skin.

5. Because of the high risk that OP pesticides pose to people, and especially to fetuses, infants, and young children, EPA took effective measures to cancel almost all the residential uses of these pesticides. This resulted in a significant and measurable reduction in poisonings to kids from roach baits, residential foggers or "bug bombs," and other homeowner uses. However, it left rural and farm children at risk from all the agriculture uses that remained. The science underlying chlorpyrifos hazards and remaining risks are detailed in this declaration.

Children are especially sensitive to chlorpyrifos poisoning

6. Infants, toddlers, and young children engage in more frequent hand-to-mouth contact than adults, and therefore have higher rates of oral exposure from pesticide-contaminated objects, dust, or soil. In addition, per pound of body weight, children eat, drink, and breathe more than adults. For example, EPA's Exposure Factors Handbook, just revised in 2011, reports that although the volume of water the average person drinks each day increases with age, when you adjust for body weight the average newborn drinks 52 mg of water per kilogram body weight per day (52 mg/kg-day), whereas a 1 year old drinks half that amount when adjusted for body weight (23 mg/kg-day), and the average adult drinks half that amount again (13 mg/kg-day),[1] EPA's Exposure Factors Handbook also reports

[1] U.S. EPA. Exposure Factors Handbook 2011 Edition (Final). U.S. Environmental

that babies 6-12 months old put their hands to their mouths an average of 19 times/hour, and 5% of babies do it 52 times/hour, whereas adults don't do it at all.[2] These age-related activities mean that infants and young children are much more likely to have greater chlorpyrifos exposures, when adjusted for body weight, than adults.

7. In addition to higher exposures per body weight, infants and children are especially susceptible to chlorpyrifos toxicity, compared with adults. The reasons for this are several, including that children's bodies have immature detoxification mechanisms compared with adults, and that chemical assault during development of critical target organs and systems can cause disruptions that are then hard-wired into the developing system. For example, research on lead and mercury proves that during neural development the nervous system is acutely vulnerable to neurotoxic assault, and exposures may result in long-term or permanent destruction or dysfunction to systems including learning, memory, and intelligence. This is also true for the developing immune system, endocrine system, and reproductive system. For example, doses of lead or mercury with no

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Protection Agency, Washington, DC, EPA/600/R-09/052F, 2011. See Chapter 3 on drinking water. http://cfpub.epa.gov/ncea/risk/recordisplay.cfm?deid=236252.

[2] U.S. EPA. Exposure Factors Handbook 2011 Edition (Final). U.S. Environmental Protection Agency, Washington, DC, EPA/600/R-09/052F, 2011. See Chapter 4 on mouthing frequency. http://cfpub.epa.gov/ncea/risk/recordisplay.cfm?deid=236252.

measurable effect on adults at all have been shown to cause permanent measurable brain damage in exposed children. Data from adult animals often cannot be directly used to predict risks to fetuses, infants, and children.

Rural and farmworker children are a particularly vulnerable population

8. Children of farmworkers and those living in agricultural communities are heavily exposed to pesticides, including chlorpyrifos. Farmworker children come in contact with pesticides through residues on their parents' skin and clothing, contaminated soil in their play areas, pesticide-laden dust tracked into their homes, food eaten directly from the fields, drift from aerial spraying, contaminated well water, and breast milk. Furthermore, these children often accompany their parents to work in the fields or help out by working themselves. Farmworker children are also exposed to pesticides prenatally, when pregnant women are exposed to pesticides during their work.

The Food Quality Protection Act

9. The National Academy of Sciences' landmark report issued in 1993, "Pesticides in the Diets of Infants and Children," urged an overhaul of EPA's pesticide program to assure the safety of children, citing organophosphate insecticides as one of the classes of pesticides of concern.[3] This National Academy

[3] Pesticides in the Diets of Infants and Children (Washington, D.C.: National Academy Press, 1993).

of Sciences study was widely cited as the catalyst for the enactment of the Food Quality Protection Act – commonly known as the FQPA. It was passed unanimously by Congress in 1996, and resulted in a significant overhaul of the pesticide regulatory framework. The FQPA recognized that infants and children were insufficiently protected under preexisting law, and it mandated an additional tenfold safety factor to address these risks.

10. The FQPA requires EPA to evaluate the "cumulative" effects of peoples' exposure to all pesticides that have a common mechanism of toxicity, when setting a "tolerance" (or maximum legally allowable limit for a pesticide on food), so the agency has spent several years developing a "cumulative risk assessment" for the OPs. Because the organophosphates all attack the human nervous system in essentially the same way, EPA has determined that they share a "common mechanism of toxicity," as described in the FQPA.

The 2007 Petition to EPA

11. In 2007, I was one of the coauthors of a document that was submitted to EPA entitled "Petition to Revoke All Tolerances and Cancel All Registrations for the Pesticide Chlorpyrifos" (the 2007 Petition). A true and correct copy of the 2007 Petition is attached to this Declaration as Exhibit B. That document was submitted to EPA on behalf of PANNA and NRDC on September 12, 2007. This declaration summarizes and highlights the scientific evidence that was submitted to

EPA in the 2007 Petition. In particular, it focuses on scientific evidence of the long lasting effects to children from early life chlorpyrifos exposure.

12. The 2007 Petition contained a robust body of scientific information laying out the human health risks associated with chlorpyrifos, and those risks particular to children and infants, which is sufficient to justify EPA revoking all tolerances and cancelling all registrations for chlorpyrifos. Scientific evidence that has emerged since the 2007 Petition was submitted further supports the revocation of all tolerances and cancellation of all registrations for chlorpyrifos.

13. The state of the science identifying many various adverse health effects associated with dietary exposure to chlorpyrifos supports a ban on chlorpyrifos and revocation of all food tolerances. Below, I summarize the overwhelming scientific evidence that was submitted to EPA in the 2007 Petition, which demonstrates that chlorpyrifos is too dangerous to be re-registered for food uses.

14. It is my understanding that EPA has not yet responded in writing to that request.

EPA's Previous Evaluations of Chlorpyrifos

15. In 2001, EPA completed the chlorpyrifos aggregate assessment, called an Interim Reregistration Eligibility Decision (IRED), which revised, but retained,

many of the pre-existing food tolerances (allowable residue limits on food).[4] In its 2002 comments on the IRED (Docket ID No. OPP-34203G), NRDC challenged the scientific limitations of the IRED, identified evidence of harm, and highlighted that there is inadequate evidence to establish a safe level at which infants and children will not suffer any developmental harm due to chlorpyrifos exposure. EPA never responded directly to NRDC's comments or other comments submitted by other public interest advocates, including PANNA and the New York Attorney General.

16. In 2006, EPA completed the cumulative risk assessment (CRA) for all organophosphates, including chlorpyrifos, and reaffirmed the chlorpyrifos IRED without change, despite new, significant published studies that had emerged during this time showing harm. Without addressing the comments by NRDC and other public interest advocates and without referencing much of the data that had been available since 2001, the Agency concluded that chlorpyrifos uses would be eligible for reregistration and that the current pesticide tolerances met the legal safety standard.[5]

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[4] 66 Fed. Reg. 57073 (Nov. 14, 2001) Organophosphate Pesticide; Availability of Chlorpyrifos Interim Risk Management Decision Document. IRED at 64-68.

[5] Memo from Debra Edwards to Jim Jones, re: Finalization of Interim Reregistration Eligibility Decisions (IREDs) and Interim Tolerance Reassessment and Risk Management Decisions (TREDs) for the Organophosphate Pesticides, and Completion of the Tolerance Reassessment and Reregistration Eligibility

17. Scientific evidence that has emerged since 2001, when the chlorpyrifos IRED was published, reinforces the earlier science showing that exposure to chlorpyrifos causes many adverse health effects. In fact, both the weaknesses in the studies relied on by EPA in the IRED and the failure to incorporate evidentiary science since 2001 undermine EPA's decision to re-register chlorpyrifos and retain its tolerances. In the 2007 Petition we summarized the pre-2001 data and identified post-2001 scientific evidence relevant to the risk assessment of chlorpyrifos. That evidence is described in more depth in the 2007 Petition. See generally Exhibit B.

18. The assessment of the health effects associated with particular pesticides includes both an aggregate assessment which analyzes the risk from multiple routes of exposures (food, water, residential uses) to a single pesticide, and a cumulative assessment which analyzes the risk from cumulative exposure to a class of pesticides that share a common mode of action. The Agency grouped chlorpyrifos with the other organophosphates to conduct its cumulative risk assessment. For the organophosphate cumulative assessment, EPA used the endpoint of cholinesterase inhibition in the rat brain to determine an acceptable maximum level of cumulative exposure to organophosphate pesticides by the oral, dermal, and inhalation route of exposure. The toxic potency of each

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Process for the Organophosphate Pesticides, July 31, 2006.

organophosphate pesticide is determined separately for each route of exposure, based on the central estimate of an oral dose that is associated with a 10% inhibition of acetylcholinesterase activity (10% AChEi), compared to control animals (not treated with pesticide), or a 15% inhibition for the dermal and inhalation routes of exposure. This is called a benchmark dose 10, or BMD10, for the oral route of exposure, and a BMD15 for the dermal and inhalation routes of exposure.[6]

Long Lasting Effects from Early Life Exposure in Children

19. The more we learn about chlorpyrifos, the more we understand its damaging effects from exposures that take place during fetal development, infancy, and childhood. Many studies published since 2001 report that chlorpyrifos is more damaging when exposures take place during prenatal or early childhood development.[7] Columbia University researchers published two studies from the

[6] Both the relative potency factors (RPF) and the POD/BMD10 were from the female rat brain ChE inhibition measurements. See Organophosphorus Cumulative Risk Assessment 2006 EPA (Jul. 31, 2006) at 42-52. Available at: http://epa.gov/oppsrrd1/cumulative/2006-op/op_cra_main.pdf.

[7] Rauh VA, Garfinkel R, Perera FP, Andrews HF, Hoepner L, Barr DB, Whitehead R, Tang D, Whyatt RW. Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics. 2006 Dec;118(6):e1845-59. Epub 2006 Nov 20.; Perera FP, Rauh V, Whyatt RM, Tang D, Tsai WY, Bernert JT, Tu YH, Andrews H, Barr DB, Camann DE, Diaz D, Dietrich J, Reyes A, Kinney PL. A summary of recent findings on birth outcomes and developmental effects of prenatal ETS, PAH, and pesticide exposures. Neurotoxicology. 2005 Aug;26(4):573-87. Review.; Whyatt RM, Rauh V, Barr

same New York City cohort of infants born to African American and Dominican women. The first study, in 2004, reported on the effects of chlorpyrifos on birth outcomes.[8] The second, published in 2006, report on child development effects.[9] Because the cohort of babies were enrolled over a number of years, the study captures changes in exposure levels related to the 2000-2001 ban of chlorpyrifos for residential use. Decreases in birth weight and length were associated with prenatal exposures to chlorpyrifos, measured in cord blood, and the follow-up study of the same children when they reached age 3 showed that children that had been exposed prenatally to the highest levels (chlorpyrifos levels of > 6.17 pg/g plasma) were significantly more likely to experience delays in cognitive and psychomotor development as well as attention problems, attention-deficit/hyperactivity disorder problems, and pervasive developmental disorder

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DB, Camann DE, Andrews HF, Garfinkel R, Hoepner LA, Diaz D, Dietrich J, Reyes A, Tang D, Kinney PL, Perera FP. Prenatal insecticide exposures and birth weight and length among an urban minority cohort. Environ Health Perspect. 2004 Jul;112(10):1125-32; Perera FP, Rauh V, Tsai WY, Kinney P, Camann D, Barr D, Bernert T, Garfinkel R, Tu YH, Diaz D, Dietrich J, Whyatt RM. Effects of transplacental exposure to environmental pollutants on birth outcomes in a multiethnic population. Environ Health Perspect. 2003 Feb;111(2):201-5.

[8] Whyatt RM, Rauh V, Barr DB, Camann DE, Andrews HF, Garfinkel R, Hoepner LA, Diaz D, Dietrich J, Reyes A, Tang D, Kinney PL, Perera FP. Prenatal insecticide exposures and birth weight and length among an urban minority cohort. Environ Health Perspect. 2004 Jul;112(10):1125-32.

[9] Rauh VA, Garfinkel R, Perera FP, Andrews HF, Hoepner L, Barr DB, Whitehead R, Tang D, Whyatt RW. Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics. 2006 Dec;118(6):e1845-59. Epub 2006 Nov.

problems. The authors report that "the proportion of delayed children in the high-exposure group was five times greater for the Psychomotor Development Index and 2.4 times greater for the Mental Development Index, increasing the number of children possibly needing early intervention services."[10] The adverse effects on birth outcomes were no longer observed among the children in the cohort who were born after the ban took effect (January 2001) and their cord blood levels of chlorpyrifos were significantly lower, underscoring the benefits of the ban. These data provide strong evidence that prenatal and early-life stage exposure to chlorpyrifos is associated with not only poor birth outcomes (lower birth weight and length), but also long-lasting, and possibly permanent, impaired cognitive development.

20. In 2011, three important studies were published in the peer-reviewed scientific literature that each reported on the long-lasting deficits in learning and IQ associated with prenatal exposure to OP pesticides generally and chlorpyrifos in particular. Each of the three studies was in a different population of children, and was conducted by a different set of academic researchers, making the concordance in results very convincing. The Mount Sinai Children's Environmental Health study reported poor cognitive development associated with prenatal organophosphate exposure (measured in mother's urine during third trimester

[10] Id.

pregnancy) measured at 1-2 years old and again at 6-9 years old among 400 New York City children.[11] The Center for the Health Assessment of Mothers and Children of Salinas (CHAMACOS) study also reported that prenatal exposure to OP pesticides (measured in mother's urine collected during pregnancy) was associated with measurably poorer intellectual development in 329 children tested at 7 years of age.[12] The third study examined 265 inner-city mothers and children enrolled in the Columbia Center for Children's Environmental Health cohort and reported evidence of deficits in memory and IQ at ages 3 and 7 years old that were specifically associated with chlorpyrifos (measured in umbilical cord blood). The authors express concern in their conclusions that chlorpyrifos continues to be of widespread use in agricultural settings. All three studies have important implications for children's long-term educational abilities and social and economic potential.[13] Moreover, the neurodevelopmental (brain development) outcomes reported for the children in these studies are similar to the reported results from

[11] Engel SM, Wetmur J, Chen J, Zhu C, Barr DB, Canfield RL, Wolff MS. Prenatal exposure to organophosphates, paraoxonase 1, and cognitive development in childhood. Environ Health Perspect. 2011 Aug;119(8):1182-8.

[12] Bouchard MF, Chevrier J, Harley KG, Kogut K, Vedar M, Calderon N, Trujillo C, Johnson C, Bradman A, Barr DB, Eskenazi B. Prenatal exposure to organophosphate pesticides and IQ in 7-year-old children. Environ Health Perspect. 2011 Aug;119(8):1189-95.

[13] Rauh V, Arunajadai S, Horton M, Perera F, Hoepner L, Barr DB, Whyatt R. Seven-year neurodevelopmental scores and prenatal exposure to chlorpyrifos, a common agricultural pesticide. Environ Health Perspect. 2011 Aug;119(8):1196-201.

animal tests, providing not only concordance across several human cohorts, but also across species. EPA's own Scientific Advisory Panel (SAP) concluded that the results of the three cohort studies published in 2011 and the animal studies together indicate that "maternal chlorpyrifos exposure would likely be associated with adverse neurodevelopmental outcomes in humans."[14] These epidemiologic studies, and particularly the CHAMACOS study of rural and agriculture populations, provide real-world evidence in human populations that EPA's current restrictions on agriculture uses of chlorpyrifos are not working. The evidence from the epidemiology shows that prenatal exposures to chlorpyrifos and other harmful OPs are not only occurring, but are occurring at levels that are causing permanent and severe effects in exposed populations.

Chlorpyrifos Is Unsafe and EPA Should Revoke All Tolerances and Cancel All Registrations for It

21. As a result of EPA's failure to act on the pending petition, NRDC members and their children are being exposed to unsafe levels of chlorpyrifos, and will continue to be as long as the chlorpyrifos registrations and food tolerances challenged in the petition remain in effect.

[14] EPA Chlorpyrifos Preliminary Human Health Risk Assessment for Registration Review. June, 2011. EPA-HQ-OPP-2008-0850-0025. P. 33.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct, to the best of my knowledge. Executed this 11th day of April, 2012, in Washington, D.C.

s/ Jennifer Sass
JENNIFER SASS, PH.D.

EXHIBIT A

Jennifer Beth Sass, Ph.D.
Natural Resources Defense Council, Senior Scientist
George Washington University, Professorial Lecturer

Full-time Employment (January 2001 to present):
Senior Scientist, Natural Resources Defense Council
1200 New York Ave, NW, Suite 400
Washington, DC. USA 20005
Tel: 202-289-6868 (main), 202-289-2362 (direct)
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Academic Affiliation (2008 to present)
Professorial lecturer
George Washington University
School of Public Health and Health Services
Department of Environmental and Occupational Health
Washington, DC. USA 20052

Short Professional Biography :

I am a Senior Scientist in the Health and Environment program of the NRDC, an environmental non-profit organization. I review U.S. government regulations of industrial chemicals and pesticides, and assess the data underlying the regulatory decisions. I am well versed in the health sciences, with degrees in Anatomy and Cell Biology, and Toxicology, and have published over three dozen articles in peer-reviewed journals. In my work with NRDC I review the science underpinning the regulation of toxic chemicals and emerging contaminants such as nanomaterials, and advocate for health-protective regulations consistent with the environmental statutes. I provide testimony and scientific briefings for the U.S. Congress and regularly participate in stakeholder and expert scientific federal advisory committees.

Education:

- Post-Doctoral Fellow, 1998-2000. Program in Human Health and the Environment, University of Maryland, Baltimore. Mentor: EK Silbergeld.

- Ph.D. 1998. Dept. of Anatomy and Cell Biology, University of Saskatchewan, Canada. Thesis title: Heat-inducible and Constitutive Expression of the 90kD Heat Shock Protein Gene, Hsp90, During Zebrafish Embryogenesis. Mentor: PH Krone.

- MSc. 1993. Dept. of Anatomy and Cell Biology, University of Saskatchewan, Saskatoon, Canada. Thesis title: Aluminum Pretreatment Impairs the Ability of Astrocytes to Protect Neurons from Glutamate Toxicity. Mentor: BHJ Juurlink.

- B.Sc. Advanced. 1989. Anatomy. University of Saskatchewan, Saskatoon, Canada.

Publications:

1. Sass, J. Putting the public into alternatives assessment. Environmental Health Policy Institute. Physicians for Social Responsibility. Online December 16, 2010. http://www.psr.org/environment-and-health/environmental-health-policy-institute/responses/putting-the-public-into-alternatives-assessment.html

2. Joshi TK, Bailar JC 3rd, Craner J, Davis D, Ehrlich R, Franco G, Frank AL, Huff J, LaDou J, Lanphear B, London L, Melnick RL, O'Neill R, Osaro E, Rosenman KD, Sass J, Smith AH, Soskolne CL, Stephens C, Stuckey R, Takaro TK, Teiteibaum D, Watterson A, Yassi A. Physician expelled from Indian Association of Occupational Health after critique. Int J Occup Environ Health. 2009 Oct-Dec;15(4):419-20.

3. Sass, JB. Supporting the need for rigorous enforceable disclosure policies for scientific journals. Commentary. Addiction 2009 Nov;104(11):1788-9.

4. Sass, JB. Key elements of effective and practical disclosure policies for health science journals. Editorial. Env Health Perspect, 2009 June;117(6):A233.

5. Sass, JB, Musu T, Burns K, Illuminato I. Nanomaterials: Brief review of policy frameworks in the U.S. and Europe and recommendations from an occupational and environmental perspective. European J Oncol, 2009; 13(4):211-218.

6. Sass, JB. Invited book review of Bending Science: How special interests corrupt public health research. T.O. McGarity and W.E. Wagner. Environ Health Perspect. 2008 Nov;116(11):1654-9

7. Sass, JB. Janssen, S. Open letter to Stephen Johnson, Administrator, U.S. Environmental Protection Agency: Ban Endosulfan. Int J Occup Environ Health, 2008, July-Sept; 14(3):236-239.

8. Huff, James and Jennifer Sass. Atrazine – A likely human carcinogen? Int J Occup Environ Health, 2007, July-Sept;13(3):356-358.

9. Balbus JM, Maynard AD, Colvin VL, Castranova V, Daston GP, Denison RA, Dreher KL, Goering PL, Goldberg AM, Kulinowski KM, Monteiro-Riviere NA, Oberdörster G,Omenn GS, Pinkerton KE, Ramos KS, Rest KM, Sass JB, Silbergeld EK, Wong BA. Meeting report: hazard assessment for nanoparticles--report from an interdisciplinary workshop. Environ Health Perspect. 2007 Nov;115(11):1654-9.

10. Guth JH, Denison RA, Sass J. Require comprehensive safety data for all chemicals. New Solut. 2007;17(3):233-58.

11. Sass JB, Wu M. Budget cuts to the U.S. EPA will reduce government data on pollutants, and increase reliance on industry data. Int J Occup Environ Health. 2007 Apr-Jun;13(2):244-6.

12. Sass JB. Recommendations for improved risk assessment approaches. J Hum Ecol Risk Assessment. 2007.

13. Sass, JB, Colangelo A. European Union bans atrazine, while the United States negotiates continued use. Int J Occup Environ Health, 2006 July;12:260-267.

14. Sass J, Simms P, Negin E. Nanotechnologies: The promise and the perils. Sustainable Development Law & Policy (SDLP) journal, 2006, Apr/May:11-16.

15. Sass, J. No small problem: It's high time for the United States to get nanotech regulations – and it needs to get them right. Bull Atom Sci, 2006, Mar/April; 62(2): 21-22

16. Sass, J.B. Credibility of Scientists: conflict of interest and bias. Env Health Perspect, 2006 March; 114(3):A147.

17. Needleman HL, Reigart JR, Landrigan P, Sass J, Bearer C, Resnik DB, Portier C. Correspondence: Benefits and Risks of Pesticide Testing on Humans and author response. Environ Health Perspect. 2005 Dec;113(12):a804-5.

18. Sass, JB. Industry efforts to weaken the EPA's classification of the carcinogenicity of 1,3-butadiene. Int J Occup Environ Health, 2005; 11:378-383.

19. Sass JB, Castleman B, Wallinga D. Vinyl chloride: Sass et al respond. Environ Health Perspect. 2005 Oct;113(10):A654-655.

20. Sass JB, Castleman B, Wallinga D. Vinyl chloride: a case study of data suppression and misrepresentation. Environ Health Perspect. 2005 Jul;113(7):809-12.

21. Sass J, Solomon G. Inappropriate influence by industry on EHP news article. Environ Health Perspect. 2005 Feb;113(2):A87-8.

22. Sass J. U.S. Department of Defense and White House working together to avoid cleanup and liability for perchlorate pollution. Int J Occup Environ Health. 2004 Jul-Sep;10(3):330-4.

23. Sass JB, Needleman HL. Human testing: Sass and Needleman respond to industry. Environ Health Perspect. 2004 May;112(6):A340-1.

24. Sass JB, Needleman HL. Industry testing of toxic pesticides on human subjects concluded "no effect," despite the evidence. Environ Health Perspect. 2004 Mar;112(3):A150-1; author reply A151-2; discussion. A152-6.

25. Sass JB, Devine JP Jr. The Center for Regulatory Effectiveness invokes the Data Quality Act to reject published studies on atrazine toxicity. Environ Health Perspect. 2004 Jan;112(1):A18; author reply A18-9.

26. Axelson O, Balbus JM, Cohen G, Davis D, Donnay A, Doolittle R, Duran BM, Egilman D, Epstein SS, Goldman L, Grandjean P, Hansen ES, Heltne P, Huff J, Infante P, Jacobson MF, Joshi TK, LaDou J, Landrigan PJ, Lee PR, Lockwood AH, MacGregor G, Melnick R, Messing K, Needleman H, Ozonoff D, Ravanesi B, Richter ED, Sass J, Schubert D, Suzuki D, Teitelbaum D, Temple NJ, Terracini B, Thompson A, Tickner J, Tomatis L, Upton AC, Whyatt RM, Wigmore D, Wilson T, Wing SB, Sharpe VA. *Re: Regulatory Toxicology and Pharmacology*. Int J Occup Environ Health. 2003 Oct-Dec;9(4):386-9; author reply 389-90.

27. Jacobson MF, Sharpe VA, Angell M, Ashford NA, Blum A, Chary LK, Cho M, Coull BC, Davis D, Doolittle RF, Egilman D, Epstein SS, Greenberg M, Hooper K, Huff J, Joshi TK, Krimsky S, LaDou J, Levenstein C, Miles S, Needleman H, Pellegrino ED, Ravanesi B, Sass J, Schecter A, Schneiderman JS, Schubert D, Soffritti M, Suzuki D, Takaro TK, Temple NJ, Terracini B, Thompson A, Wallinga D, Wing S. *Editorial policies on financial disclosure*. Nat Neurosci. 2003 Oct;6(10):1001.

28. Sass J. MacLennan et al report on an elevated incidence of prostate cancer among workers in a triazine manufacturing plant. J Occup Environ Med. 2003 Apr;45(4):343-4; author reply 344

29. Sass J. Continued insensitivity to conflicts of interest at IARC. Int J Occup Environ Health, 2003 Jan-Mar; 9(1); 88-9. discussion 89.

30. Sass J. Lead IARC towards compliance with WHO/IARC Declaration of Interests (DOI) policy. Int J Occup Environ Health. 2002 Jul-Sep;8(3):277-8.

31. Axelson O, Castleman B, Epstein S, Franco G, Giannasi F, Grandjean P, Greenberg M, Hooper K, Huff J, Jacobson M, Joshi TK, Kulkarni GK, LaDou J, Mazaheri M, Mekonnen Y, Melnick R, Mirabelli D, Ofrin R, Partanen T, Pott F, Sass J, Soskolne CL, Suplido ML, Terracini B, Tomatis L, Ungvary G, Watterson A, Wesseling C, Yassi A. Re: Implementation of WHO Guidelines on Disclosure of Interest by members of WHO Expert Panels. Int J Occup Environ Health. 2002 Jul-Sep;8(3):271-3.

32. Sass JB, Greer L. Re: concern that working group members who will be assessing styrene have financial conflicts of interest. Int J Occup Environ Health. 2002 Apr-Jun;8(2):153-5.

33. Choich JA, Sass JB, Silbergeld EK. A novel system applying the 2-deoxyglucose method to fish for characterization of environmental neurotoxins. Toxicology Mechanisms and Methods. 2002; 12: 35-43

34. Gurney S, Sass J. Public trust requires disclosure of potential conflicts of interest. Nature. 2001 Oct 11;413(6856):565.

35. Sass JB, Haselow DT, Silbergeld EK. Methylmercury-induced decrement in neuronal migration may involve cytokine-dependent mechanisms: a novel method to assess neuronal movement in vitro. Toxicol Sci. 2001 Sep;63(1):74-81.

36. Sass JB, Martin CC, Krone PH. Restricted expression of the zebrafish hsp90alpha gene in slow and fast muscle fiber lineages. Int J Dev Biol. 1999;43(8):835-8.

37. Krone PH, Lele Z, Sass JB. Heat shock genes and the heat shock response in zebrafish embryos. Biochem Cell Biol. 1997;75(5):487-97. Review.

38. Sass JB, Krone PH. HSP90alpha gene expression may be a conserved feature of vertebrate somitogenesis. Exp Cell Res. 1997 Jun 15;233(2):391-4.

39. Krone PH, Sass JB, Lele Z. Heat shock protein gene expression during embryonic development of the zebrafish. Cell Mol Life Sci. 1997 Jan;53(1):122-9. Review.

40. Sass JB, Weinberg ES, Krone PH. Specific localization of zebrafish hsp90 alpha mRNA to myoD-expressing cells suggests a role for hsp90 alpha during normal muscle development. Mech Dev. 1996 Feb;54(2):195-204.

41. Krone PH, Sass JB. HSP 90 alpha and HSP 90 beta genes are present in the zebrafish and are differentially regulated in developing embryos. Biochem Biophys Res Commun. 1994 Oct 28;204(2):746-52.

42. Sass JB, Ang LC, Juurlink BH. Aluminum pretreatment impairs the ability of astrocytes to protect neurons from glutamate mediated toxicity. Brain Res. 1993 Sep 10;621(2):207-14.

43. Sass JB, Ang LC, Juurlink BH. A simple, yet versatile, co-culture method for examining neuron-glia interactions. J Neurosci Methods. 1993;47(1-2):115-21.

44. Ang LC, Bhaumick B, Munoz DG, Sass J, Juurlink BH. Effects of astrocytes, insulin and insulin-like growth factor I on the survival of motoneurons in vitro. J Neurol Sci. 1992 Jun;109(2):168-72.

Book Chapters:

Soffriti M, Sass J, Castleman B, Gee D. 2012. Vinyl Chloride – A Saga of Secrecy and the Value of Animal Testing. Late Lessons from Early Warnings: Science, Precaution and Politics. European Environmental Agency report. April, 2012

Sass JB, D Mergler, EK Silbergeld. 2000. Environmental toxins and neurological disease. In: *Diseases of the Nervous System: Clinical Neuroscience and Therapeutic Principles*. 3rd edition. Eds. AK Asbury, G McKhann, WI McDonald, PJ Goadsby, JC McArthur. Cambridge Univ Press, NY, NY. Chapter 111.

Reports

- *Strengthening Toxic Chemical Risk Assessments to Protect Human Health*. An NRDC issue paper by Sarah Janssen, Jennifer Sass, Ted Schettler, Gina Solomon. February 2012.
- *The Delay Game: How the industry ducks regulation of the most toxic substances*. An NRDC report by Jennifer Sass and Daniel Rosenberg. October, 2011
- *Atrazine: Poisoning the Well – Atrazine continues to contaminate surface water and drinking water in the United States*. An NRDC Issues Paper by Mae Wu, Mayra Quirindongo, Jennifer Sass, Andrew Wetzler. April 2010
- *Asleep at the switch: How EPA is ignoring atrazine contamination in surface and drinking water in the Central United States.* An NRDC Issues Paper by Mae Wu, Mayra Quirindongo, Jennifer Sass, Andrew Wetzler. August, 2009
- *Effective and practical disclosure policies: NRDC paper on workshop to identify key elements of disclosure policies for health science journals*. An NRDC paper by Jennifer Sass. June, 2009. http://www.nrdc.org/health/disclosure/
- *Deepest Cuts: Repairing Health Monitoring Programs Slashed Under the Bush Administration*. An NRDC Issues Paper by Miriam Rotkin-Ellman, Mayra Quirindongo, Jennifer Sass, Gina Solomon. December 2008. http://www.nrdc.org/health/deepestcuts/
- *Nanotechnology's invisible threat: small science, big consequences*. An NRDC Issues Paper by Jennifer Sass. May, 2007. http://www.nrdc.org/health/science/nano/contents.asp

U.S. Congressional Testimony and Briefings

- Briefing before the US Senate Committee on Environment and Public Works on recommendations of the National Academies for improving risk assessments. Washington, DC. February 28, 2011

- Briefing before the US Senate Committee on Commerce, Science, and Transportation. Green Chemistry: merging business and sustainability. Washington, DC. June 26, 2009

- Testimony before the US House Energy and Commerce Committee, Subcommittee on Oversight and Investigations at hearings entitled, "Science Under Siege: Scientific Integrity at the Environmental Protection Agency". Washington, DC. September 18, 2008

- Testimony before the US Senate Committee on Environment and Public Works, Subcommittee on Transportation Safety, Infrastructure Security, and Water Quality hearing entitled, "Pharmaceuticals in the Nation's Water: Assessing Potential Risks and Actions to Address the Issue". April 15, 2008

- Briefing before the US Senate Committee on Environment and Public Works. Perspectives on Nanotechnology: Business, Government, and Public Health. Washington, DC. May 30, 2007

- Testimony before the US House of Representatives Committee on Science and Technology. Legislative Hearing on the EPA Fiscal Year 2008 Research and Development Budget Proposal. March 14, 2007.

National Academies of Science:

- Invited panelist to address the National Research Council committee to Review of the Federal Strategy to Address Environmental, Health, and Safety Research Needs for Engineered Nanoscale Materials. BEST-K-07-02-A. May, 2008

- Prepared comments and presented testimony to the National Research Council Committee on the Health Risks of Phthalates. BEST-K-07-07-A. December, 2007

- Prepared comments and presented testimony to the National Research Council Committee on Improving Risk Analysis Approaches Used by the U.S. EPA. BEST-K-05-02-A. June, 2007

- Prepared comments and presented testimony to the Institute of Medicine Roundtable on Environmental Health Sciences, Research and Medicine. National Institute of Environmental Health Science Roadmap. June, 2006

- Prepared comments and presented testimony to the National Research Council Committee on Ensuring the Best Science and Technology Presidential and Federal Advisory Committee Appointments. Call for Comments on Science and Technology Presidential and Federal Advisory Committee Appointments. July 21, 2004

- Prepared comments and presented testimony to the National Research Council on scientific and regulatory issues for the Committee on Environmental Decision Making: Principles and Criteria for Models (BEST-K-03-02-A). March, 2004

- Prepared comments and presented testimony to the National Research Council on scientific issues and environmental concerns, for the "Committee on Coeur d'Alene River Basin" Superfund Site Assessment and Remediation (BEST-K-02-04-A). Additional comments identified concern that numerous members of the committee had close ties to polluters. January, 2004

- Prepared comments and presented testimony to the National Research Council on the scientific issues, for the "Committee to Assess the Health Implications of Perchlorate Ingestion" (BEST-K-03-05-A). Additional comments identified concern that numerous members of the committee had close ties to perchlorate polluters. October, 2003

- Prepared comments and presented testimony to the National Research Council raising concern of industry influence on the provisional committee appointments and charge questions for the NAS advisory panel to address the Toxicologic Risk of Fluoride in Drinking Water (BEST-K-02-05-A). May, 2003

- Prepared comments and presented testimony to the National Academies on the scientific issues, for the "Provisional Committee Appointments for NAS Advisory Panel - Use of Third Party Toxicity Research with Human Research" (STLP-Q-02-02-A). Additional comments identified concern that numerous members of the committee had close ties to pesticide manufacturers. December 3, 2002

Service on U.S. Federal scientific and stakeholder committees:

- Invited panelist. EPA Office of Water. Arsenic Small Systems Working Group. 2012.

- Invited panelist. EPA Office of Water. Drinking Water Strategy – Contaminants as a group process. Stakeholder meeting. September, 2010

- Selected to serve on the Leadership Council of the CDC National Conversation on Public Health and Chemical Exposures. We met throughout 2010 and will continue our work into 2011. The goal is to successfully engage a broad range of groups and individuals representing impacted communities, health professionals, regulatory agencies, and industry to develop an action agenda with clear, achievable recommendations to strengthen government's efforts to protect health and the environment from chemical hazards.

- Invited participant on a working group of the President's Council of Advisors on Science and Technology (PCAST), to review the National Nanotechnology Initiative. 2010

- Invited participant on the federal advisory committee to provide Peer Review of the EPA Nanotechnology White Paper. Coordinated by Versar, Inc. (Keith Drewes). 2006

- Member of the National Toxicology Program Nanotechnology Working Group, NTP Board of Scientific Counselors. National Institute of Environmental Health Sciences. 2005.

- Member of the Public Interest Partners of the National Institute of Environmental Health Sciences (NIEHS PIP). 2005 to 2011.

- Invited member of the National Toxicology Program U.S. Department of Health and Human Services High-Throughput Screening Assays Workshop. December 14-15, 2005. Crystal City, Virginia

- Member of the Interim Ad-Hoc Work Group on Nanoscale Materials, National Pollution Prevention and Toxics Advisory Committee (NPPTAC), U.S. EPA. July-October, 2005

- Invited member of the National Toxicology Program Retreat. Dept of Health and Human Services. To provide input on the Draft paper, Toxicology in the 21st Century: The Role of the National Toxicology Program. Greensboro, NC. August, 2004

- Participant in the Technical Peer Review Workshop on the EPA Risk Assessment Forum Draft Framework for Cumulative Risk Assessment. June, 2002

- Member of the EPA/USDA Pesticide Program Dialogue Committee (PPDC). This Committee provides a forum for a diverse group of stakeholders to provide feedback to the pesticide program on various pesticide regulatory, policy and program implementation issues. Summer 2001 to present

- Member of the EPA/USDA Committee to Advise on Reassessment and Transition (CARAT). The purpose of CARAT to provide advice on strategic approaches for pest management planning, transition, and tolerance reassessment for pesticides as required by the Food Quality Protection Act (FQPA). This committee advises EPA and USDA on ways to ensure smooth implementation of FQPA through use of sound science, consultation with stakeholders, increased transparency, and reasonable transition for agriculture. Summer 2001

- Member of the EPA Expert Peer Review for the *Draft Framework for Cumulative Risk Assessment* document, US EPA NCEA-F-1098. August 2, 2001. Workshop discussion Arlington, VA. August 2001

Selected Recent Testimony and Comments to the U.S. Environmental Protection Agency:

Pyrethroid Cumulative Risk Assessment. EPA-HQ-OPP-2011-0746. January, 2012.

Chlorpyrifos preliminary human health risk assessment. EPA-HQ-OPP-2008-0850. October, 2011

Pesticides; Policies concerning nanomaterials. EPA–HQ–OPP–2010-0197. August, 2011

Revisions to EPA's rule on protection of subjects in human research involving pesticides. EPA-HQ-OPP-2010-0785. April, 2011.

Proposal to conditionally register nanosilver pesticide product. September, 2010

Proposed Rule to increase public availability of the identities of the inert ingredients in pesticide products. EPA-HQ-OPP-2009-0635. April 2010

EPA Policy paper on revised risk assessment methods for agriculture workers, children of workers in agriculture fields, and pesticides with no food uses. EPA-HQ-OPP-2009-0889. April 2010

EPA Draft guidance for pesticide registrants on pesticide drift labeling. EPA-HQ-OPP-2009-0628. March, 2010.

Draft Toxicological Review of Trichloroethylene: In support of the summary information in the Integrated Risk Information System. EPA-HQ-ORD-2009-0791. January, 2010.

FIFRA Scientific Advisory Panel on atrazine. EPA-HQ-OPP-2009-0851. February, 2010.

Fungicides mancozeb, maneb, metiram, and thiram. EPA-HQ-OPP-2009-0431. November, 2009

Atrazine. EPA-HQ-OPP-2009-0759. October, 2009

Perchlorate in drinking water. EPA-HQ-OW-2009-0297. September, 2009

NRDC petition to cancel endosulfan. EPA-HQ-OPP-2002-0262. June, 2009

Testimony and Comments to Regulatory Agencies (other than EPA):

1. Presented testimony and comments at the Maine State Legislature on the health hazards of bisphenol A. March, 2011.

2. Submitted comments in response to the March, 2009 Presidential Memo on Scientific Integrity: Request for Public Comment. May, 2009.

3. Submitted scientific and legal comments to the New Jersey Department of Environmental Protection on a drinking water standard for perchlorate. December, 2007

4. Submitted scientific and legal comments to the FDA on nanoparticles in sunscreens, cosmetics, and personal care products. December 21, 2007. Docket No. 1978N-0038

Selected Recent Invited Speaker:

1. Invited panelist. Society of Toxicology and EPA Office of Research and Development. Workshop on Contemporary Concepts in Toxicology – Building for Better Decisions. May, 2012.

2. Invited speaker at The Kavli Science Journalism Workshop. Nano: the newest technology. Massachusetts Institute of Technology (MIT), June 2012.

3. Invited speaker at University of Idaho College of Law, Moscow Idaho. March, 2012

4. Invited speaker and workshop participant, Arizona State University, College of Law. The biggest issues for the smallest stuff: regulation and risk management of nanotechnology. Phoenix, AZ. March, 2011.

5. Invited speaker, DC EcoWomen. Washington, DC March, 2011.

6. Invited speaker, The Appropriate Use of Science in Public Policy. Hosted by the Professionals for the Public Interest (PftPI). Washington, DC October, 2010

7. Invited speaker, graduate class on Science and Policy. Virginia Tech, Blacksburg VA. October, 2010

8. Invited panelist. Nanotechnology: Should we sweat the little things? Society of Environmental Journalists, 20th Annual Conference. Missoula, MT October, 2010.

9. Invited participant. Workshop on Assessing Consistency in Epidemiology Data for Application in Regulatory Risk Assessment. Johns Hopkins School of Public Health (Ron White), Baltimore MD. September, 2010

10. The 10th Transatlantic Consumer Dialogue (TACD) annual meeting. Invited speaker on nanomaterials. Brussels, June, 2009

11. Science, Technology, and Public Policy Program at the Ford School of Public Policy at the University of Michigan. Invited speaker. Title: Nanotoxicology: a review of the science and policy. January, 2009

12. NIEHS AFGE Local 2923 and the NIEHS Diversity Council. Labor Day Keynote Lecturer. Title: Occupational safety, public health, and environmental protection: The historical role of women in making the connection. Research Triangle Park, NC. August, 2008.

13. Region VI Pretreatment Association, U.S. EPA, and State EPA Region VI. Title: Nanomaterials in waste water. Invited presenter. Oklahoma City, OK. August, 2008.

14. Global conference on occupational and environmental cancer prevention. Invited speaker. Title: An international environmental NGO assessment of environmental cancers and their prevention. Scotland. April, 2008.

15. Resources for the Future, and Wilson Center Project on Emerging Nanotechnologies. Nanotechnology and Nature: Reducing risks and reaping rewards. Washington, DC. June 6, 2007

16. Beyond Pesticides 25th National Pesticide Forum. New Opportunities for Protecting Health and the Environment. Nanotechnologies. Chicago, Il. June 2, 2007

17. American Bar Association Annual Meeting. Section of Environment, Energy, and Resources. Regulation of nanotechnology: size does matter. San Diego, CA. October, 2006

18. Johns Hopkins Bloomberg School of Public Health. Risk Policy, Management, and Communications course. Professor, Ronald White. Baltimore, MA. November/December 2003, 2004, 2005, 2006, 2007, 2008

Provided Peer-Review for Scientific and Professional Journals

Accountability in Research
American Journal of Public Health
Archives of Environmental & Occupational Health
Environmental Health Perspectives
European Environmental Agency, Late Lessons from Early Warnings
International Journal of Occupational and Environmental Health
Journal of Nanoparticle Research
Public Health Reports

Selected Recent Professional and Public Service

1. Coming Clean Collaborative. Workgroup leader, Emerging Technologies. 2007, 2008, 2009, 2010, 2011, 2012

2. Center for the Environmental Implications of Nanotechnology (CEINT). Member of the External Advisory Board. 2009, 2010

3. Public Interest Partners, National Institute of Environmental Health Sciences (NIEHS). Liaison with the NIEHS National Advisory Environmental Health Sciences Council (NAEHS). 2007, 2008, 2009, 2010

4. Invited expert on the North American Commission for Environmental Cooperation meeting of experts to guide the further development of its Special Report on Toxic Chemicals and Children's Health in North America. Montreal, Canada. November, 2004

5. Organizer and session moderator for the annual general conference of the Association for Science in the Public Interest (ASIPI). George Mason University, VA. June, 2003

6. Reviewer of applicants for the American Association for the Advancement of Science/ National Institutes of Health Science Policy Fellowship Program. March, 2003, 2004

7. Invited participant in the North American Free Trade Agreement (NAFTA) Trilateral Workshop on Risk Assessment and Children's Environmental Health. Oaxaca, Mexico. February, 2003

8. Invited observer of the International Agency for Research on Cancer (IARC) Monographs on the Evaluation of Carcinogenic Risks to Humans. Preparation of Volume 84: Some drinking-water disinfectants and contaminants, including arsenic. Lyon, France. October, 2002

9. Member of the Roundtable Workshop to discuss Genomics and Environmental Policy, organized by the Woodrow Wilson International Center for Scholars, Washington, DC. Purpose is to discuss the impacts and implications of advances in genomics on environmental protection and policy. May, October, 2002

10. Member of the LifeLine Advisory Committee. This committee advises and reviews the development of the LifeLine model for cumulative risk assessment, developed by Hampshire Research Institute. Coordinators are Paul Price and Christine Chaisson. Summer 2001

11. Co-chair of the organizing committee for the annual general meeting of the Association for Science in the Public Interest, Richmond, VA. May, 2001

Selected Recent Scientific Conference Presentations

1. Sass, JB. 2011. Panelist. Natural Resources Defense Council (NRDC) perspective on transparency, speed and stakeholders involvement in EPAs chemical risk assessment. Society for Risk Analysis Annual Meeting. December, Charleston, SC.

2. Sass JB. 2009. Panelist. The Regulatory Frontier: Addressing products of nanotechnology. Abstract 660. Society of Toxicology Annual Meeting. March, Baltimore, MD.

3. Sass JB. 2008. Panelist. Data gaps and research needs for improving risk analysis of nanoscale materials and nanotechnology. Society of Risk Analysis. September, Washington, DC.

4. Sass JB. 2006. Panelist. Nanotechnology challenges. American Public Health Association Annual Meeting. November, Boston MA

5. Sass JB. 2005. Vinyl Chloride Carcinogenicity and EPA's Chemical Assessment Process. Abstract 121339. American Public Health Association Annual Meeting. December, Philadelphia, PA.

6. Sass JB, Colangelo A. 2005. U.S. Regulation of Atrazine: Taking Care of Business. Collegium Ramazzini Annual Meeting. Living in a Chemical World. September, Bologna, Italy.

7. Sass J, Colangelo A. 2005. EPA Review of Atrazine Cancer Risks: Taking Care of Business. Meeting of Atrazine and the health of humans and wildlife: state of the science and future research needs. University of Iowa. April, Iowa City, Iowa

8. Sass, JB. 2004. Panelist. Scientific Integrity in Regulation. American Public Health Association Annual Meeting. Session 3214.0. November, Washington, DC

9. Sass, JB. 2003. Panelist. Social Determinants of Health: sound science for sale? (session 4259.0). Presentation title, “Are the regulated industries regulating themselves…where is government?” Abstract 70559. American Public Health Association Annual Meeting, November. San Francisco, CA

10. Sass, JB. 2003. Panelist. Increase Influence of Industry in American and International Assessments of Toxic Chemicals. First International Scientific Conference on Occupational and Environmental Health at the National Institute of Occupational and Environmental Health. November. Hanoi, Viet Nam

Selected Recent Community Service and Activities

- Garrett Park Conservation Action Network. 2008-ongoing
- Garrett Park Conservation Trust, Vice-President. 2009-2011
- Wilderness First Responder certified; Wilderness First Aid certified, 2010
- ACA certified whitewater kayak instructor, American Canoe Association, 2009-ongoing
- Solo oar-raft the Grand Canyon, Colorado River, 225 miles. 2009
- Volunteer organizer with the Potomac White Water Festival. 2008-2011
- Runner and fund-raiser for the *National AIDS Marathon*. Completed the Marine Corps Marathon, Washington DC. 2005
- Runner and fund-raiser for the *National AIDS Marathon*. Completed the Baltimore Marathon, Baltimore MD. 2004

EXHIBIT B

PETITION TO REVOKE ALL TOLERANCES AND CANCEL ALL REGISTRATIONS FOR THE PESTICIDE CHLORPYRIFOS

Filed 12 September 2007

The Natural Resources Defense Council (NRDC) and Pesticide Action Network North America (PANNA) petition the U.S. Environmental Protection Agency (EPA) to revoke all tolerances and cancel all registrations for the pesticide chlorpyrifos. This petition is filed pursuant to 21 U.S.C. § 346a(d).

I. Introduction

Chlorpyrifos is one of the most widely used insecticides in the United States. It is used on various food and feed crops, on golf courses, as a non-structural wood treatment, and as an adult mosquitocide. Agriculturally, approximately 10 millions pounds are applied annually, with use on corn comprising the largest market (using approximately 5.5 million pounds ai). [1]

Chlorpyrifos belongs to a class of pesticides called organophosphates, which EPA has grouped together based on their common mechanism of toxicity. The devastating effects of this class of pesticides, originally designed as wartime nerve agents including sarin gas, are attributed to their inactivation of an enzyme called cholinesterase.[2] This enzyme is responsible for the timely deactivation of the nerve signaling protein acetylcholine.

Acetylcholine is a messenger of the nervous system, a "neurotransmitter," which carries the signal from a nerve cell to its target. Important targets of acetylcholine include muscles, sweat glands, the digestive system, and even heart and brain cells. In particular, acetylcholine signals activity of the "rest and digest" portions of the nervous system (the parasympathetic system) that stimulates digestion, slows the heart rate, and helps the body to conserve energy. The organophosphate pesticides, including chlorpyrifos, block the ability of cholinesterase to deactivate acetylcholine after its message is delivered. The resulting accumulation of acetylcholine causes over-activation of all its targets. Clinical symptoms of organophosphate poisoning can include: eye pupil contraction, increased salivation, nausea, dizziness, confusion, convulsions, involuntary urination and defecation, and, in extreme cases, death by suffocation resulting from loss of respiratory muscle control.

The state of the science identifying many various adverse health effects associated with dietary exposure to chlorpyrifos supports a ban on chlorpyrifos and revocation of all food tolerances. This petition summarizes the overwhelming scientific evidence that chlorpyrifos is too dangerous to be re-registered for food uses.

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[1] "Chlorpyrifos Facts." EPA website, <www.epa.gov/oppsrrd1/REDs/factsheets/chlorpyrifos_fs.htm, 8 Mar 2007. All home uses of chlorpyrifos have been canceled "except ant and roach baits in child-resistant packaging." All uses for termite control were required to be phased out by December 31, 2005. IRED, p.71

[2] As chemical weapons, the production and stockpiling of organophosphate nerve agents are outlawed by the United Nations' 1993 Convention on the Prohibition of the Development, Production, Stockpiling and Use of Chemical Weapons and on Their Destruction. ¶71(b)..

II. Legal Standard

EPA regulates pesticides under two statutes, the Federal Food, Drug, and Cosmetic Act (FFDCA), 21 U.S.C. § 346a and the Federal Fungicide, Insecticide, and Rodenticide Act (FIFRA), 7 U.S.C. § 136 *et seq.* The Food Quality Protection Act of 1996 ("FQPA") significantly amended both the FFDCA and FIFRA by mandating that health-based and child-protective standards drive decisions about acceptable levels of pesticide residues in food and the environment. FIFRA requires that pesticides must be registered to be sold in the United States.[3] EPA may not register a pesticide unless the chemical will perform its intended function without causing any "unreasonable adverse effects on the environment."[4]

The FFDCA, as amended by the FQPA, authorizes EPA to set tolerances (maximum allowable levels) for pesticide residues in food or to grant exemptions from the requirement to have a tolerance.[5] EPA may "establish or leave in effect a tolerance for a pesticide chemical residue in or on a food only if the Administrator determines that the tolerance is safe."[6] The term "safe" means that "there is a reasonable certainty that no harm will result from aggregate exposure" to the pesticide, "including all anticipated dietary exposures and all other exposures for which there is reliable information."[7] A pesticide may not be used on a particular food unless there is a tolerance or exemption for that food.[8] The Food and Drug Administration and the U.S. Department of Agriculture are charged with enforcing these regulations by randomly sampling fruits and vegetables for exceedances of tolerances or use of unregistered pesticides or banned pesticides.

The FFDCA explicitly requires that EPA, in establishing a tolerance, must assess the risk that a pesticide poses to infants and children in particular.[9] Before EPA can establish a tolerance, the Agency shall "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure" to the pesticide, and shall "publish a specific determination regarding the safety of the pesticide chemical residue for infants and children."[10] In ensuring that the statutory safety standard is met, EPA must consider available information concerning "the special susceptibility of infants and children," including "neurological differences between infants and children and adults, and effects of *in utero* exposure to pesticide chemicals."[11] EPA must also base its tolerance decision on available information about "food consumption patterns unique to infants and children" and the "cumulative effects on infants and children of [pesticides] that have a common mechanism of toxicity."[12] EPA acknowledges that, when setting

[3] 7 U.S.C. § 136a.
[4] 7 U.S.C. § 136a(c)(5)(C).
[5] 21 U.S.C. §§ 345a(b) & (c).
[6] *Id.* § 346a(b)(2)(A)(i).
[7] *Id.* § 346a(b)(2)(A)(ii).
[8] *Id.* § 346a(a)(1).
[9] *Id.* § 346a(b)(2)(C).
[10] *Id.* §§ 346a(b)(2)(C)(ii)(I) & (II).
[11] *Id.* § 346a(b)(2)(C)(i)(II).
[12] *Id.* §§ 346a(b)(2)(C)(i)(I) & (III).

new tolerances under the standard, it "must now focus explicitly on exposures and risks to children and infants."[13]

Furthermore, "an additional tenfold margin of safety for the pesticide chemical residue and other sources of exposure shall be applied for infants and children to take into account potential pre- and post-natal toxicity and completeness of the data with respect to exposure and toxicity to infants and children."[14] EPA can depart from this requirement and use a different margin of safety "*only if*, on the basis of *reliable data*, such margin will be safe for infants and children."[15]

Tolerance decisions are driven by the level of pesticide residue detected on food, which is the amount of pesticide that remains on a commodity after a pesticide is applied at a rate that meets or exceeds effective pest control.[16] They are "not based primarily on health considerations.... Their primary purpose is to ensure compliance with good agricultural practice."[17] On the other hand, reference doses (RfD), which represent the amount of pesticide residue that is safe for consumers to eat, are set, if at all, after tolerances. Based on residue data from food and drinking water and considering complexities, such as cooking, if the dietary exposure exceeds the RfD, EPA informs the registrant that the tolerance is unacceptably high. The registrant is tasked with proposing mitigation options, such as a lower application rate or cancellation of that use. As such, the pesticide control framework was established to maintain pesticide residues on food not at safe levels but at or below tolerance levels.

III. Factual Background

In 2001, EPA completed the chlorpyrifos aggregate assessment, called an Interim Reregistration Eligibility Decision (IRED), which revised, but retained, many of the pre-existing food tolerances (allowable residue limits on food).[18] In its 2002 comments on the IRED (Docket ID No. OPP-34203G), NRDC challenged the scientific limitations of the IRED, identified evidence of harm, and highlighted that there is inadequate evidence to establish a safe level at which infants and children will not suffer any developmental harm due to chlorpyrifos exposure. EPA never responded directly to NRDC's comments or other comments submitted by other public interest advocates, including the Pesticide Action Network North America (PANNA) and the New York Attorney General (Docket ID No. OPP-34203G).

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[13] EPA, Fact Sheet: Protecting Children from Pesticides (Jan. 2002) (www.epa.gov/pesticides/factsheets/kidpesticide.htm) ("The 1996 Food Quality Protection Act set tougher standards to protect infants and children from pesticide risks.")
[14] 21 U.S.C. § 346a(b)(2)(C).
[15] *Id.* (emphasis added).
[16] J. Sass and S. Kegley. Call with EPA to discuss chlorpyrifos. From HED: Jack Housenger, Anna Lowit, and Tom Moriarty; from RD: Venus Eagle; from SRRD: Pete Caulkins, Margaret Rice, and Tom Myers; from OGC: Mark Dyner and Jon Fleuchaus. July 17, 2007
[17] Philip J. Landrigan and others, Pesticides In The Diets Of Infants And Children (Washington, D.C.: National Academy Press, 1993), 9.
[18] 66 Fed Reg 57073 (Nov 14, 2001) Organophosphate Pesticide; Availability of Chlorpyrifos Interim Risk Management Decision Document. IRED at 64-68.

In 2006, EPA completed the cumulative risk assessment (CRA) for all organophosphates (OPs), including chlorpyrifos, and reaffirmed the chlorpyrifos IRED without change, despite new, significant published studies that emerged during this time showing harm. Without addressing the comments by NRDC and other public interest advocates and without referencing much of the data that had been available since 2001, the Agency concluded that chlorpyrifos uses would be eligible for reregistration and that the current pesticide tolerances met the legal safety standard.[19] Because EPA failed to respond to any of NRDC's comments, this petition incorporates by reference the January 14, 2002 NRDC comments and those of other public health advocates.

According to EPA, tolerances are generally reassessed under two possible scenarios. First, an application to register a new use for a pesticide forces EPA to review the aggregate assessment and determine whether the new use 'fits' into the aggregate risk evaluation (i.e. the aggregate exposure from all use scenarios is at or below the RfD); second, during registration review, which occurs about every fifteen years, must reconsider the aggregate risk evaluation.[20] Tolerances are not reassessed based on new data, new science, or new evidence of harm. However, scientific evidence that has emerged since 2001 when the chlorpyrifos IRED was published reinforce the earlier science showing that exposure to chlorpyrifos causes many adverse health effects. In fact, both the weaknesses in the studies relied on by EPA in the IRED and the failure to incorporate evidentiary science since 2001 undermine EPA's decision to re-register chlorpyrifos and retain its tolerances. In this petition we summarize the pre-2001 data and identify relevant post-2001 scientific evidence relevant to the risk assessment of chlorpyrifos.

IV. A Risk Assessment Must Account for the Full Spectrum of Toxicity

The assessment of the health effects associated with particular pesticides includes both an aggregate assessment, which analyzes the risk from multiple routes of exposures (food, water, residential uses) to a single pesticide, and a cumulative assessment, which analyzes the risk from cumulative exposure to a class of pesticides that share a common mode of action. The Agency grouped chlorpyrifos with the other organophosphates to conducts its cumulative risk assessment. For the organophosphate cumulative assessment, EPA used the endpoint of plasma and red blood cell cholinesterase inhibition in dams to determine an acceptable maximum level of cumulative exposure to organophosphate pesticides (identified as a 10% effect level, or benchmark dose 10, BMD10).

Alternately, for the individual aggregate assessment of chlorpyrifos, EPA identified the critical endpoint as structural alterations in brain development in exposed rodent pups at

[19] Memo from Debra Edwards to Jim Jones, re: Finalization of Interim Reregistration Eligibility Decisions (IREDs) and Interim Tolerance Reassessment and Risk Management Decisions (TREDs) for the Organophosphate Pesticides, and Completion of the Tolerance Reassessment and Reregistration Eligibility Process for the Organophosphate Pesticides, July 31, 2006.

[20] J. Sass and S. Kegley. Call with EPA to discuss chlorpyrifos. From HED: Jack Housenger, Anna Lowit, and Tom Moriarty; from RD: Venus Eagle; from SRRD: Pete Caulkins, Margaret Rice, and Tom Myers; from OGC: Mark Dyner and Jon Fleuchaus. July 17, 2007

the lowest dose tested to determine an acceptable maximum level of aggregate exposure to chlorpyrifos (identified as the RfD).[21] The Agency determined that there was demonstrated evidence of neuropathology and increased susceptibility following pre-natal exposure to chlorpyrifos.[22] Since the developmental neurotoxicity test (DNT) did not identify a no-effect level, and to account for possible non-cholinergic effects in the brain, EPA retained the FQPA factor of 10X. [23] However, this petition reviews scientific evidence that a 10X factor is insufficient, and, as explained below, no safe level of early-life exposure to chlorpyrifos can be supported.

For the organophosphate cumulative assessment, EPA used only the endpoint of cholinesterase inhibition in female rat brain at 21-days of exposure. The Agency argues that there was no evidence of differences between adults and pups for this endpoint and eliminated the FQPA factor by dropping it to 1X. However, as discussed below, the Agency's explanation for this decision does not reflect a true representation of the data used by EPA.

A. Genetic Evidence of Vulnerable Populations

As part of the risk calculation for a particular pesticide, EPA will often include an intra-species variability factor to account for the variation between different people's responses to the same exposure (both chemical and dose). The same dosage of chlorpyrifos may be very harmful to one person and have no effect on another person. This is because of individualized factors that include differences in nutritional status, health or disease status, activity level, lifestyle, exposure to other chemicals or agents, and inherent genetic differences in the activity of the enzymes that break down toxic chemicals in the body. Conventionally, the Agency uses a standard intra-species factor of 10X, presuming no more than a 10-fold difference in susceptibility across a diverse human population.

Paraoxonase (PON1) is a protein (enzyme) that behaves very differently from one individual to the next, and aids in recovering from pesticide toxicity. PON1 detoxifies many of the organophosphates, particularly chlorpyrifos, through catalyzing the hydrolysis of its toxic oxon metabolite. In other words, PON1 breaks down the toxic by-products of chlorpyrifos that are produced during its metabolism, so that they do not build up in the body. A slow-acting genotype of PON1 is less efficient at detoxifying the oxon and is therefore associated with increased pesticide toxicity.[24]

Published epidemiologic studies by Furlong and colleagues in 2003 and 2006 report that the age-related activity of PON1 may impair the ability for perinatal and juvenile animals

[21] IRED at 17
[22] IRED at 16
[23] Makris S, Raffaele K, Sette W, Seed J. A retrospective analysis of twelve developmental neurotoxicity studies submitted to the USEPA Office of Prevention, Pesticides, and Toxic Substances (OPPTS). Draft 11/12/98. Available at http://www.epa.gov/scipoly/sap/meetings/1998/december/neuro.pdf
[24] Lee, BW, London, L, Paulauskis, J, Myers, J, Christiani, DC. Association Between Human Paraoxonase Gene Polymorphism and Chronic Symptoms in Pesticide-Exposed Workers. J Occup Environ Med, 2003 Feb; 45(2)

and humans to recover from pesticide toxicity.[25, 26] In fact, the authors reported in their 2006 paper a 164-fold variation in sensitivity to chlorpyrifos between the most sensitive newborn and the least sensitive mother.[27] Although EPA claims to have reviewed this study for the OP CRA , the study supports an intraspecies factor of over 164X whereas the Agency applied only a 10X intraspecies factor to all the organophosphates.[28] In the OP CRA,The Agency specifically acknowledged, and subsequently disregarded, the Furlong et al. study, instead relying on a 2002 study that used a physiologically-based pharmacokinetic (PBPK) model for chlorpyrifos to find that the "response was relatively insensitive to changes in oxonase activity at low doses." [29] Despite EPA's stated preference for human data, and despite the availability of significant informative data derived from unintentionally exposed people (occupational and environmental epidemiologic studies, human biomonitoring [internal dose], and human passive dosimetry [external measurements]), in this case the Agency relied on the model to support its assessment. PBPK models are only as reliable as the data used to design them; they are therefore meant to help bridge data gaps, not override robust data.

EPA's treatment of the PON1 studies with respect to the calculation of the intra-species uncertainty factor provides a stunning example of the Agency turning a blind eye to relevant, robust data. Furthermore, using an intra-species variability factor of 100X or higher – as the results from the Furlong study should prescribe – would drive the tolerances below practicable levels of detections. Practically, tolerances set below the level of detection available for the most sensitive detection methods makes the tolerance unenforceable. EPA should not have ignored the result of the Furlong study and should have applied an intra-species variability factor of at least 150X in the aggregate and cumulative assessments; practically, the Agency should revoke all tolerances for chlorpyrifos.

B. Long-Lasting Effects from Early Life Exposure in Children

Many studies published since 2001 report that fetal exposure to chlorpyrifos is more damaging than adult exposure.[30] Columbia University researchers published two studies

[25] Costa LG, Richter RJ, Li WF, Cole T, Guizzetti M, Furlong CE. Paraoxonase (PON 1) as a biomarker of susceptibility for organophosphate toxicity. Biomarkers. 2003 Jan-Feb;8(1):1-12. Review.
[26] Furlong CE, Holland N, Richter RJ, Bradman A, Ho A, Eskenazi B. PON1 status of farmworker mothers and children as a predictor of organophosphate sensitivity. Pharmacogenet Genomics. 2006 Mar;16(3):183-90.
[27] Furlong CE, Holland N, Richter RJ, Bradman A, Ho A, Eskenazi B. PON1 status of farmworker mothers and children as a predictor of organophosphate sensitivity. Pharmacogenet Genomics. 2006 Mar;16(3):183-90.
[28] CRA at Section I.B page 55
[29] Organophosphorus Cumulative risk assessment – 2006 Update, available at < http://www.epa.gov/pesticides/cumulative/2006-op/index.htm>, 55.
[30] Rauh VA, Garfinkel R, Perera FP, Andrews HF, Hoepner L, Barr DB, Whitehead R, Tang D, Whyatt RW. Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics. 2006 Dec;118(6):e1845-59. Epub 2006 Nov 20.; Perera FP, Rauh V, Whyatt RM, Tang D, Tsai WY, Bernert JT, Tu YH, Andrews H, Barr DB, Camann DE, Diaz D, Dietrich J, Reyes A, Kinney PL. A summary of recent findings on birth outcomes and developmental effects of prenatal ETS, PAH, and pesticide exposures. Neurotoxicology. 2005 Aug;26(4):573-87. Review.; Whyatt RM, Rauh V, Barr DB, Camann DE, Andrews HF, Garfinkel R, Hoepner LA, Diaz D, Dietrich J, Reyes A, Tang D,

from a single New York City (NYC) cohort reporting on the effects of chlorpyrifos on birth outcomes[31] and child development.[32] The authors report on a cohort of NYC African American and Dominican women and babies enrolled over a number of years, that capture changes in exposure levels related to the 2000-2001 ban of chlorpyrifos for residential use. Decreases in birth weight and length were associated with cord blood levels of chlorpyrifos, and the follow-up of children when they reached age 3 showed that the more highly (prenatally) exposed children (chlorpyrifos levels of > 6.17 pg/g plasma) were significantly more likely to experience delays in cognitive and psychomotor development as well as attention problems, attention-deficit/hyperactivity disorder problems, and pervasive developmental disorder problems. The authors report that "the proportion of delayed children in the high-exposure group was five times greater for the Psychomotor Development Index and 2.4 times greater for the Mental Development Index, increasing the number of children possibly needing early intervention services."[33] The adverse effects on birth outcomes were no longer observed among the children in the cohort who were born after the ban took effect (Jan 2001) and concentrations in cord blood were significantly lower, underscoring the benefits of the ban. These data provide strong evidence that prenatal and early-life stage exposure to chlorpyrifos is associated with not only poor birth outcomes (lower birth weight and length), but also long-lasting, and possibly permanent, impaired cognitive development.

In addition to the sensitivity of early life exposures (pre- and peri-natal) to chlorpyrifos, there are data reporting that infants born to mothers with genetically low activity of the PON1 detoxifying enzyme may be an especially vulnerable population. Berkowitz and colleagues from Mount Sinai School of Medicine determined pesticide exposure in a cohort of over 400 women in NYC by a prenatal questionnaire and measurement of maternal blood and urinary metabolites and fetal cord blood. The authors correlated this self-reported exposure information with birth outcomes and found that maternal detectable chlorpyrifos exposure and low PON1 activity correlated with a significant, albeit small, reduction in newborns' head circumference.[34] The authors point to pre-established evidence that small head size is predictive of impaired cognitive ability to

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Kinney PL, Perera FP. Prenatal insecticide exposures and birth weight and length among an urban minority cohort. Environ Health Perspect. 2004 Jul;112(10):1125-32; Perera FP, Rauh V, Tsai WY, Kinney P, Camann D, Barr D, Bernert T, Garfinkel R, Tu YH, Diaz D, Dietrich J, Whyatt RM. Effects of transplacental exposure to environmental pollutants on birth outcomes in a multiethnic population. Environ Health Perspect. 2003 Feb;111(2):201-5.

[31] Whyatt RM, Rauh V, Barr DB, Camann DE, Andrews HF, Garfinkel R, Hoepner LA, Diaz D, Dietrich J, Reyes A, Tang D, Kinney PL, Perera FP. Prenatal insecticide exposures and birth weight and length among an urban minority cohort. Environ Health Perspect. 2004 Jul;112(10):1125-32

[32] Rauh VA, Garfinkel R, Perera FP, Andrews HF, Hoepner L, Barr DB, Whitehead R, Tang D, Whyatt RW. Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics. 2006 Dec;118(6):e1845-59. Epub 2006 Nov

[33] Rauh VA, Garfinkel R, Perera FP, Andrews HF, Hoepner L, Barr DB, Whitehead R, Tang D, Whyatt RW. Impact of prenatal chlorpyrifos exposure on neurodevelopment in the first 3 years of life among inner-city children. Pediatrics. 2006 Dec;118(6):e1845-59. Epub 2006 Nov

[34] Berkowitz GS, Wetmur JG, Birman-Deych E, Obel J, Lapinski RH, Godbold JH, HOlzman IR, Wolff MS. 2004. *In Utero* Pesticide Exposure, Maternal Paraoxonase Activity, and Head Circumference, Env. Health Persp., 112(3):388-91

support their suggestion that the infants of mothers with low PON1 enzyme activity may be an especially vulnerable population.

EPA failed quantitatively to incorporate these important evidentiary data that were published since the 2001 IRED was completed, which report a significant association between real-world chlorpyrifos exposures and real, developmental harm resulting from pre-birth and early childhood exposures. As noted earlier, FQPA imposes a duty on EPA to "focus explicitly on exposures and risks to children and infants."[35] The failure to consider quantitatively the full spectrum of diverse impacts of chlorpyrifos exposure to fetuses is a direct violation of EPA's mandate.

C. No Safe Level in Rodent Developmental Neurotoxicity Study

As discussed above, a substantial body of scientific evidence demonstrates the fetotoxic, neurotoxic, and immunotoxic properties of chlorpyrifos and its oxon metabolite, associated with pre-natal and early life exposures. These exposures have been shown to result in long-lasting, possibly permanent damage to the nervous system. There is no evidence that there is a safe or acceptable level of exposure to chlorpyrifos during pre-birth and early life stages. In fact, EPA staff experts concluded in the EPA human health risk assessment of chlorpyrifos:

> "the weight of the evidence raises concern for an increase in both the sensitivity and susceptibility of the fetus or young animal to adverse biochemical, morphological, or behavioral alterations from chlorpyrifos treatment during brain development. With respect to cholinesterase inhibition, an increase in sensitivity of the young compared to adults was seen all along the dose response curve, even at relatively low doses. There is a clear differential response (2- to ~5-fold) in the young compared to the adult animal after an acute treatment to a relatively low dose of chlorpyrifos. There is also increased sensitivity found after repeated dosing (up to 9-fold), but at the LD10 [lethal dose that results in a 10% death rate] and MTD [maximum tolerated dose]. It is important to point out that *an uncertainty remains concerning the magnitude of the differential response*, given that newborn animals (less than PND 7) have not been characterized for sensitivity. *Results of multiple studies have consistently shown that the developing brain is susceptible to chlorpyrifos treatment*. Effects on the developing CNS that are indicative of the unique susceptibility to the young animal include changes in macromolecular synthesis, altered cell signaling and muscarinic receptor down regulation, as well as morphological alterations in brain development. An uncertainty remains regarding the NOAELs for the susceptibility effects. The

[35] EPA, Fact Sheet: Protecting Children from Pesticides (Jan. 2002) (www.epa.gov/pesticides/factsheets/kidpesticide.htm) ("The 1996 Food Quality Protection Act set tougher standards to protect infants and children from pesticide risks.").

> effects observed raise a high degree of concern that the fetus or young animal is particularly susceptible to adverse outcome if exposed to chlorpyrifos."[36]

The assessment of EPA scientific experts points to substantial scientific evidence that early life exposures to chlorpyrifos are extensively more harmful than adult exposures, and that the magnitude of the differential response is uncertain. This assessment from EPA staff scientists strongly supports the use of the default 10X FQPA factor.

D. Endocrine Disrupting Effects

Thyroid hormone is essential for virtually every function in the body, including reproduction and neurodevelopment. Both animal and human studies have reported that chlorpyrifos may interfere with thyroid hormone function. In a 2006 study of sub-fertile men, chlorpyrifos exposure was associated with reduced levels of thyroid stimulating hormone (TSH) and thyroxine.[37] In a 2005 study of rat pituitary cells, which are normally stimulated to grow after exposure to thyroid hormone, cell growth was inhibited by co-exposure to chlorpyrifos.[38] In an earlier study (1998), exposure to chlorpyrifos in ewes was associated with reduced thyroxine (thyroid hormone) concentrations.[39] More troubling, these effects resulted from exposures at levels similar to those found in the general population, indicating that chlorpyrifos can reduce thyroid hormone and cause endocrine disruption at environmentally relevant levels. In addition to causing infertility, reductions in thyroid hormone concentrations, even at subclinical levels, can result in permanent neurological effects on the developing nervous system of a fetus or newborn.[40, 41]

Studies also indicate that chlorpyrifos can affect the reproductive hormones estrogen and testosterone. Chlorpyrifos is a weak estrogen-like substance.[42] Pituitary cells from the rat that are normally stimulated to grow after estrogen exposure were found to grow after chlorpyrifos exposure.[43] This growth was blocked by a potent estrogen receptor

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[36] EPA. Human health risk assessment: Chlorpyrifos. June 8, 2000. p 131. emphasis is added.
[37] Meeker JD, Barr DB, Hauser R. 2006 Thyroid hormones in relation to urinary metabolites of non-persistent insecticides in men of reproductive age. Reprod Toxicol. 22(3):437-42.
[38] Ghisari M, Bonefeld-Jorgensen EC. 2005 Impact of environmental chemicals on the thyroid hormone function in pituitary rat GH3 cells. Mol Cell Endocrinol. 244(1-2):31-41
[39] Rawlings, N.C., Cook, S.J., Waldbillig, D., 1998. Effects of the pesticides carbofuran, chlorpyrifos, dimethoate, lindane, triallate, trifluralin, 2,4-d, and pentachlorophenol on the metabolic endocrine and reproductive endocrine system in ewes. J. Toxicol. Environ. Health A 54, 21–36.
[40] Pop VJ, Brouwers EP, Vader HL, Vulsma T, van Baar AL, de Vijlder JJ. 2003 Maternal hypothyroxinaemia during early pregnancy and subsequent child development: a 3-year follow-up study Clin Endocrinol 59(3):282-8.
[41] Haddow JE, Palomaki GE, Allan WC, Williams JR, Knight GJ, Gagnon J, O'Heir CE, Mitchell ML, Hermos RJ, Waisbren SE, Faix JD, Klein RZ. 1999 Maternal thyroid deficiency during pregnancy and subsequent neuropsychological development of the child. N Engl J Med. 341(8):549-55.
[42] Andersen, H.R., Vinggaard, A.M., Rasmussen, T.H., Gjermandsen, I.M., Bonefeld-Jorgensen, E.C., 2002. Effects of currently used pesticides in assays for estrogenicity, androgenicity, and aromatase activity in vitro. Toxicol. Appl. Pharmacol. 179, 1–12.
[43] Ghisari M, Bonefeld-Jorgensen EC. 2005 Impact of environmental chemicals on the thyroid hormone function in pituitary rat GH3 cells. Mol Cell Endocrinol. 244(1-2):31-41

antagonist, suggesting that chlorpyrifos stimulates the growth of these pituitary cells via the estrogen receptor and is an estrogen agonist. In human studies, exposure to chlorpyrifos has been shown to be associated with lower levels of testosterone, poorer sperm quality, and increased sperm DNA damage.[44, 45]

Gonadotropin-releasing hormone (GnRH) is a hormone released by the hypothalamus. It acts as a primary regulator of reproduction by controlling the release of luteinizing hormone and follicle stimulating hormone from the pituitary gland, thereby ultimately controlling androgen and estrogen levels. In experiments with a cell line model for GnRH neurons, exposure to chlorpyrifos was found to alter the biosynthesis of GnRH, potentially disrupting the entire hypothalamic-pituitary-gonadal axis.[46]

According to the IRED, EPA did not consider the endocrine disrupting effects of chlorpyrifos because the development of an Endocrine Disruptor Screening Program (EDSP) has not been completed. As a consequence, it neglects analyzing an entire category of potential adverse health effects. In fact, the risk assessment omits a group of studies that, taken together, suggest that chlorpyrifos may be an endocrine disrupting chemical, capable of interfering with multiple hormones controlling reproduction and neurodevelopment.

There is precedent for the Agency to consider endocrine disrupting effects in a human health risk assessment in the absence of a final EDSP. For example, in the RED for atrazine, the Agency examined the potential endocrine disrupting effects of atrazine on amphibians, undermining any agency claim that existing studies of the endocrine disrupting effects cannot be considered in its human health risk assessments. Accordingly, given the studies suggesting that chlorpyrifos has the potential to cause endocrine disrupting effects, EPA should have quantitatively incorporated these endpoints in its risk assessment of chlorpyrifos.

E. Cancer risks

The 2004 National Institutes of Health Agriculture Health Study, a very robust prospective epidemiology study of pesticide applicators in the Midwest, reported chlorpyrifos-specific findings that have been ignored by EPA despite their high relevance to the risk analyses and registration decisions. The incidence of lung cancer was statistically significantly associated with both chlorpyrifos lifetime exposure-days and chlorpyrifos intensity-weighted exposure days. After adjusting for other pesticide exposures and demographic factors, "individuals in the highest quartile of chlorpyrifos lifetime exposure-days (>56 days) had a relative risk of lung cancer of 2.18 (95%

[44] Meeker JD, Ryan L, Barr DB, Hauser R. Exposure to non-persistent insecticides and reproductive hormones in adult men. Epidemiology 2006;17:61–8.
[45] Meeker JD, Singh NP, Ryan L, et al. Urinary levels of insecticide metabolites and DNA damage in human sperm. Hum Reprod 2004;19:2573–80.
[46] Gore AC 2002 Organochlorine pesticides directly regulate gonadotropin-releasing hormone gene expression and biosynthesis in the GT1-7 hypothalamic cell line. Mol Cell Endocrinol. 192(1-2):157-70.

CI=1.31-3.64), significantly higher than those with no chlorpyrifos exposure."[47] These data were not referenced in the final aggregate assessment of chlorpyrifos or the OP CRA, but are highly relevant and so should have been.

F. Potential adverse effects below 10% cholinesterase inhibition

The OP CRA evaluated the cumulative toxicity of chlorpyrifos and its related organophosphate pesticides assuming that if the Agency regulated so as to allow no more than a 10% level of cholinesterase inhibition (10% ChEI) in the female adult rodent brain, this would be protective of all adverse effects at all life stages. That is, the Agency presumed that there are no other adverse effects that occur with doses lower than the dose eliciting a 10% ChEI in the female adult rodent brain. However, scientific studies published both prior to and since the IRED was completed in 2001 have reported that fetal and newborn exposure to chlorpyrifos affects diverse cellular functions by mechanisms of toxicity that are independent of cholinesterase inhibition. This is important because while the systemic toxicity that results from cholinesterase inhibition is reasonably well characterized, it does not explain why rodents exposed pre- and perinatally seem to recover from cholinesterase inhibition relatively rapidly, yet display persistent and more severe damage to the central nervous system.[48] Accumulating scientific evidence points to non-cholinergic mechanisms that disrupt multiple brain targets.[49] Many of these critical targets are vulnerable even at doses below those that elicit 10-20% cholinesterase inhibition. Some of the relevant studies are listed below:

- Scientists first reported in 1994, and then confirmed in 2001 that chlorpyrifos inhibited the production of the cellular second messenger Cyclic Adenosine Monophosphate (cAMP) in rat brain.[50] This has serious implications for many important cellular functions. For example, cAMP is required for normal function of hormones like glucagon (increases blood sugar levels) and adrenaline (regulates the stress response by increasing heart rate, elevating blood sugar, and depressing the immune system). cAMP is also required for regulating normal calcium movement in the body. Disruption of normal cAMP function may be associated with progression of some cancer types, including melanoma.[51,52]

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[47] Lee et al, Cancer Incidence Among Pesticide Applicators Exposed to Chlorpyrifos in the Agricultural Health Study, Journal of the National Cancer Institute, Vol 96, No. 23, December 1, 2004, p. 1781-9

[48] Slotkin TA, Cousins MM, Tate CA, Seidler FJ. Persistent cholinergic presynaptic deficits after neonatal chlorpyrifos exposure. Brain Res. 2001 Jun 1;902(2):229-43.

[49] Pope CN. Organophosphorus pesticides: do they all have the same mechanism of toxicity? J Toxicol Environ Health B Crit Rev. 1999 Apr Jun;2(2):161-81. Review.

[50] Huff RA, Corcoran JJ, Anderson JK, Abou-Donia MB. Chlorpyrifos oxon binds directly to muscarinic receptors and inhibits cAMP accumulation in rat striatum. J Pharmacol Exp Ther. 1994 Apr;269(1):329-35; Huff RA, Abu-Qare AW, Abou-Donia MB. Effects of sub-chronic in vivo chlorpyrifos exposure on muscarinic receptors and adenylate cyclase of rat striatum. Arch Toxicol. 2001 Oct;75(8):480-6.

[51] Dumaz N, Hayward R, Martin J, Ogilvie L, Hedley D, Curtin JA, Bastian BC,Springer C, Marais R. In Melanoma, RAS Mutations Are Accompanied by Switching Signaling from BRAF to CRAF and Disrupted Cyclic AMP Signaling. Cancer Res. 2006 Oct 1;66(19):9483-91.

[52] Abramovitch R, Tavor E, Jacob-Hirsch J, Zeira E, Amariglio N, Pappo O, Rechavi G, Galun E, Honigman A. A pivotal role of cyclic AMP-responsive element binding protein in tumor progression. Cancer Res. 2004 Feb 15;64(4):1338-46.

- Scientists reported in 2007 that in neonatal rats exposed to four daily doses of 1 mg/kg chlorpyrifos on days 1-4 after birth displayed life-stage and gender-specific alterations in the expression of genes important for nerve cell growth, cAMP-related cell signaling, programmed cell death (apoptosis), oxidative stress, and neurotransmitter synthesis. This dose and treatment regime is below the threshold dose that is associated with growth retardation and systemic toxicity and elicits less than 20% ChEI in exposed newborn rats.[53]
- In 2006, scientists reported that chlorpyrifos disrupted serotonin pathways in the developing rat brain at doses spanning the threshold for cholinesterase inhibition.[54] Interestingly, the study reported altered expression of transcription factors in both the forebrain (an area with many cholinergic neurons) and in the cerebellum (an area poorly innervated with cholinergic neurons), suggesting that there are severe impacts on non-cholinergic targets of chlorpyrifos in the brain, presumably through a non-cholinergic mechanism of toxicity.
- Scientists reported in 2006 an observed loss of non-cholinergic cerebellum neurons and permanent sensorimotor deficits in adult rodents exposed to chlorpyrifos *in utero*, demonstrating long-lasting effects from early life exposures to chlorpyrifos.[55] In this work, pregnant Sprague-Dawley rats were treated with 1.0 mg/kg daily dermal exposures to chlorpyrifos, and offspring were evaluated at 90 days after birth, corresponding to a human adult age. This study provides evidence that exposures during vulnerable windows of development can result in adverse impacts that extend into adulthood.
- In 2007, researchers reported that neonatal rats exposed to four daily doses of 1 mg/kg chlorpyrifos on days 1-4 after birth displayed regional alterations in the expression of the fibroblast growth factor family of genes across the brain and brain stem.[56] The proteins that are coded from these genes play critical roles in neural cell development, brain assembly and recovery from neuronal injury.

The broad spectrum of neurotoxic effects indicate that chlorpyrifos toxicity is far more complex than would be predicted if only its direct impairment of cholinesterase activity were considered.

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[53] Slotkin TA, Seidler, FJ. 2007. Comparative developmental neurotoxicity of organophosphates in vivo: Transcriptional responses of pathways for brain cell development, cell signaling, cytotoxicity and neurotransmitter systems. Brain Res Bull, May 30;72(4-6):232-74. Epub 2007 Jan 25.
Crumpton TL, Seidler FJ, Slotkin TA. Developmental neurotoxicity of chlorpyrifos in vivo and in vitro: effects on nuclear transcription factors involved in cell replication and differentiation. Brain Res. 2000 Feb 28;857(1-2):87-98.

[54] Slotkin TA, Tate CA, Ryde IT, Levin ED, Seidler FJ. Organophosphate insecticides target the serotonergic system in developing rat brain regions: disparate effects of diazinon and parathion at doses spanning the threshold for cholinesterase inhibition. Environ Health Perspect. 2006 Oct;114(10):1542-6

[55] Abou-Donia MB, Khan WA, Dechkovskaia AM, Goldstein LB, Bullman SL, Abdel-Rahman A. In utero exposure to nicotine and chlorpyrifos alone, and in combination produces persistent sensorimotor deficits and Purkinje neuron loss in the cerebellum of adult offspring rats. Arch Toxicol. 2006 Sep;80(9):620-31. Epub 2006 Feb 16.

[56] Slotkin TA, Seidler FJ, Fumagalli F. Exposure to organophosphates reduces the expression of neurotrophic factors in neonatal rat brain regions: similarities and differences in the effects of chlorpyrifos and diazinon on the fibroblast growth factor superfamily. Environ Health Perspect. 2007 Jun;115(6):909-16. Epub 2007 Feb 27.

A review published in 2003 by Duke University Professor Abou-Donia of OP poisoning incidents includes clinical reports of long-term impairment of cognitive and neurobehavioral performance associated with long-term exposure to the pesticides.[57] Permanent clinical symptoms that have been reported includes anxiety and deficits in learning, memory, and concentration. [58] In addition, individuals exposed to low, subclinical levels of chlorpyrifos have reported persistent long-term deficits in concentration, word finding, and short-term memory.[59] Two separate studies in 1996 and 1997 reported clinical cases of long-term cognitive and neuropsychological deficits in sheep dipper workers exposed to organophosphate pesticides.[60, 61] Dr. Abou-Donia suggests that the observed long-term effects are more likely to be a result of neuronal cell damage and death from apoptosis and oxidative stress, rather than from transient cholinesterase inhibition. [62]

Neither EPA's aggregate risk assessment (IRED) nor the OP CRA cite or quantitatively incorporate the results of the aforementioned laboratory studies and clinical reports. Without quantitatively incorporating low-dose risks of non-cholinergic effects, EPA's contention that the acute and chronic dietary point of departures (BMD10) are protective is unproven and is likely to underestimate significantly the long-lasting impairments resulting from early life exposure to chlorpyrifos.

EPA ought to heed experts who warned: "the fact that alterations in neurodevelopment occur with organophosphate exposures below the threshold for cholinesterase inhibition reinforces the inadequacy of this biomarker [cholinesterase inhibition] for assessing exposure or outcome related to developmental neurotoxicity."[63] EPA's own Scientific Advisory Panel (SAP) in 2002 had raised the same concern, stating "reliance on a single biochemical assay to measure brain damage may become problematic."[64] Accordingly, the Agency must consider non-cholinergic neurotoxicity in the CRA and IRED assessments when establishing the safe level (RfD) and allowable commodity tolerances. Taking into consideration the full toxicity spectrum of chlorpyrifos will lead to the scientifically-defensible conclusion that it is too dangerous to be reregistered.

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[57] Abou-Donia, MB. Organophosphorus ester-induced chronic neurotoxicity. Arch Environ Health, 2003; 58(8): 484-497

[58] *Id.*

[59] Kaplan JG, Kessler J, Rosenberg N et al. Sensory neuropathy associated with Dursban (chlorpyrifos) exposure. Neurology 1993; 43:2193-2196

[60] Beach JR, Spurgeon A, Stephens R, et al. Abnormalities on neurological examination among sheep farmers exposed to organophosphate pesticides. Occup Environ Med, 1996; 53(8): 520-525

[61] London L, Myers JE, Neil V, et al. An investigation into neurological and neurobehavioral effects of long-term agrochemical use among deciduous fruit farm workers in the Western Cape, South Africa. Environ Res, 1997; 73(1-2):132-145

[62] Abou-Donia, MB. Organophosphorus ester-induced chronic neurotoxicity. Arch Environ Health, 2003; 58(8): 484-497

[63] Slotkin TA, Tate CA, Ryde IT, Levin ED, Seidler FJ. Organophosphate insecticides target the serotonergic system in developing rat brain regions: disparate effects of diazinon and parathion at doses spanning the threshold for cholinesterase inhibition. Environ Health Perspect. 2006 Oct;114(10):1542-6.

[64] Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held June 26-27, 2002. Released on July 19, 2002, 26.

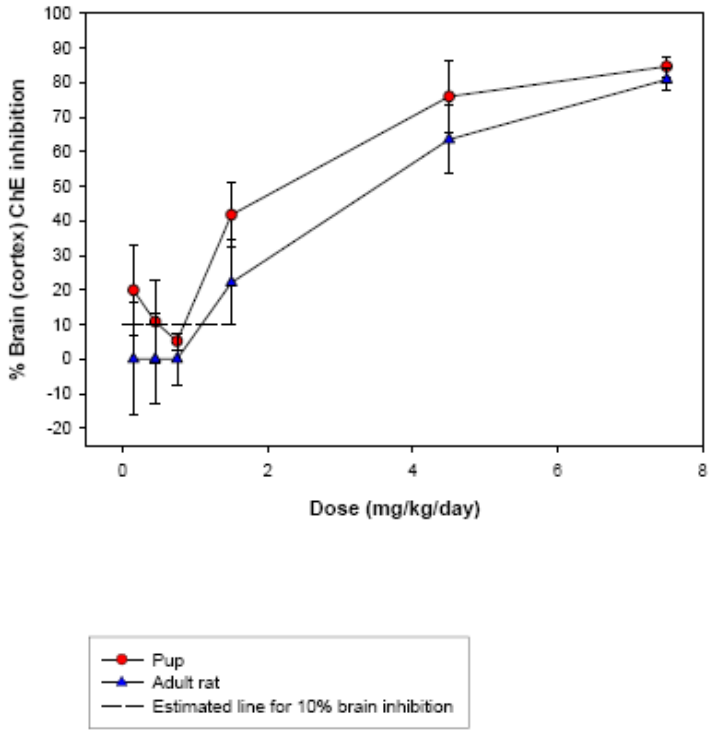
III. CRA Misrepresents Risks, Fails to Apply FQPA

The CRA failed to apply any FQPA factor to adjust for early life exposures, citing a 2000 study that EPA interprets to show no difference in response between pups and adult rats at the dose estimated to result in 10% inhibition.[65]

In addition to relying on limited data, EPA resorted to inaccurate interpretations of that data to support its decisions. EPA approached the determination of an FQPA factor by screening for data "which measured brain cholinesterase inhibition in juvenile and adult rats following repeat dosing."[66] For all organophosphate pesticides *except* chlorpyrifos, EPA then determined a benchmark dose. However, for chlorpyrifos, EPA used data from a paper by Zheng et al.[67] authored and provided by FIFRA SAP member Carey Pope, to identify a 10% brain cholinesterase inhibition point.[68] EPA relied solely on this one study to eliminate the FQPA factor for repeat exposures, stating that "at this dose, there is no difference in response between pups and adult rats." However, review of these data in both the original published manuscript, and as presented in the cumulative risk assessment, shows that there is an obvious difference between juvenile and adult responses to chlorpyrifos. (See Figure 1, below.)

[65] *Id.*
[66] Organophosphorus Cumulative risk assessment – 2006 Update, available at < http://www.epa.gov/pesticides/cumulative/2006-op/index.htm>, 59.
[67] Zheng Q, Olivier K, Won YK, Pope CN. 2000. Comparative cholinergic neurotoxicity of oral chlorpyrifos exposures in preweaning and adult rats. Toxicological Sciences, 55(1): 124-132
[68] Oklahoma State University, Fig I.B-3, Cumulative Risk Assessment at 63

Figure I.B-3 Plot of chlorpyrifos data from Zheng et al (2000).



In fact, Zheng et al. report that neonates are more sensitive than adults to chlorpyrifos associated ChEI.

First, the authors observed that after acute chlorpyrifos exposure, neonates were much more sensitive than adults: "Following acute CPF [chlorpyrifos] exposure, more extensive ChE [cholinesterase] inhibition was noted in neonates than in adults (especially in the brain) with NOELs based on ChE inhibition in adult tissues being 1 to ≥10-fold higher than in neonates."[69] These results are consistent with many other reports in the scientific literature: "It is apparent from a number of studies that neonatal rats are more sensitive to acute toxicity following either oral or subcutaneous acute high dosages of CPF (Atterberry et al, 1997; Moser and Padilla, 1998; Pope and Chakraborti, 1992; Pope et al, 1991)." They also note that signs of toxicity and lethality generally develop several hours, rather than immediately, after an acute exposure to chlorpyrifos.

The authors also reported that neonates were more sensitive than adults following repeat exposure scenarios: "With repeat exposures, NOELs based on ChE inhibition in adults were only 0.2 - 2-fold higher than in neonates." However, using the endpoint of body

[69] Zheng Q, Olivier K, Won YK, Pope CN. 2000. Comparative cholinergic neurotoxicity of oral chlorpyrifos exposures in preweaning and adult rats. Toxicological Sciences, 55(1): 124-132

weight changes following repeat doses, the authors noted that "the NOEL for adults was 5-fold higher than for neonates."[70]

EPA has mischaracterized these data. Rather, these data support using a 10X FQPA factor based on acute exposures using brain cholinesterase endpoints, a 2X FQPA factor based on repeat exposures using brain cholinesterase endpoints, and a 5X FQPA factor based on repeat exposure using body weight endpoints. EPA has presented an incomplete and therefore inaccurate interpretation of these data to support for its decision to remove the FQPA factor altogether.

IV. Over-Reliance on Registrant Data

Chlorpyrifos is one of the most studied of all the organophosphate pesticides. And, as demonstrated above, all the evidence of adverse health effects arising from the exposure to chlorpyrifos supports banning all uses of chlorpyrifos and revoking all food tolerances. Yet, despite this plethora of publicly-available data, the Agency cherry picked the data, ignoring robust, peer-reviewed data in favor of weak, industry-sponsored data to determine that chlorpyrifos could be re-registered and food tolerances be retained. EPA's re-registration and tolerance reassessment decision is not scientifically defensible because it is based on a strained and biased interpretation of an incomplete data set.

As with all scientific inquiry, greater confidence is ascribed to results of studies that are repeatable, supplied by multiple lines of evidence, and drawn from multiple, well-designed, well-conducted studies of adequate statistical power. To that end, all of the studies identified in this petition are published and publicly-available in peer-reviewed scientific literature, indicating that they were subject to public and professional scrutiny and are therefore likely to be reliable. These data showing adverse impacts of chlorpyrifos and other organophosphate pesticides on fetal and childhood development from non-cholinergic effects satisfy all three prongs for strong scientific validity because they a) arise from multiple laboratories (independent lines of evidence), b) are based on studies *in vitro*, in whole animals, and in humans (multiple lines of evidence), and c) show agreement across studies regarding the reported adverse outcomes (repeatability) and the mechanisms of action (biological plausibility). These data fulfill the scientific criteria for establishing causality, highlighting the breadth of robust data available to, yet ignored by, the Agency regarding chlorpyrifos.

Where EPA should have relied on its strongest scientific evidence, it led off with its weaker database and relied on the odd claim of scant organophosphate data to justify its decision not to refine the intra-species factor. More egregiously, despite having data on chlorpyrifos, the Agency chose to ignore that data and retain a weak intra-species factor for chlorpyrifos. As illustrated by the PON1 study discussed in the previous section, the Agency chose to ignore strong evidence of harm at doses below those that inhibit cholinesterase, despite evidence of susceptibility in exposed children.

[70] Zheng Q, Olivier K, Won YK, Pope CN. 2000. Comparative cholinergic neurotoxicity of oral chlorpyrifos exposures in preweaning and adult rats. Toxicological Sciences, 55(1): 124-132

V. EPA Failed to Incorporate Inhalation Routes of Exposure

In its aggregate assessment, EPA considered exposures from food, drinking water, and residential uses of chlorpyrifos. However, for some populations that include children and pregnant women, inhalation of chlorpyrifos-contaminated air may be one, if not the most, significant source of chlorpyrifos exposure. Although EPA was advised of these public data prior to 2006, it failed to incorporate quantitatively this scientific evidence of air exposures into the aggregate assessment.[71]

Available monitoring data show that for volatile and semi-volatile pesticides (vapor pressure > 10^{-7} mm Hg at 20-25°C), post-application drift typically accounts for 80-95% of the total off-site airborne pesticide movement. Chlorpyrifos falls solidly into this category of pesticides, with a vapor pressure of 10^{-5} mm Hg. Air monitoring studies conducted by the California Air Resources Board (ARB) and by communities working with PANNA indicate that post-application volatilization typically peaks between two and 24 hours after the start of an application for volatile and semi-volatile pesticides and may persist for days above levels of concern. ARB published its work on air monitoring for chlorpyrifos in 1998.[72] PANNA published its chlorpyrifos air monitoring results for Lindsay, California in July 2006, before the finalization of the OP CRA.

A. State of California Data Documents Air Contamination

The California ARB has documented widespread presence of chlorpyrifos in the air using both near-field and ambient air monitoring.

1. Near-Field Monitoring

The California ARB measured air concentrations of chlorpyrifos near an orange grove treated with chlorpyrifos, with the application taking place during two separate events separated by a day.[73] Three-day, time-weighted average concentrations at the monitoring stations ranged from 5,312 to 8,112 ng/m^3 (depending on the location of the monitoring station). See Figure 1. Translation of these concentrations into Reference Exposure Levels (RELs) that take into account breathing rate and body weight indicated that these concentrations exceeded the acute 24-hour REL for a one-year-old child by a factor of 31

[71] PANNA provided EPA with the results of the ARB monitoring demonstrating problematic exposure from volatilization drift for multiple pesticides on several occasions, including in several formal comment letters to EPA on molinate (Docket ID # OPP-34232, included here by reference), several legal petitions,[71] in comments submitted to US EPA for the OP CRA docket in October of 2006 (Docket ID # EPA-HQ-OPP-2006-0618), and in a presentation to EPA staff (EFED and HED) on May 9, 2002. PANNA published a report presenting and analyzing the ARB data in May of 2003. S.E. Kegley, A. Katten, and M. Moses, *Secondhand Pesticides: Airborne Pesticide Drift in California*, Californians for Pesticide Reform (San Francisco, CA 2003),

[72] *Report for the Application and Ambient Air Monitoring of Chlorpyrifos (and the Oxon Analogue) in Tulare County during Spring/Summer 1996*, California Air Resources Board, Test Report #C96-040 and # C96-041, April 7, 1998, http://www.cdpr.ca.gov/docs/empm/pubs/tac/chlrpfs.htm.

[73] *Report for the Application and Ambient Air Monitoring of Chlorpyrifos (and the Oxon Analogue) in Tulare County during Spring/Summer 1996*, California Air Resources Board, Test Report #C96-040 and # C96-041, April 7, 1998, http://www.cdpr.ca.gov/docs/empm/pubs/tac/chlrpfs.htm.

to 48 and the acute 24-hour REL for adults by a factor of 1.4 to 2.1.[74] Concentrations of chlorpyrifos were still above both the adult and child RELs at the downwind site at the end of the monitoring period, at 4,900 ng/m^3 (29 times the child REL and 1.3 times the adult REL). These data indicate that those who live, work, or go to school near application sites risk acute nervous system toxicity from airborne exposure to this pesticide. The developing fetus, infants and children are especially at risk because their nervous systems are still developing.

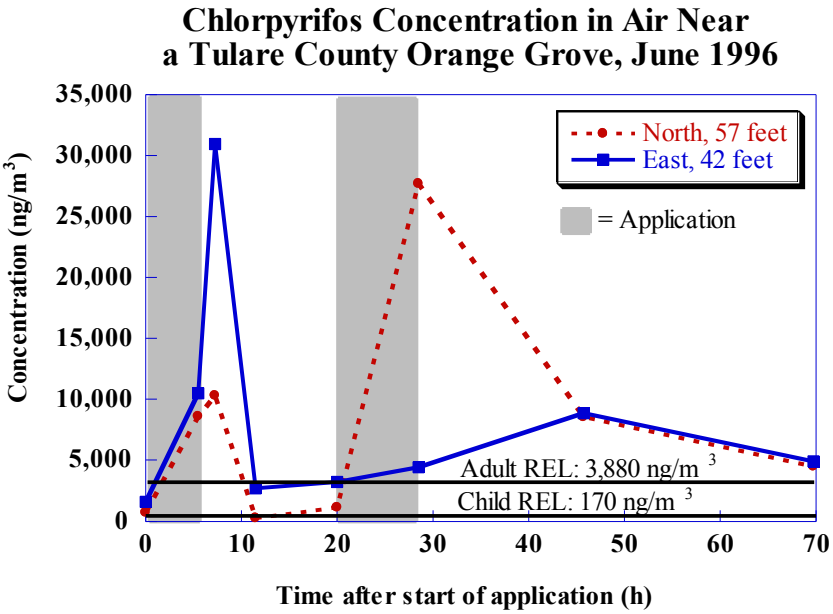


Figure 1: Chlorpyrifos air concentrations peaked approximately 2.5 hours after the end of the first application and again during the second application. Substantial volatilization continued for several days after application and exceeded 24-hour RELs for both adults and children for much of the sampling period.

ARB only conducted a single application site monitoring study for chlorpyrifos; however, the fact that the application occurred in two distinct time periods provides essentially two applications in one study. The similar peak concentrations observed for the two

[74] In order to compare observed concentrations of chlorpyrifos in air with concentrations likely to be associated with adverse effects, the US EPA inhalation NOAELs for acute and sub-chronic exposures to chlorpyrifos of 0.1 mg/kg-day (based on plasma and red blood cell cholinesterase inhibition)74 were used to calculate Reference Exposure Levels (RELs) for a sensitive receptor, a one-year-old infant weighing 7.6 kg, breathing on average 4.5 m^3 of air per day. This calculation takes into account the 10-fold intraspecies, 10-fold interspecies and 10-fold FQPA uncertainty factors used by US EPA for chlorpyrifos.

$$\text{Acute REL (ng/m}^3\text{)} = \frac{\text{Inhalation NOEL (mg/kg-day)} \times 10^6\,\text{ng/mg} \times \text{body wt. (kg)}}{(UF_{inter} \times UF_{intra} \times UF_{FQPA}) \times \text{breathing rate (m}^3\text{/day)}} = \frac{0.1\ \text{mg/kg-day} \times 10^6\,\text{ng/mg} \times 7.6\ \text{kg}}{(10 \times 10 \times 10) \times 4.5\ \text{m}^3\text{/day}} = 170\ \text{ng/m}^3$$

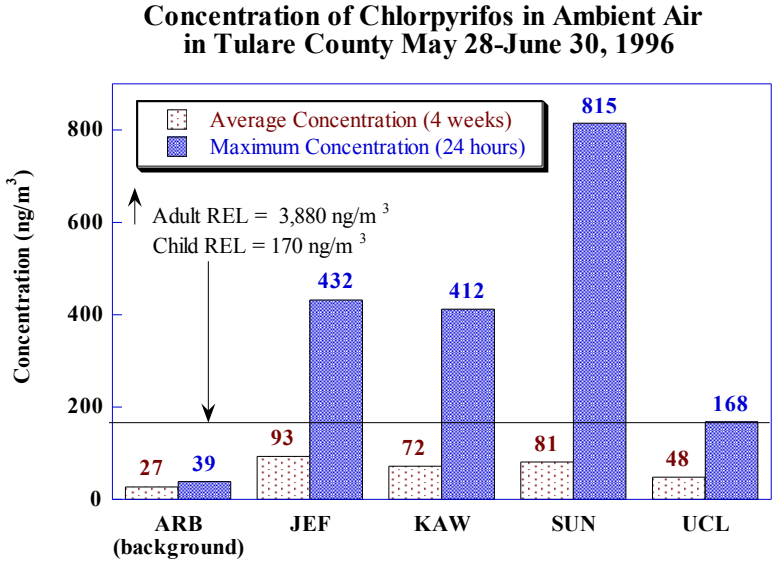
The calculated concentration is the equivalent of a concentration in air below which no adverse effects on cholinesterase inhibition are anticipated by US EPA. Note, however, that the developmental neurotoxicity observed for chlorpyrifos (see Section 1 above) is not mediated by cholinesterase inhibition and may occur at lower doses.

applications under different wind conditions (30,950 ng/m^3 vs. 27,700 ng/m^3) suggest that peak air concentrations may be quite predictable based on the vapor pressure of the pesticide, a fact consistent with other work in the peer-reviewed literature.[75]

The breakdown product chlorpyrifos oxon was observed in 100% of the samples, but the toxicity of this substance was not taken into account in this analysis because no RELs are available for comparison. However, because the oxon is more acutely toxic than the parent compound, neurotoxic effects associated with breathing air contaminated with both chlorpyrifos and its oxon at the measured levels will be greater than chlorpyrifos concentrations alone would suggest.

2. Ambient Monitoring

During the summer of 1996, the ARB sampled seasonal concentrations of chlorpyrifos in ambient air in Tulare County, California by placing monitoring stations on several schools that were somewhat distant from direct applications but located in regions of high use.[76] Monitoring occurred over the course of four and a half weeks, which serves as an estimate of sub-chronic exposure. Average concentrations over the full time frame of the monitoring study were below both adult and child sub-chronic RELs, averaging 38% of the one-year-old child REL over all sites. See Figure 2. The maximum measured 24-hour concentrations equaled or exceeded the 24-hour acute child REL at four of the five monitoring sites and ranged from 23% to 485% of the 24-hour acute child REL. The monitoring report was published by ARB in 1998, but was not incorporated into EPA's aggregate assessment.



[75] JE Woodrow, JN Seiber, LW Baker, Correlation Techniques for Estimating Pesticide Volatilization Flux and Downwind Concentrations, *Envi. Sci. Tech.*, **1997,** 31: 523-529.

[76] *Report for the Application and Ambient Air Monitoring of Chlorpyrifos (and the Oxon Analogue) in Tulare County during Spring/Summer 1996,* California Air Resources Board, Test Report #C96-040 and # C96-041, April 7, 1998, http://www.cdpr.ca.gov/docs/empm/pubs/tac/chlrpfs.htm.

Figure 2: Chlorpyrifos concentrations in air in Tulare County, CA in Summer 1996 measured by the CA ARB. Averages are for 4 days per week of sampling over the 4-week period. Monitoring sites included ARB, the ARB office in downtown Visalia; JEF, Jefferson Elementary School in Lindsay; KAW, Kaweah School in Exeter; SUN, Sunnyside Union Elementary School in Strathmore; UCL, University of California, Lindcove Field Station.

Using these ARB data, scientists at the California Department of Health Services concluded in a peer-reviewed paper in 2002 that short-term chlorpyrifos exposure estimates exceeded the acute REL for 50% of children in the exposed general populations.[77] The researchers noted that farm workers and their children likely experience higher exposures and risks than individuals in the general population. Furthermore, "[p]esticide exposures and risks are characterized for the communities around the air monitoring locations. However, the potential for exposures in other residential areas clearly exist . . ." In addition, the authors indicate that census data suggest "a potential for exposures and risks, similar to those calculated in this risk assessment, for hundreds of thousands of people in California." [78]

B. Community Air Monitoring Shows Widespread Contamination

Since 2004, PANNA has been working with rural communities to conduct air monitoring at people's homes, schools and workplaces.[79] Chlorpyrifos is one of the primary pesticides that has been found in these communities. Data collected in Lindsay, California in June and July of 2004, 2005, and 2006, and in Washington State in 2006 demonstrate that daily exposure to chlorpyrifos can be substantial, and regularly exceeds the "acceptable" 24-hour acute dose for a one-year-old child established by the EPA. This information has been transmitted to EPA staff through personal communications with staff, presentations at public meetings, and in Spray Drift Work Group meetings. The 2004 and 2005 results from the Lindsay, California study were published on July 14, 2006.[80]

Of the 104 samples collected in Lindsay, California during the summer of 2004, 11% were above the 24-hour acute and sub-chronic child REL. The highest concentration observed for a 24-hour period was 1,340 ng/m^3 (7.9 times the 24-hour acute child REL). Of the 108 samples in the same area during the next summer (2005), 23% were above the 24-hour acute and sub-chronic child REL. The highest concentration observed for a 24-hour period in 2005 was 1,120 ng/m^3 (6.6 times the 24-hour acute child REL). These data are consistent with results obtained by the ARB for ambient air monitoring conducted in

[77] S. Lee, R. McLaughlin, M. Harnly, *et al.*, Community exposures to airborne agricultural pesticides in California: Ranking of Inhalation Risks, *Env Health Persp*, 2002, 110: 1175–84.
[78] S. Lee, R. McLaughlin, M. Harnly, *et al.*, Community exposures to airborne agricultural pesticides in California: Ranking of Inhalation Risks, *Env Health Persp*, 2002, 110: 1175–84.
[79] *Drift Catcher Results*, Pesticide Action Network, www.panna.org/campaigns/driftCatcherResults.html
[80] K Mills and SE Kegley, *Air Monitoring for Chlorpyrifos in Lindsay, California, June-July 2004 and July-August, 2005*, Pesticide Action Network North America (San Francisco, CA, July 14, 2006).

1996 (see above).

Although the observed 24-hour average concentrations were below the adult RELs, adults living in the houses where the monitoring stations were located experienced symptoms of acute OP poisoning. This observation suggests the following: 1) the NOELs EPA determined from industry toxicology studies are inaccurate and do not reflect the true toxicological endpoints; and/or 2) using a 24-hour averaging time does not protect people from poisoning resulting from shorter-term exposures at higher concentrations. In any case, it is clear that inhalation exposure is high enough to cause acute poisonings of bystanders and that EPA's failure to account for inhalation exposures in its aggregate risk assessment is a serious flaw in the risk assessment process.

C. Inhalation Exposure to Chlorpyrifos Far Exceeds Dietary Exposure

In areas of high chlorpyrifos use, inhalation is the primary source of exposure, dwarfing all other sources. A comparison of dietary exposure estimated by EPA for the most-exposed (99.9th percentile) children to inhalation exposure reported by ARB and PANNA from measurements in several different locations and seasons is illuminating.

The highest acute dietary exposures for infants are estimated by EPA to result in a dose that is 50% of the acute Population Adjusted Dose (PAD). In contrast, inhalation exposures estimated from ARB monitoring data indicate that infants living very close to an application site during the day the application takes place are exposed to a dose that is over 75 times higher than the acute PAD. The ambient air monitoring conducted in Lindsay, California and the Yakima Valley in Washington State[81] indicate that the highest 24-hour exposures (comparable to the 99.9th percentile acute dietary exposure) would result in a dose that ranges from 404–793% of the acute PAD. These data show that EPA is failing to account for the vast majority of exposure when it assumes inhalation exposure is zero for rural residents in areas of high chlorpyrifos use.

VI. Exporting Hazards

Unless chlorpyrifos is banned, and all tolerances cancelled, chlorpyrifos will continue to be used, often unsafely, in other countries thus creating a health and environmental hazard in those countries and on contaminated food re-entering the US. Although chlorpyrifos is listed as a "restricted use" pesticide in the US, it is exported in high volume: 7 to 9 million pounds annually since 1997 (8,570,694 in 2000).[82] Between 1997 and 2000, nearly 65 million pounds of severely restricted or forbidden pesticides in the US were exported; more than 22 tons per day – and more than half were exported to

[81] C Dansereau, SE Kegley, K Tupper, A Wang and M. Perez, *Poisons on the Wind: Community Air Monitoring for Chlorpyrifos in the Yakima Valley*, Farm Worker Pesticide Project and Pesticide Action Network North America (San Francisco, CA December 2006).

[82] Smith, C. 2001. Pesticide exports from U.S. ports, 1997-2000. *Int J Occ Environ Health*, 7(4): 266-274. Table 6, data from California EPA.

developing countries for agriculture use.[83] The International Labor Organization estimates that 60 to 90% of children estimated to be working in Africa (80 million), Asia (152 million), and Latin America (17 million) work in agriculture. These children are exposed to toxic pesticides in the fields, from drinking and washing water, through contaminated clothing, and in their homes.[84] The U.N. Commission on Human Rights stated that "[a]llowing the export of products recognized to be harmful is immoral."[85] The mitigation requirements in this IRED include respirators with an organic-vapor removing cartridge and a pesticide-approved prefilter, chemical-resistant outer-clothes, enclosed-cab machinery, emergency equipment readily available, and storage containments for discarding single-use chemically-resistant over-clothes. It is inconceivable that these are "readily available" to mixers, loaders, applicators, and fieldworkers in developing countries. US labeling requirements will have no mitigation effects for these men, women, and children workers. Cancellation of these dangerous pesticides is the most prudent and health-protective solution.

VII. Conclusion

Just a few months prior to the August, 2006 release of the CRA, the Local Presidents of EPA Unions representing scientists, risk managers, and related staff took the unusual step of sending a letter to Administrator Johnson expressing significant concerns about the EPA's risk analyses for organophosphates and identifying undue influence of pesticide registrants on its decision-making processes for these pesticides.[86] Particular concerns raised by the EPA Union leaders included the failure of EPA adequately to address exposures to infants and children who live near treated fields, including the children of farm workers. Moreover, the letter alerted Administrator Johnson that Pesticide Program staff "feel besieged by political pressure exerted by Agency officials perceived to be too closely aligned with the pesticide industry and former EPA officials now representing the pesticide and agricultural community; and by the USDA…."[87] The letter concluded that "until EPA can state with scientific confidence that these pesticides will not harm the neurological development of our nation's born and unborn children, there is no justification to continue to approve the use of the remaining OP [organophosphate] and carbamate pesticides."[88]

Separately, NRDC also voiced serious concerns about the limitations of the data set used by EPA for the aggregate and cumulative assessments.[89] Many of these concerns were discussed at length by the FIFRA SAP and reported in 2002. Two members of the panel "felt strongly that the studies presented by the Agency have limited application to

[83] article by C. Smith according to customs records
[84] US Newswire. 2001. U.N. human rights investigator deems U.S. export of banned pesticides 'immoral'. December 17, 16 :09. http://www.usnewswire.com
[85] U.N. Special Rapporteur Fatma Zora Ouhachi-Vesely. In : US newswire, December 17, 2001. op cit.
[86] Union Letter to EPA Administrator. May 24, 2006 http://www.nrdc.org/media/docs/060525.pdf
[87] Union letter at 3
[88] Union letter at 3
[89] NRDC comments on the Revised Cumulative Risk Assessment of the Organophosphate Pesticides. Docket OPP-2002-0230. April 28, 2002

understanding the effects of OP insecticides, specifically in children."[90] The SAP was also concerned about the failure to fully incorporate pre- and post-natal effects of organophosphates associated with children's brain function. The SAP reported that "[n]ot to include data on these outcomes excludes important variables in the assessment and therefore introduces important specification error. Wilson's work and the work of many others have shown that *systematically measured behavior may demonstrate toxicological effects at lower doses than those that yield phenotypic or biochemical alterations*."[91] Significantly, the SAP concluded that EPA's assessment contained "substantial measurement and specification errors, and as a consequence, underestimates the risk of OPs for child health."[92] In its final determinations, EPA failed to acknowledge these important limitations and chose not to adjust the uncertainty factors.

Without incorporating published literature describing the chronic impacts of long-term, low-level doses of organophosphate pesticides, particularly early-life exposures, EPA is making critical decisions about chlorpyrifos based on only a fragment of the whole story. Together with the decision to ignore robust data, this approach of deliberately selecting for the weakest data dumbs down the Agency's registration decision to the lowest common denominator.

Robust data shows that any use restriction on chlorpyrifos would still not be health-protective and that all food tolerances must be revoked. EPA's decision to reregister chlorpyrifos and retain food tolerances violates FIFRA and the FFDCA. EPA failed to consider important studies and improperly disregarded others. Furthermore, the Agency relied on a biased selection of available, weak data, in favor of the robust data, leading to an unsupported risk assessment.

As a result of EPA's actions, NRDC and PANNA members and their children are being exposed to unsafe levels of chlorpyrifos, and will continue to be as long as the chlorpyrifos registrations and food tolerances challenged in this petition remain in effect. We therefore request that EPA expedite its consideration of this petition in every way possible. If EPA intends to solicit public comment before making a decision on this petition, we request that the Agency do so promptly. EPA's past history of significant delay in responding to pesticide petitions and tolerance objections filed by NRDC constitutes a pattern and practice of unlawful agency inaction that harms NRDC and PANNA and its members.

Based on all of the foregoing comments, NRDC and PANNA petition EPA to revoke all tolerances and cancel all registrations for the pesticide chlorpyrifos. We reserve the right to supplement this petition based on new information.

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[90] Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held June 26-27, 2002. Released on July 19, 2002, 26.
[91] *Id* (emphasis is added).
[92] Transmittal of Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held June 26-27, 2002. Released on July 19, 2002 (emphasis is added).

Respectfully submitted,

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Dated: 12 September 2007

cc: Administrator Stephen Johnson
General Counsel Roger Martella
James Gulliford
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ATTACHMENT 2

January 14, 2002

Public Information and Records Integrity Branch
Information Resources and Services Division (7502C)
Office of Pesticide Programs
Environmental Protection Agency
1200 Pennsylvania Ave., N.W.
Washington, DC 20460

Docket Number: OPP-34203G

Submitted by Email: opp-docket@epa.gov

RE: **Comments on the Chlorpyrifos Interim Reregistration Eligibility Decision Document (IRED)**

Federal Register: November 14, 2001; Volume 66, Number 220, Pages 57073-57074

Dear Sir or Madam:

We submit the following comments on behalf of the Natural Resources Defense Council (NRDC), the World Wildlife Fund (WWF), and the Farmworker Justice Fund, Inc. NRDC uses law, science, and the support of more than 500,000 members nationwide to protect the planet's wildlife and wild places and to ensure a safe and healthy environment for all living things. NRDC has no direct or indirect financial or fiduciary interest in the manufacture or sale of chlorpyrifos. WWF is a non-profit organization with over 1.2 million members in the United States. WWF is dedicated to using the best available scientific knowledge to preserve the diversity and abundance of life on Earth by conserving endangered spaces, safeguarding endangered species, and addressing global threats to the planet's web of life. The Farmworker Justice Fund, Inc. is a national, nonprofit advocacy center that seeks to improve living and working conditions for migrant and seasonal farmworkers and their families. For the past two decades, it has urged the EPA to reduce or eliminate the use of hazardous pesticides.

Unless otherwise noted or referenced, all page number references in the text of these comments refer to the Interim Reregistration Eligibility Decision (IRED) for Chlorpyrifos, Case No. 0100.

SUMMARY OF COMMENTS:

ALL CHLORPYRIFOS TOLERANCES MUST BE REVOKED & CHLORPYRIFOS MUST BE CANCELLED

EPA has found that there is ***not*** a "no observable effect level" (NOEL), or even a "no observable adverse effect level" (NOAEL), for the developmental effects of chlorpyrifos on young animals, and therefore on human infants and children. In other words, EPA has no scientific basis upon which to conclude that there is a fully safe level at which infants and children will not suffer developmental harm due to chlorpyrifos exposure. Moreover, EPA has no basis to derive a Q* or to use any other quantitative risk assessment methodology that will derive a negligible risk level (i.e. 1 in 1 million lifetime risk) for chlorpyrifos' developmental effects. Therefore, EPA simply cannot make a legal finding that any specific chlorpyrifos level on food is "safe" for infants and children, or that there is a "reasonable certainty of no harm" to infants and children, at any specific level. Thus, as a legal matter, under FFDCA §408(b)(2), EPA must revoke all tolerances and cancel all food uses, for chlorpyrifos.

Moreover, even assuming for the sake of argument that EPA may set tolerances for chlorpyrifos, notwithstanding the legal prohibition on EPA's setting a tolerance for a substance that has no NOEL or NOAEL (and for which a Q* cannot be derived and therefore a cancer-like risk assessment is impossible), EPA has failed to adequately apply safety factors and to consider many routes of exposure in the IRED. EPA must apply not only the traditional two 10-fold safety factors for interspecies variability and for intra-species variability, but also must apply an additional safety factor of at least 10 for gaps in exposure data, assumptions, and underestimates of risk, as detailed in these comments, and an FQPA safety factor of at least 30 to account for potential pre-and post-natal ***toxicity*** to infants and children (10X) and to account for significant gaps in ***exposure data*** for infants and children and the lack of a NOEL (3X). EPA has failed to adequately consider important exposure routes for millions of infants and children, including children living on farms and who accompany their parents into farm fields (see discussion of farm children below), exposure from mosquito spraying, drift, and drinking water. Moreover, EPA has failed to consider cumulative exposure to organophosphates and carbamates that have a common mechanism of toxicity.

Finally, EPA must cancel all uses of chlorpyrifos under FIFRA. All food uses must be cancelled due to excessive risk and the cancellation of all tolerances under the FFDCA. Even if theoretically an extremely low tolerance could be set for chlorpyrifos after application of all appropriate safety factors, the chemical would have to be applied at such low rates and in such a manner that it would not be efficacious. Moreover, the food and non-food uses of chlorpyrifos result in worker exposure; non-occupational drift, air, and water exposure; other non-food human exposure; and ecological risks that pose "unreasonable adverse effects on the environment" and thus must result in cancellation of all uses of chlorpyrifos.

INTRODUCTION

Uses and Risks of Chlorpyrifos

Chlorpyrifos is an organophosphate pesticide used at approximately 21-24 million pounds active ingredient (a.i.) annually in the United States. In addition to food crops, chlorpyrifos is used as a cattle eartag, as a treatment for lawns, turf, and ornamentals, and for pasture, woodland, and lots/farmsteads. Turf use accounts for 2.5 million pounds annually. By agreement with the registrants (June, 2000)[i], almost all the residential uses (including household, schools, parks) of chlorpyrifos are being cancelled; public health uses such as mosquito and "childproof" ant-and-roach bait are residential uses that remain eligible for reregistration (IRED p.8). Termite control accounted for 5 million pounds annually, and is slated to be withdrawn or phased-out (prohibited Dec. 31, 2005; IRED p.7). These voluntary phase-outs are predicted to eliminate almost half the annual chlorpyrifos use in the United States (IRED p.12). Prior to these phase-outs, chlorpyrifos was the fourth most commonly used pesticide in US households, and comprised a full 25% of pesticide pounds in residential settings. However, these voluntary phase-outs and withdrawals in the US will not reduce the amount of chlorpyrifos exported annually, which has remained steady at approximately 8-9 million pounds annually since 1997.[ii]

Non-agricultural outdoor uses remaining include golf courses (rate of application reduction), road medians, food processing plants, manufacturing plants, ship holds, railbox cars, and non-structural wood treatments such as fenceposts, utility poles, landscape timber, poles, posts, and processed wood products (IRED p.10). NRDC believes that many of these remaining uses, especially wood treatments, pose significant exposure risks to children.

Chlorpyrifos is used in highest amounts on corn (5.53 million lbs a.i.), cotton (670 thousand), apples (550 thousand), alfalfa (480 thousand), oranges (460 thousand), and peanuts (316 thousand). Crops with the highest percentage of crop treated are Brussels sprouts (73%), cranberries (46%), apples (44%), broccoli (41%), and cauliflower (31%). This list includes some of the foods most consumed by children, making children's exposure risk particularly high.[iii] Chlorpyrifos is applied aerially, by chemigation, groundboom, hand wand, airblast sprayer, and other methods.

The EPA risk summary speculates that dietary risk, acute and chronic, would now be below the Agency's level of concern, presuming full compliance with risk mitigation recommendations. Prior to mitigation, EPA admits that acute exposures exceed the level of concern for infants, toddlers, children, and females. After mitigation, EPA believes that these risk groups no longer exceed the level of concern (IRED p.19).

However, there are considerable risks remaining for workers, even presuming full compliance with so-called "mitigation measures." For mixers, loaders, and applicators some risks remain elevated, even assuming that maximum personal protection equipment (PPE) and engineering controls are used. In real life, however, maximum recommended PPE is often not used. Under many scenarios current restricted entry intervals (REIs) also are insufficient.[iv] The IRED notes that risks may be mitigated by a combination of additional PPE and engineering controls, and by reductions in application rates (IRED p.3). Again, past experience shows that recommended application rates, along with recommended PPE controls, are often disregarded. Post application risks, also exceeding the level of concern, may be mitigated by reducing application rates and by lengthening REIs. Again, this is unlikely to be sufficient to protect worker risk.

Furthermore, in measuring the extent of exposure and in determining aggregate exposure, EPA should acknowledge farmworker children to be a major, identifiable subgroup of consumers whose unique, increased level of exposures must be taken into account. These nearly 1,000,000 children are deserving of protection under the "reasonable certainty of no harm" health standard under the law.

Although EPA says the drinking water risk is below the level of concern, the Agency notes that there have been cases of high levels of drinking water well contamination associated with localized applications of chlorpyrifos as a subterranean termiticide. This is being addressed, EPA

says, by eliminating all termiticidal uses. However, despite EPA's assertions that only termiticidal use leads to water contamination problems, USGS and others have found contamination of ground and surface water with chlorpyrifos and its metabolites, and EPA's own modeling shows that it is likely that in certain areas of heavy use, chlorpyrifos (and its metabolites) present significant water risks. There is no evidence that the water risks of chlorpyrifos and its metabolites are limited to termiticidal use. This evidence of surface and groundwater contamination with chlorpyrifos is discussed below.

Drift/Air Exposure

In certain "sentinel" populations, such as farmworker children who live in a pesticide-rich environment, registered, non-residential, non-dietary sources may account for most of a child's exposure to pesticides, regardless of whether there is registered indoor use. Pesticides applied aerially must be assessed for its effects on people affected by pesticide drift or sloppy application. Reports in the medical literature describe numerous preventable illnesses and deaths among children with such "take-home" exposures. NRDC's report, *Trouble on the Farm*, documents the scientific evidence supporting the potential for take-home exposures from pesticides, even when not registered for residential use (this report is hereby incorporated by reference). These exposures are particularly important for children given their greater potential susceptibility, hand-to-mouth behavior and other behaviors in the home.

Significant concentrations of organophosphate pesticides (of at least 1.0 ng/m^3, and at least 2.4 ng/m^3 in one site in California) have been detected in winter fog, outside of the growing season (Glotfelty and others, 1987;[v] Seiber and others, 1993[vi]). This phenomenon may be due to volatilization or wind erosion, as is the case for other pesticides (Glotfelty and others, 1990c; Wu, 1981).[vii] Evidence suggests that photochemical reactions lead to the production of oxygen analogs (oxons) of chlorpyrifos in air during daylight, which are incorporated into the nighttime fog (Glotfelty and others, 1990a[viii]). Chlorpyrifos has been detected in rain at concentrations up to 180 ng/L, in air up to 199/ng/m^3, and in fog up to 14,200 ng/L.[ix] The maximum concentration found in rain frequently exceeded EPA's water quality criteria for freshwater aquatic organisms (83 ng/L – acute; 41 ng/L - chronic).[x] In addition, several studies have shown that drift is a significant concern. Thus, EPA must consider airborne exposure as a source in conducting its assessment of risk.

Ecological Risks

Ecological risks from chlorpyrifos use remain extremely worrisome; the Agency notes high risks, acute and reproductive toxicity, to birds, fish, mammals, and aquatic invertebrates. Some risk quotients exceed 1000 times the acceptable limit (IRED pp. 52-58). It is highly toxic to honey bees, which are killed if present even within 24 hours after application (IRED p. 54). Given that animals and insects, and especially honeybees, pollinate over ¾ of the staple crop plants worldwide, and have an estimated economic value to world agriculture of $200 billion annually, it is no surprise that any decline in honeybee populations is considered a serious threat to world food supplies.[xi] The IRED states that mitigation will require reduced application rates, increased retreatment intervals, reduced seasonal maximum allowable rates, and no-spray setback zones (IRED, p. 4). NRDC believes that these recommendations are unlikely to be adhered to, as discussed further in these comments. EPA estimates that risks to invertebrates will remain of concern, despite these mitigation efforts. Further, bioconcentration of chlorpyrifos will likely pose an acute and reproductive risk to aquatic birds and mammals, in spite of mitigation efforts.

EPA's Decision

The Agency's IRED concludes that chlorpyrifos may be reregistered under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) for agricultural uses because the registrants agreed to cancel most residential uses, and because EPA intends to require some mitigation measures for the remaining agricultural uses. Under the Federal Food, Drug and Cosmetic Act (FFDCA), EPA relies on the same rationale to keep in effect those tolerances that the registrant continues to support.

The IRED states that with the addition of label restrictions and amendments detailed in the IRED, all currently registered uses of chlorpyrifos except open-pour dust formulations may continue. In the current IRED, EPA says the tolerance for tomatoes will be revoked, and tolerances for apples and grapes will be lowered to 0.01 ppm each (IRED p.18). In an effort to further mitigate elevated ecological risk, the IRED recommends some crop-specific measures, including reducing the maximal number of liquid applications per season as follows: alfalfa, from eight to four; citrus, to two; corn, to three, with a reduced application rate from 7.5 to 3 lbs a.i./A; cotton, from six to three, with a reduced application rate from 6 to 3 lbs a.i./A. Further, spray drift warnings and no-spray zones will be included on labels.

The Agency has determined that, with presumed full compliance with mitigation recommendations, a 10X FQPA factor for infants, children, and females aged 13-50 (IRED p.16),

and a 100X uncertainty factor for acute and chronic reference doses (IRED p.15) will provide a sufficient margin of safety.

EPA CANNOT LAWFULLY ESTABLISH OR LEAVE IN EFFECT TOLERANCES FOR CHLORPYRIFOS

EPA has found that there is ***no*** NOEL or NOAEL for the developmental effects of chlorpyrifos on human infants and children. EPA therefore has no scientific basis upon which to conclude that there is a fully safe level at which infants and children will not suffer developmental harm because of chlorpyrifos exposure. Moreover, EPA has no basis to derive a Q* or to use any other quantitative risk assessment methodology that will derive a negligible risk level (i.e. 1 in 1 million lifetime risk) for chlorpyrifos' developmental effects. Therefore, EPA simply cannot make a legal finding that any specific chlorpyrifos level on food is "safe" for infants and children, or that there is a "reasonable certainty of no harm" to infants and children, at any specific level. Thus, as a matter of law, under FFDCA §408(b)(2), EPA must revoke all tolerances, and cancel all food uses, for chlorpyrifos.

Even assuming for the sake of argument that EPA can set tolerances for chlorpyrifos, notwithstanding the legal prohibition on EPA's setting a tolerance for a substance that has no NOEL or NOAEL (and for which a Q* cannot be derived and therefore a cancer-like risk assessment is impossible), EPA has failed adequately to apply safety factors and to consider many routes of exposure in the IRED. EPA must apply not only the traditional two 10-fold safety factors for interspecies variability and for intra-species variability, but also must apply an additional safety factor of at least 10 for the data gaps and buried assumptions that underestimate risk, as detailed further below, and an FQPA safety factor of at least 30 to account for potential pre-and post-natal ***toxicity*** to infants and children (10X) and to account for significant gaps in ***exposure data*** for infants and children and the lack of a NOEL (3X). EPA has failed adequately to consider important exposure routes for millions of infants and children, including children living on farms and who accompany their parents into farm fields (see farm children section below), exposure from mosquito spraying, drift, and drinking water exposure. Moreover, EPA has failed to consider cumulative exposure to organophosphates and carbamates that have a common mechanism of toxicity; under FFDCA §408(b)(2), no tolerance may be established or left in place without considering such cumulative risks.

EPA must cancel all uses of chlorpyrifos under FIFRA. All food uses must be cancelled due to excessive risk and the cancellation of all tolerances under the FFDCA. Even if theoretically an extremely low tolerance could be set for chlorpyrifos after application of all appropriate safety factors, the chemical would have to be applied at such low rates an in such a manner that it would not be efficacious. Moreover, the food and non-food uses of chlorpyrifos result in worker exposure; non-occupational drift, air, and water exposure; other non-food human exposure; and ecological risks that pose "unreasonable adverse effects on the environment" and thus must result in cancellation of all uses of chlorpyrifos.

CHLORPYRIFOS TOXICITY CONSIDERATIONS

The Agency has seriously underestimated the risks posed by chlorpyrifos use, thereby undermining its conclusion that chlorpyrifos can continue to be used in a way that prevents "unreasonable adverse effects on the environment," FIFRA § 3(c)(5), and provides "a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue," FFDCA § 408(b)(2)(A). EPA rationalized the continued use of chlorpyrifos for agriculture applications, because EPA considers that dietary risks are not of concern, residential risks are no longer of concern due to voluntary withdrawals of household uses, and EPA considers that risks posed to workers and the environment are acceptable, although they exceed the level of concern in a number of scenarios.

While supporting the voluntary withdrawals of residential uses, schools, and parks, which is predicted to reduce by half the total pounds of chlorpyrifos used annually, NRDC takes issue with the above assumptions. Our concern about exposure to chlorpyrifos remains high, notwithstanding EPA's intended cancellation and mitigation actions. Unfortunately, the Agency has understated the adverse health effects caused by chlorpyrifos, the amount of exposure that people endure without effect, and the amount of chlorpyrifos to which people are exposed. With chlorpyrifos and other developmental neurotoxic chemicals, risk to the fetus, infant, and child comes primarily from the timing of exposure. Even a very small dose, for even a short duration, during a developmental period of vulnerability will result in permanent neural dysfunction. There is no demonstrated reliable threshold of safety for this highly toxic chemical, as indicated in the IRED, where a no-effect level could not be determined for developmental neurotoxicity[xii] (IRED p. 15-16). In addition, the Agency has overestimated the effectiveness of some mitigation recommendations. First, EPA assumes that pesticide applicators will reliably use personal protective equipment (PPE) and will always abide by specified re-entry intervals (REIs) – a

waiting time between applying the product and returning to the field – even though these mitigation measures are not enforced and are commonly ignored. Second, the Agency does not adequately account for "take-home" exposures from agriculture uses (domestic contact with pesticides tracked home on the clothes, shoes, and skin of workers) or exposures from spray drift (wind-blown pesticides from field or aerial applications that subsequently impact people). These points are described in greater detail in NRDC's comments submitted to the Agency October 16, 2000.[xiii]

There is demonstrated evidence of neuropathology and increased vulnerability of fetuses when exposed to chlorpyrifos (IRED p. 16; Makris et al.[xiv]). However, given that in these experiments neuropathology was seen in the neonates at the lowest dose tested, these studies were unable to identify an offspring NOAEL in the DNT (IRED p. 16). In that study, structural alterations in brain development, which would result in permanent brain dysfunction, were seen at the lowest doses tested (IRED p. 17), strongly indicating that a 10X FQPA margin of safety is insufficient to protect fetuses, neonates, and children from irreversible chlorpyrifos-induced brain damage. Furthermore, Congress was clear in stating that under the FQPA, EPA must establish tolerances based on a No Effect Level (NOEL); *not* the NOAEL. If there is no NOEL, EPA cannot simply *assume* that there is a safe level based upon no evidence, and then apply safety factors. Whereas Congress did allow EPA to consider setting tolerances for non-threshold *carcinogens* at a level that poses no greater than a 1 in 1 million lifetime cancer risk, EPA has no scientific basis upon which to set a tolerance for chlorpyrifos that would pose a 1 in 1 million risk of suffering developmental problems, cannot establish a NOEL, and thus cannot set or leave in place a tolerance for chlorpyrifos.

Of additional concern, the chlorpyrifos metabolite, TCP, is more persistent than the parent compound (IRED p.20), is of greater toxic potency to fetuses than adults (IRED p.16), and exceeds chronic DWLOCs for children (IRED p.16). The toxicity of the chlorpyrifos metabolite was not considered in the chlorpyrifos dietary assessment (IRED p.17). This omission represents a very serious risk to tens of thousands of exposed vulnerable fetuses, neonates, and children.

Given these very serious concerns, NRDC believes that EPA simply cannot establish or leave in effect any tolerance for chlorpyrifos. Assuming arguendo that EPA can do so, at a minimum EPA must use an additional 30X FQPA margin of safety – in addition to the other uncertainty factors – to protect children and to account for demonstrated fetotoxicity, uncertainty in extrapolating from

effect-levels to NOAELs, and very substantial gaps in exposure and toxicity data (detailed in these comments). The current IRED recommends an uncertainty factor of 100X for the acute and chronic reference dose (IRED p. 15). NRDC believes that if the agency decides to establish tolerances notwithstanding the lack of NOELs (a decision that we believe would not be lawful), the Agency is clearly obligated to apply a 1000X UF, in addition to a 30X FQPA margin of safety, comprised of: 10X for interspecies extrapolation, 10X for intraspecies variability, and 10X to adjust for the lack of consideration of metabolite-induced toxicity, uncertainty in extrapolating from subchronic to lifetime effects, and uncertainty associated with other non-conservative assumptions detailed in these comments.[xv] NRDC believes that there are no "safe" levels of chlorpyrifos for fetuses, infants, and children, as demonstrated by the Agency's own DNT studies. Exposure to the developing nervous system to neurotoxic chemicals during periods of heightened vulnerability, even at low levels or short durations, will disrupt normal structure and function and will cause permanent disruption of neural function (see numerous papers by Abou-Donia et al, and Slotkin et al [xvi]).

DEVELOPMENTAL TOXICITY OF CHLORPYRIFOS IS UNDERESTIMATED

The action of chlorpyrifos as a developmental neurotoxic chemical is undisputed. The EPA human health risk assessment of chlorpyrifos states:

> In conclusion, the weight of the evidence raises concern for an increase in both the sensitivity and susceptibility of the fetus or young animal to adverse biochemical, morphological, or behavioral alterations from chlorpyrifos treatment during brain development. With respect to cholinesterase inhibition, an increase in sensitivity of the young compared to adults was seen all along the dose response curve, even at relatively low doses. There is a clear differential response (2- to ~5-fold) in the young compared to the adult animal after an acute treatment to a relatively low dose of chlorpyrifos. There is also increased sensitivity found after repeated dosing (up to 9-fold), but at the LD10 and MTD. It is important to point out that an uncertainty remains concerning the magnitude of the differential response, given that newborn animals (less than PND 7) have not been characterized for sensitivity. Results of multiple studies have consistently shown that the developing brain is susceptible to chlorpyrifos treatment. Effects on the developing CNS that are indicative of the unique susceptibility to the young animal include changes in macromolecular synthesis, altered cell signaling and muscarinic receptor down regulation, as well as morphological alterations in brain development. An uncertainty remains regarding the NOAELs for the susceptibility effects. The effects observed raise a

high degree of concern that the fetus or young animal is particularly susceptible to adverse outcome if exposed to chlorpyrifos.[xvii]

A substantial body of scientific evidence exists demonstrating the fetotoxic, neurotoxic, and immunotoxic properties of chlorpyrifos and its oxon metabolite, with treatment in utero or perinatal resulting in permanent damage to the nervous system (see numerous papers by Abou-Donia et al, and Slotkin et al [xviii]). NRDC is extremely concerned, in light of the experimentally demonstrated, permanent, effects of chlorpyrifos in the developing nervous system, that the chlorpyrifos IRED has underestimated risk, used central tendency estimates, used non-conservative assumptions, and ignored data gaps in estimating exposure risk. These concerns are detailed further below.

The IRED Does Not Account for the Toxicity of Metabolites and Stereoisomers

Though EPA has abundant data for dietary exposure to chlorpyrifos, its PDP and FDA databases only include monitoring data for residues of the chlorpyrifos parent compound. As stated in the IRED, the Agency has concluded that the primary metabolite of chlorpyrifos, 3,5,6-trichloro-2-pyridinol (TCP), does not induce cholinesterase inhibition, and exhibits effects only at doses higher than those producing ChEI with chlorpyrifos, and therefore, is less toxic than chlorpyrifos (58 Fed. Reg. 19,354(April 14, 1993)) (IRED p. 16). However, compared with chlorpyrifos, TCP is stated to be "more mobile and significantly more persistent in many soils, especially under anaerobic conditions" (IRED p.20). Further, the Agency states in the IRED that, "upper-bound estimated environmental concentrations of TCP exceeded chronic DWLOCs for children" (IRED p. 16). This is especially disconcerting, given the "evidence of increased susceptibility of rabbit fetuses relative to dams" (IRED p.16). Recently published experimental evidence of the toxicity of chlorpyrifos oxon and TCP in animals[xix], and in a human cell line[xx] emphasize the need for inclusion of metabolites in the risk assessment. The impact of these metabolites on developing animals – where even short-lived compounds could conceivably have irreversible effects on the nervous system – heightens the need for prudence in carrying out cumulative assessments. EPA appears to have no requirement for chemical-specific pharmacokinetic studies in fetal animals that would aid in discerning the contribution of toxic metabolites, such as the chlorpyrifos oxon and TCP, to children's risk. Until data are available that are specific to parent compound/metabolite mixtures for chlorpyrifos, from all exposure sources, any risk assessment involving this chemical would be incomplete, and likely less protective of public health than is required by law. Under FQPA, EPA is to set tolerances so as to "ensure that there is a reasonable

certainty that no harm will result to infants and children from aggregate exposure to the pesticide chemical residue."[xxi] NRDC therefore asserts that the Agency is unjustified in its determination that "residues of TCP are not of concern for the chlorpyrifos dietary assessment," and "can therefore be excluded from the tolerance expression." (IRED p. 17).

Buried Assumptions Underestimate Risk

Many OP assessments contain buried or outdated assumptions that receive little or no recognition in the risk characterization, despite their less-than-health-protective impact on final risk estimates. These assumptions tend towards a lesser, rather than a greater, certainty of no harm to infants and children. EPA should acknowledge as much. Outside scientists have criticized many of these assumptions as not being sufficiently protective.[xxii]

Some specific examples from the chlorpyrifos and other OP assessments, drawn from the draft SOPs, illustrate the problem better:

- *EPA assumes all "toddlers," aged 1 to 6 years, weigh 33 pounds (15 kg).* EPA bases its assumption on the mean or average weight for three year-olds. Because home pesticide exposures and risks are calculated on a per pound basis, EPA's baseline assumption about a toddler's weight will tend toward risk estimates that understate true risks for smaller toddlers and younger infants whose brains and nervous systems are developing more rapidly and are therefore more vulnerable than those of older children.
- *EPA bases its estimate of a toddler's exposure to a chlorpyrifos-contaminated surface due to hand-to-mouth activity on the ludicrous assumption that this activity occurs just 1.56 times per hour.* Then-Assistant Administrator L. Goldman, a pediatrician, as well as EPA's latest scientific advisory panel all singled out this assumption as being particularly inadequate. The latter panel also indicated that assessment of a child's mouthing behavior would be incomplete if it focused only on hand-to-mouth exposures. Children put objects other than fingers in their mouth, and these objects may carry pesticide residues and be ingested. Children also eat "feral" food — food that's dropped on the floor and which picks up residues from contaminated linoleum, carpet or other household surfaces. In any case, the hand-to-mouth value EPA currently uses as its "conservative" assumption is roughly 16 times lower than the value obtained from a recent study of 30 children. Those children put fingers in their mouths 26 times per hour on average, with some children showing the behavior up to 70 times per hour[xxiii]. While EPA has proposed to change its guidelines to a value of 20 times per hour, this is still an average value. Moreover, until

> the change is final, the agency continues to use the dramatically low value of 1.56 times per hour — a value which any parent will recognize as ridiculously low.

Since FQPA's mandate for a reasonable certainty of no harm means that EPA must set tolerances so as to fully protect even exceptionally exposed children (i.e. EPA must set tolerances to protect *all* children, including all "major identifiable subgroups" of children at greater risk), and not merely the average child, use of these central tendency estimates are inappropriate. These concerns apply equally to the characterization of risk from residential exposure of agriculture uses such as spray drift, residential residue track-in, exposures to farm worker children, and exposures to children in and around schools.

It is clear the above central tendency assumptions are not health-protective of more highly-exposed and susceptible populations; EPA must stop making any claims of conservatism, and should instead acknowledge that current assessments using the draft standard operating procedures may be likely to understate true exposure for many children.

Reliance on Unenforced and Unreliable Mitigation Downplays Exposure

It is disturbing that the risks to workers for mixing, loading and applying chlorpyrifos to agriculture crops exceed safe levels, even when presuming full compliance with mitigation recommendations. Furthermore, it is widely recognized that training and monitoring workers for use of PPE is inadequate, and moreover, workers in hot fields often cannot tolerate PPE. EPA therefore should not assume that PPE will mitigate the risk from certain scenarios when real life experience suggests PPE cannot be relied upon. These particular chemical uses may not be found "safe", and by definition cause "unreasonable adverse effects on the environment" under FIFRA. Therefore EPA should issue a Notice of Intent to Cancel (NOIC) and proposal to revoke their tolerances.

The Agency has proposed to mitigate occupational risks partially through lengthening the re-entry interval (REI) for most crops. Although this seems at face value to be a health-protective step, in practice it has no mitigation value whatsoever. There is no enforcement for REIs, and therefore, lengthening such REIs, like requirements for increased PPE, are routinely ignored[xxiv]. Poor compliance with a mitigation effort that may sound reasonable on paper will not protect exposed individuals. It is clearly stated under FIFRA §3(C)(5) that "The Administrator shall register a pesticide if the Administrator determines that…..when used in accordance with widespread and

commonly recognized practice it will not generally cause unreasonable adverse effects on the environment". This passage illustrates that EPA cannot rely on mitigation methods, such as PPE and REIs, which are unenforced and poorly followed.

Exporting Hazards

Although chlorpyrifos is listed as a "restricted use" pesticide in the US, it is exported in high volume: 7 to 9 million pounds annually since 1997 (8,570,694 in 2000).[xxv] In fact, a recent article by C. Smith states that, according to customs records, between the years 1997 through –2000, nearly 65 million pounds of severely restricted or forbidden pesticides in the United States were exported; more than 22 tons per day. More than 55% of these products were exported to developing countries for agriculture use. The International Labor Organization estimates that 60 to 90% of children estimated to be working in Africa (80 million), Asia (152 million), and Latin America (17 million) are working in agriculture. These children are exposed to toxic pesticides in the fields, from drinking and washing water, through contaminated clothing, and in their homes.[xxvi] The U.N. Commission on Human Rights stated that "[a]llowing the export of products recognized to be harmful is immoral."[xxvii] The mitigation requirements in this IRED include respirators with an organic-vapor removing cartridge and a pesticide-approved prefilter, chemical-resistant outerclothes, enclosed-cab machinery, emergency equipment readily available, and storage containments for discarding single-use chemically-resistant overclothes. It is inconceivable that these are "readily available" to mixers, loaders, applicators, and fieldworkers in developing countries. Clearly, new labeling requirements will have no mitigation effects for these men, women, and children workers.

No Estimate of Risk Associated with Greenhouse and Nursery Uses

Of further concern, the IRED states that post application risks to nursery and greenhouse workers were unassessed, due to lack of data (IRED pp. 40, 89). This lack of data is insufficient justification to ignore the obvious risk of such registered uses. NRDC maintains that this must be considered an important gap in data required to conduct a complete risk assessment, and contributes to the underestimation (in this case, complete ignorance) of risk in this IRED.

RISK TO FARMWORKER CHILDREN

Farmworker Children Are an Identifiable High-Risk Group

In measuring the extent of exposure and in determining aggregate exposure, EPA should acknowledge farmworker children to be a major, identifiable subgroup of consumers whose

unique, increased level of exposures must be taken into account.[xxviii] NRDC's report, *Trouble on the Farm*, documents the scientific evidence supporting the potential for take-home exposures from pesticides, even when not registered for residential use (this report is hereby incorporated by reference). These exposures are particularly important for children given their greater potential susceptibility, hand-to-mouth behavior and other behaviors in the home. These nearly 1,000,000 children are deserving of protection under the "reasonable certainty of no harm" health standard under the law.

EPA's refusal to apply a sufficient margin of safety for children (at least 30X) in its chlorpyrifos assessment is inconsistent with the need for additional protections for the fetuses of pregnant farmworker women who may be exposed while their mothers are at work, and the risks facing neonates who are brought to the fields to accompany their parents due to lack of day care. These babies, who face exposure to an extremely potent neurotoxin at vulnerable stages of development are *not* employees and may *not* be disregarded on the grounds they face an *occupational* risk (Farmworker Justice Fund, Comments to the EPA's Pesticide Docket on the Preliminary Risk Assessment for Chlorpyrifos, December 23, 1999).

The legal analysis submitted by Farmworker Justice Fund to the Pesticides Docket for the chlorpyrifos risk assessment remains relevant:

> "In setting, modifying or revoking tolerances, the FQPA directs the EPA to consider, *inter alia*, *'a*vailable information concerning the ... effects of *in utero* exposure to pesticide chemicals.' § 408 (b)(2)(C)(I)(II). In the case of threshold effects, FQPA also directs the EPA to add an additional 10-fold (or other) margin of safety for infants and children 'to take into account potential pre- and post-natal toxicity and completeness of the data with respect to exposure and toxicity to infants and children.' *Id.* at 408(b)(2)(C)(ii). In explaining its method of implementing the 10-fold safety factor to the SAP, the EPA expressly stated that it would *not* consider prenatal exposures to the unborn children of pregnant farmworker women because such exposures are 'occupational' and hence, not within the contemplation of the FQPA. *See Presentation for the FIFRA Scientific Advisory Panel by Office of Pesticide Programs, Health Effects Division on FQPA Safety Factor for Infants and Children* (March 1998). The statutory language which directs the EPA to consider the effects of 'in utero' or 'pre-natal' exposures to pesticides makes *no* exception for occupational exposures. Nor could such an exception make sense since it is patent that a fetus or unborn child cannot work. In an

> analogous context, the California Supreme Court has held that a child, who was injured in utero when his pregnant mother was exposed to carbon monoxide at work, could not be prevented from filing suit in tort by the workers compensation bar, which prohibits an employee from suing his or her employer. *Snyder v. Michael's Stores Inc,* 16 Cal.4th 991, 945 P.2d 781, 68 Cal.Rptr.2d 476 (1997). The Court dismissed the notion that the unborn child could be deemed an 'employee' as 'wholly without merit.' The Court also noted that every other court to consider this question - except one - had reached the same conclusion (and the only exception was a lower California court whose decision was effectively overruled by the *Snyder* case). Since an unborn child cannot be an 'employee,' its pesticide exposure cannot be 'occupational.'"

Thus, any prenatal exposure to the fetuses of farmworkers must be considered in the determination to set the FQPA margin of safety for infants and children. Although the Agency applied a 10X FQPA factor, given the extremely neurotoxic action of chlorpyrifos, the demonstrated increased vulnerability of fetuses and neonates, and the lack of a NOEL,[xxix] NRDC maintains that this is woefully insufficient to provide a margin of safety for these vulnerable sub-groups.

Drift and Take-Home Exposures to Chlorpyrifos

Even setting aside the issue of whether an unborn child in a farmworker mother's womb is exposed as a result of "occupational" activity when the mother is in the fields, it is clear beyond peradventure that EPA must consider fetal and childhood exposure of farm children and fetuses as a result of drift, take-home exposure, drinking water exposure in farm areas, in-field "day care," and other farm child exposure resulting from proximity to the fields. These comments are discussed in detail in the NRDC Report *Trouble on the Farm.*[xxx]

The scientific literature and common sense demonstrate that children and others experience substantial potential exposures through drift from crop spraying, and through take-home exposures when, for example, pesticides are exhaled from parents' lungs or brought home on boots, work clothes, etc. [xxxi] [xxxii] Pesticides used on lawns, gardens and nearby farms end up in soil and are tracked into the home on shoes and pets. One common lawn herbicide, 2,4-D, has been found to persist in carpet dust up to a year after lawn application.[xxxiii] For methyl-parathion, for example, 34% of original residues remained in clothes even after 10 launderings—a level high

enough to kill insects and present a health risk to humans.[xxxiv] Chlorpyrifos is expected to cause long-lasting take-home exposure problems (IRED p. 41).

EPA's current policy, as stated in the spray and dust drift label policy, is clear. The Agency is obligated to protect public health from all pesticide exposures, including those resulting from spray and dust drift:

> "EPA's position on pesticide drift is that applicators must not allow pesticide spray or dust to drift from the application site and contact people, animals, and certain sensitive sites, including structures people occupy at any time and the associated property, parks and recreation areas, nontarget crops, aquatic and wetland areas, woodlands, pastures, or rangelands. The Agency believes this is prudent public policy. It sets high but appropriate standards for applicators to protect people and the environment."[xxxv]

In certain "sentinel" populations, such as farmworker children who live in a pesticide-rich environment, these non-dietary sources may account for most of a child's exposure regardless of whether there is registered indoor use. Reports in the medical literature describe numerous preventable illnesses and deaths among children with such "take-home" exposures. NRDC's report, *Trouble on the Farm*, documents the scientific evidence supporting the potential for take-home exposures from pesticides, even when not registered for residential use (this report is hereby incorporated by reference). These exposures are particularly important for children given their greater potential susceptibility, hand-to-mouth behavior and other behaviors in the home.

EPA is now considering building into its pesticide risk assessments the fact that worker risks may "spill over" to the families of workers and to fetuses that workers may carry on and off the job. For example, pregnant women working or living on or near farms may very well have pesticide exposures that clearly fall within the purview of the FFDCA section 408 aggregate safe exposure requirement—particularly (but not exclusively) when the exposures occur off the work site due to take–home/drift exposures. Current EPA practice, including the current chlorpyrifos risk assessment, is to consider an additional FQPA 10X margin of safety, when applicable, only to consumers of food crops and not to exposed workers and their families. Given the certainty of drift and take–home exposures with many pesticides, including chlorpyrifos, the Agency is obligated to expand its exposure assessment to include these risks from agriculture use. EPA's failure to incorporate these real-life exposures into its risk assessments will result in final risk estimates that are not adequately protective of human health. If data are lacking to quantify such

exposures, it is essential to incorporate an additional margin of safety to assure that use of the chemical is consistent with a reasonable certainty of no harm to children until more precise data can be generated, as required by the FQPA.

As detailed in the above discussion, it is invalid to presume, as the IRED does, that by canceling residential uses of chlorpyrifos, there will no longer be a risk of exposure to fetuses and children. NRDC asserts that so long as agriculture uses remain, the risk of exposure to fetuses and children is certain.

DIETARY EXPOSURE IS UNDERESTIMATED

EPA's Acute Dietary Exposure Estimate Is Flawed and Unlawful

For chlorpyrifos, as with other OPs, the Agency used estimates of the percentage of crops treated (%CT) from BEAD (Biological Economic Analysis Division) to derive the acute dietary risk estimate, rather than assuming the more conservative approach of 100% crop treated (IRED p. 18; Acute dietary risk assessment[xxxvi]). Using %CT in estimating acute risk violates governing law. The FQPA specifically authorized EPA to consider the percent of crop treated (%CT) "when assessing *chronic* dietary risk . . . only if the Administrator" makes four specific findings about data reliability. FFDCA § 408 (b)(2)(F) (emphasis added). By contrast, the law nowhere allows EPA to use %CT in assessing *acute* dietary risk. Of course, the law's distinction between chronic and acute risks makes perfect sense. Congress understood that the likelihood that a person will experience harm from chronic exposures over time could be affected by the overall percentage of that crop treated with that chemical. On the other hand, in examining the acute harm resulting from a single exposure, it is irrelevant whether a large or small percentage of that crop was treated with that pesticide. *Any* amount of a crop treated at a level causing acute harm could not be characterized as assuring a "reasonable certainty of no harm," making it completely inappropriate and unlawful to use %CT in assessing *acute* dietary risks. Using %CT to assess acute risk is in direct violation of the FQPA.

In the case of chlorpyrifos, BEAD estimated a maximum of 53% crop treated for apples, and only 1% CT for grapes.[xxxvii] By using %CT, rather than assuming 100% CT, the Agency has underestimated risk for affected individuals by as much as 100X for grapes (used 1%CT), and 1.8X for apples (used 53% CT). This is especially important given that the mitigation measures proposed in this IRED include reducing tolerances for both apples (currently 1.5 ppm) and grapes (currently 0.5 ppm) to 0.01 ppm. Simple math reveals that the Agency has underestimated the

risk estimate for grapes by 100X, and then reduced the tolerance by 50X, resulting in an overall increase by 50X in allowable risk. For apples, the Agency has underestimated risk by 50X, and then reduced the tolerance by 150X, resulting in a real-reduction of only 100X. Since apples and grapes are acknowledged to be the largest contributors to dietary exposure (especially for children), this is extremely worrisome[xxxviii] (IRED p. 18). Further, the acute dietary risk assessment for chlorpyrifos indicates that without mitigation, exposure from residues on fresh apples exceeds the allowable risk for children 1-6 yrs by over 300X (aPAD=364%)[xxxix] (IRED p. 19), rendering the above 50X increase completely inappropriate and the above 100X reduction wholly inadequate.

The mitigation proposed, which is limited to the above described insufficient tolerance changes and new labeling requirements, which are unenforced and poorly adhered to, is unlikely to result in any substantial reductions in exposure.

Drinking Water Exposures Do Not Account for Toxic Metabolites

For drinking water, the chlorpyrifos metabolite TCP is not included in the tolerance expression, despite being identified in a number of environmental fate studies. Compared with chlorpyrifos, TCP is stated to be "more mobile and significantly more persistent in many soils, especially under anaerobic conditions" (IRED p.20). Further, the Agency states in the IRED that, "upper-bound estimated environmental concentrations of TCP exceeded chronic DWLOCs for children" (IRED p. 16). This is especially disconcerting, given the "evidence of increased susceptibility of rabbit fetuses relative to dams" (IRED p.16). Given the demonstrated evidence of exposure to TCP in food and drinking water, and the increased vulnerability of children and pregnant women, NRDC believes the Agency is unjustified in not including TCP exposure in its tolerance assessment.

Under FQPA, drinking water exposures (through ingestion, inhalation, and dermal absorption from hand-washing and showers) at least must be estimated. "Refinement" of drinking water data in a risk assessment may be an appropriate long-term goal, but it is scientifically unjustified for EPA to intentionally circumscribe the scope of the risk assessment by ignoring these exposures in the interim. Where EPA lacks drinking water monitoring data for specific chemicals or toxic metabolites, it should make quantitative or qualitative estimates and be frank in its description of both the assumptions, the uncertainties, and the limitations of the data.

Inclusion of complete, real-world drinking water exposures to chlorpyrifos and its metabolites such as TCP at sites of maximum likely exposure in an aggregate exposure assessment is critical for two additional reasons. First, EPA decision-makers should be presented with a risk assessment that reflects the entire range of real-life exposures to the chemical in question. Second, because exposure to OP-contaminated drinking water will tend to add to the estimated risk, its inclusion in the preliminary risk assessment is necessary to demonstrate, in the most transparent way, the urgency of taking immediate steps to reduce that risk. When EPA fails to incorporate real-life drinking water OP exposures into its risk assessments, it will tend toward risk estimates that are less than health-protective. If data are lacking to quantify drinking water exposure to individual OPs, the use of an additional 10X margin of safety is essential to assure that use of the chemical is consistent with a reasonable certainty of no harm to children until more precise data can be generated.

Water Monitoring Data Is Not Designed to Detect Peak Levels

The IRED states that surface water estimates were based on the USGS NAWQA monitoring data, which reported the maximum dissolved chlorpyrifos concentration was 0.4 ppb (IRED p.21).

Surface Water

Combined USGS data for state, local, national, and multi-state studies that measured chlorpyrifos concentrations in surface water detected the pesticide at 7 of 108 (6%) sites sampled.[xl] *Most of these data do not include TCP or other chlorpyrifos metabolites*. Chlorpyrifos has medium runoff potential due to its relatively low water solubility, 2 mg/L, (Becker and others, 1989[xli]; Goss, 1992[xlii]), though some of its metabolites are more soluble and persistent A chlorpyrifos flux as a percentage of use of 0.15 has been measured in the Minnesota River.[xliii] Chlorpyrifos is also, of course, used in non-agricultural settings, and can thus drift or runoff directly into surface water bodies in areas of high population density. In two out of nine studies that measured chlorpyrifos concentrations in surface waters, its concentration exceeded EPA's water quality criteria for aquatic organisms in some samples (0.083 µg/L [83 ng/L] – acute; 0.041 µg/L [41 ng/L] – chronic).[xliv,xlv,xlvi,xlvii]

Ground Water

Data from the Mid-Continent Pesticide Study[xlviii] shows that chlorpyrifos was present in the ground water in 4.2% of the wells sampled.[xlix] Cohen and others (1990) found the chlorpyrifos transformation product 3,5,6-Thrichloro-2-pyridinol in ground water in Cape Cod, Mass.[l]

Chlorpyrifos has been detected in 0.6% of wells sampled, according to the U.S. EPA's Pesticides in Ground Water Database.[li] Long (1989) detected chlorpyrifos in the ground water of 30% of 56 sites examined beneath pesticide mixing and loading facilities in Illinois.[lii] The maximum concentration detected was 0.5 µg/L. Habecker (1989)[liii] detected a maximum surface soil concentration of chlorpyrifos of 41,000 µg/kg at pesticide mixing and loading sites in Wisconsin. Krapac and others[liv] detected a maximum surface soil concentration of 26,000 µg/kg at agrichemical facilities in Illinois.

EPA Must Consider Exposures at Areas of High Water Contamination.

With the exception of the extraordinarily high exposures that would be suggested by the data for mixing and loading areas, these monitoring data generally are not targeted to areas of anticipated maximum exposure, which are the areas that FQPA requires that EPA consider in setting "safe" tolerances considering aggregate exposure. EPA, therefore, must at least estimate such maximum exposure levels using modeling.

Using the PRZM/EXAMS screening model, estimated 90-day average and peak chlorpyrifos concentrations were 6.7 and 40 ppb respectively. However, EPA did not rely upon these model estimates. Rather, the IRED estimated acute exposures at 0.026 to 0.4 ppb, based on the 95th percentile of the monitoring data. This is a full 100X lower than the 40 ppb estimate derived from the PRZM/EXAMS model.

NRDC contends that, contrary to the assertions of conservatism in the IRED (IRED pp. 21-22), these estimates likely underestimate actual levels substantially. Water monitoring sample sites are not necessarily correlated with chlorpyrifos use sites, and in particular, may miss sites where multiple fields are treated with chlorpyrifos resulting in pooled runoff into a common water source. In fact, the IRED states, "it is not clear that they [monitoring data] represent the most vulnerable groundwater where chlorpyrifos is used most intensively" (IRED p.22). Monitoring of surface water is likely to be subject to the same problem. Levels of chlorpyrifos in pooled runoff sites are likely to be many times higher than single field sites. Similarly, data collection is not timed to correspond with worst-case scenarios, such as closely following chlorpyrifos applications, or following large storm runoff events, and thus most often misses these highly toxic environmental exposures.

AGGREGATE RISK ESTIMATE IS INADEQUATE

Under the FFDCA, a pesticide tolerance can only be established if "there is a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue, including all anticipated dietary exposures and other exposures for which there are reliable information." FFDCA § 408(b)(2)(A)(ii). Aggregate exposure is the total exposure to a single chemical or its residues that may occur from dietary (i.e., food and drinking water), residential and other non-occupational sources, and from all known or plausible exposure routes (oral, dermal and inhalation).

The FQPA requires assessment of both dietary and non-dietary, non-occupational pesticide exposures in the aggregate, and FIFRA demands that EPA protect against pesticides' unreasonable health and environmental effects. Both good science and public health protection demand routine assessment of aggregate exposure because real-world exposures aggregate – that is, pesticides are used in a wide variety of settings and formulations likely to result in multiple exposure sources. Yet EPA has failed to do an adequate aggregate assessment in its chlorpyrifos assessment.

In addition to food and water exposures, the aggregate assessment must take into account exposures due to air drift and migration of contaminated soil, especially in agricultural areas, residential exposures from registered uses (including golf courses, greenhouses, and nurseries), residential "take-home" exposures to families of those directly exposed to the OP through its agricultural uses, as well as exposures from uses that do not conform with the label, where there is an indication that these uses occur.

When lacking actual data on any these various sources of non-dietary exposure, EPA should not simply assume that the particular route of exposure is unimportant or nonexistent, as it has chosen to do with respect to chlorpyrifos. For example, in the chlorpyrifos IRED, the Agency has ignored the contribution to risk from chlorpyrifos metabolites, which are highly toxic and persistent (IRED p.17), from spray drift and take-home exposures associated with agriculture uses (IRED p. 41), and from greenhouse and nursery uses (IRED p. 40). The risk characterization should clearly note that failure to include all possible routes of exposure will tend to bias final estimates of aggregate risk so that they will understate rather than overstate true risks. In other words, EPA's risk estimates are less rather than more protective of public health. Moreover, when there are not actual data to confirm the absence of exposure to a pesticide across any particular route, EPA must incorporate an additional safety factor to account for this lack of complete exposure data.

LEVEL OF REGULATION MUST ENSURE A REASONABLE CERTAINTY OF NO HARM FOR *ALL* FETUSES, INFANTS, AND CHILDREN

EPA may not sacrifice the hundreds or thousands of children who may exceed the reference dose for a particular OP. Under FQPA, the burden is upon the advocate of a tolerance to prove (and upon EPA to find) that there is a reasonable certainty that no children will be harmed in EPA 's pesticide decisions. Thus, if the best evidence suggests that hundreds or even thousands of children will exceed the reference dose for an OP, EPA is forbidden by statute to find a reasonable certainty of no harm to these particular infants and children, and the Agency should not issue a tolerance at that level.

Instead, in EPA's proposed approach – which is styled as a "highly refined" Monte Carlo risk analysis – the agency regulates dietary residues of individual OPs at the 99.9th percentile. EPA seeks to mask in this approach the fact that even regulation at the 99.9th percentile, for a pesticide commonly used on a ubiquitous children's food, means that 0.1% of all American children under age six (around 24,000 children in all) could exceed the chronic RfD every day, based on the best information available to the agency. Further, a child exposed to multiple organophosphate pesticides may fall within the 99.9th percentile for one, but lie above the safety threshold when cumulative OP risks are calculated. No reading of the statute will support any approach that allows hundreds or thousands of children to exceed the reference dose. Regulating dietary residues of chlorpyrifos at the 99.9th percentile directly violates the plain statutory language of the FQPA.

HUMAN TESTING

In 1999, chlorpyrifos registrants submitted to EPA a never-published, non peer-reviewed study in which volunteers had been intentionally dosed with chlorpyrifos for the purpose of discovering a no-effect level, and therefore influencing regulatory levels for this nerve system poison.[lv] In previous years, human tests of chlorpyrifos have been performed on small groups of prison "volunteers" as well, and the results submitted for consideration by EPA. The human tests submitted by chlorpyrifos registrants should be rejected by EPA. The purpose of using results from human testing is to justify the establishment of less stringent safety standards. Intentionally dosing human subjects with known poisons – with no medical benefit to the subjects –is illegal, unethical, and unscientific.

Human testing of pesticides violates the Nuremberg Code, adopted by American judges in the wake of the Nuremberg "Doctors Trials" of Nazis following World War II. The Nuremberg Code has been relied on by state and federal courts as establishing rock bottom minimum legal standards for human testing. Furthermore, EPA's consideration of the chlorpyrifos human tests would clearly be unethical and in contravention of the Helsinki Declaration, the Common Rule, FIFRA, and the EPA Scientific Advisory Panel/Science Advisory Board report on human testing. Finally, the scientific value of the results of the chlorpyrifos human tests is negligible at best. The tests involved so few subjects that the risks to the broader population –especially the most vulnerable subgroups – cannot be meaningfully assessed. The IRB process used to justify the tests, the "voluntary" consent process used, and many other aspects of the chlorpyrifos human tests were in clear violation of ethical and legal requirements of the Nuremberg Code, Helsinki Declaration, the Common Rule, FIFRA, and the EPA Scientific Advisory Panel/Science Advisory Board report on human testing.

We note that, in December 2001, the Agency requested that the National Academy of Sciences conduct a review of the scientific and ethical issues posed by use of these studies.[lvi] The Agency has stated that during the Academy's deliberations, and until a policy is in place, the Agency will not consider or rely on any such studies, whether previously or newly submitted.[lvii] NRDC urges the Agency to adhere to this position, and to refrain from any consideration of these studies. EPA must reject the use of human tests, consistent with sound scientific practice and pursuant to EPA's ethical and legal obligations.

Thank you for the opportunity to provide these comments.

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[iii] Consumers Union of US, Inc. Washington, DC. 1998. Worst First; High-risk insecticides, children's foods and safer alternatives. p. 13

[iv] Kate Hallward, Anne Katten, Margaret Reeves and Kristin Schafer, *Facing Poison in the Fields: California Farmworkers and Pesticides* (1999); Shelley Davis and Rebecca Schleifer, *Indifference to Safety* (1999)
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[xiii] NRDC comments submitted by A. Quintero to the Chlorpyrifos Docket: OPP-34203C, October 16, 2000.

[xiv] Makris S, Raffaele K, Sette W, Seed J. A retrospective analysis of twelve developmental neurotoxicity studies submitted to the USEPA Office of Prevention, Pesticides, and Toxic Substances (OPPTS). Draft 11/12/98.

[xv] this list of buried assumptions and underestimates of risk is similar to that cited in the EPA Trichloroethylene Health Risk Assessment, Review Draft, August, 2001, section 1.4

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ATTACHMENT 3

NRDC
THE EARTH'S BEST DEFENSE

DUE: October 5, 2011

Docket EPA-HQ-OPP-2008-0850

COMMENTS FROM THE NATURAL RESOURCES DEFENSE COUNCIL ON THE CHLORPYRIFOS PRELIMINARY HUMAN HEALTH RISK ASSESSMENT FOR REGISTRATION REVIEW Document ID EPA-HQ-OPP-2008-0850-0025

These comments are submitted on behalf of our members. The Natural Resources Defense Council (NRDC) is among the nation's most effective environmental action groups, combining the grassroots advocacy of 1.3 million members and online activists with the courtroom and scientific expertise of more than 350 lawyers, scientists and other professionals on staff. NRDC has no financial interest in chlorpyrifos or any other pesticide product that may be the subject of these comments.

INTRODUCTION

A decade ago, the EPA took an important step towards protecting human health by banning indoor uses of chlorpyrifos, otherwise known as 'Dursban.'. As a result, children today are not being exposed to these chemicals from roach killers, flea bombs, and other dangerous household pesticides. Unfortunately, chlorpyrifos is still widely used in agriculture at about 10 million pounds annually, putting farm workers and their families, agricultural communities, and food consumers at risk.

NRDC petitioned the U.S. Environmental Protection Agency (EPA) in 2007 to cancel the remaining uses of chlorpyrifos. As part of a settlement of the subsequent NRDC lawsuit, EPA was scheduled to release an updated assessment of the health risks of chlorpyrifos by June,

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2011– a deadline that has already passed. EPA needs to protect human health, especially mothers, infants, and children, from chlorpyrifos by cancelling all remaining uses, including agriculture uses, and revoking all tolerances of this dangerous war-era chemical.

Unless otherwise specified, these comments refer to the 2011 Chlorpyrifos Preliminary Human Health Risk Assessment for Registration Review (hereinafter referred to as the Preliminary Risk Assessment or PRA).

The 2011 PRA contains a number of significant weaknesses and some improvements compared with the 2000 assessment. These are discussed in more detail in the body of these comments, but can be summarized as follows:

A. ***New epidemiology on harm to kids*** – EPA has identified several important new studies that have been published showing that kids exposed to chlorpyrifos and other organophosphate pesticides during prenatal and early-life have measurable delays in cognitive achievement, motor control, social behavior, and intelligence measures. These studies support NRDC's call to cancel all uses of chlorpyrifos, and EPA should act immediately to do so.
B. ***FQPA kids safety factor removed*** - EPA has inappropriately reduced the FPQA factor from 10X in the 2000 assessment to 1X in the 2011 PRA.
C. ***Dietary risk estimates are weakened*** - The 2011 PRA uses acute and chronic dietary risk estimates for food that are 7-10 fold weaker than the values used in the 2000 assessment.
D. ***Drinking water exposures unsafe*** - Exposure estimates from drinking water exceed the level of concern (i.e. may be unsafe) for all infant scenarios, and for all populations at the high end crop-application rate combinations.
E. ***Residential bystander exposures from field volatilization unsafe*** – EPA assessed bystander inhalation exposure from field volatilization of applied chlorpyrifos and found air concentrations exceeded levels of concern for several ambient air concentrations, acute application site air concentrations, and intermediate-term application site air concentrations.
F. ***Occupational exposures unsafe –*** Of 305 occupational exposure scenarios assessed by EPA, one-quarter had risk estimates of concern even with the maximum level of personal protection and engineering controls considered. And, 43% has risk estimates that were only 'acceptable' when some level of personal protection was required.

Glossary of terms:

For dietary risk assessments (other than cancer) the Agency uses the uncertainty factor (UF) to calculate an acute or chronic reference dose (acute RfD or chronic RfD) where the RfD is equal to the NOAEL divided by the appropriate UF (RfD=NOAEL/UF).

For dietary risk assessments (other than cancer) with FQPA concerns, the acute or chronic Population Adjusted Dose (aPAD or cPAD) is a modification of the RfD to accommodate the

FQPA factor. The FQPA factor is applied to the RfD by dividing the RfD by the FQPA factor (PAD=RfD/FQPA). Risk is presented as the %PAD, where %PAD= (exposure/PAD)x100%.

Residential (non-dietary) risk is measured by a Margin of Exposure (MOE), which measures how close the residential exposure comes to the NOAEL, from animal studies. Generally MOEs greater than 100 do not exceed the Agency's level of concern (this incorporates the standard uncertainty factors of 10X for interspecies variability and 10X for intraspecies variability).

For non-dietary risk assessments (other than cancer) the UF is used to determine the level of concern, LOC. For example, when 100 is the UF, the LOC is 100. To estimate risk, a ratio of the NOAEL to exposures (MOE=NOAEL/exposure) is calculated and compared to the LOC.

The Drinking Water Level of Comparison (DWLOC) represents the maximum allowable contribution to the human diet that may be attributed to residues of a pesticide in drinking water, after dietary exposure is subtracted from the aPAD or cPAD. Risks from drinking water are assessed by comparing the DWLOC to the estimated environmental concentrations (EECs) in surface and ground water. Generally, the Agency has no risk concerns when the EECs are below the DWLOC. DWLOC=(water exposure X body weight)/ (Liters of water x 10^3)

DETAILED COMMENTS

A. New studies on kids show harm from prenatal and early life exposure

EPA has correctly identified three important new scientific studies published in the last year that demonstrate lasting impairments in children exposed to chlorpyrifos and/or other related OP (organophosphate) pesticides during pre-natal or early life stages, but has failed to commit to cancelling all remaining chlorpyrifos uses, despite this evidence of harm. The three studies are (PRA at 30):

- *The Columbia study*, from Columbia University, NYC. This study population includes multi-ethnic urban low income mother-infant pairs, and focuses on chlorpyrifos in infant cord blood. (Rauh et al, 2006)

- *The Mt. Sinai study*, from Mt. Sinai School of Medicine, NYC. This study population includes multi-ethnic urban low income mother-infant pairs, and assesses non-specific OP metabolites in maternal urine linked with associated health outcomes of children that were exposed in utero. (Engel et al, 2011)

- *The CHAMACOS study*, from University of California Berkeley Center for Health Assessment of Mothers and Children of Salinas, CHAMACOS. This study population is a farm worker/ agriculture worker population, and assesses non-specific OP metabolites in maternal urine linked with associated health outcomes of children that were exposed in utero. (Eskenazi et al, 2007)

All three of the studies were done by federally-funded university scientists and published in the government-supported high quality scientific journal, Environmental Health Perspectives, with updated published in 2011 as follows:

> The Columbia study update: *Seven-year neurodevelopmental scores and prenatal exposure to chlorpyrifos, a common agricultural pesticide.* Rauh V, Arunajadai S, Horton M, Perera F, Hoepner L, Barr DB, Whyatt R. Environ Health Perspect. 2011 Aug;119(8):1196-201.
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> The Mt. Sinai study update: *Prenatal exposure to organophosphates, paraoxonase 1, and cognitive development in childhood*. Engel SM, Wetmur J, Chen J, Zhu C, Barr DB, Canfield RL, Wolff MS. Environ Health Perspect. 2011 Aug;119(8):1182-8.
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> The CHAMACOS study update: *Prenatal Exposure to Organophosphate Pesticides and IQ in 7-Year-Old Children*. Bouchard MF, Chevrier J, Harley KG, Kogut K, Vedar M, Calderon N, Trujillo C, Johnson C, Bradman A, Barr DB, Eskenazi B. Environ Health Perspect. 2011 Aug;119(8):1189-95.

EPA notes that "The Columbia results are associated with high chlorpyrifos cord blood levels, while the CHAMACOS and Mt. Sinai teams correlated increasing levels of maternal urinary [OP metabolites] with reported mental delays in children" (PRA at 30, 31). Moreover, EPA points out that "the neurodevelopmental outcomes reported for children in these epidemiology studies are qualitatively similar to the behavioral outcomes in animal studies (following gestational and/or postnatal exposures to chlorpyrifos" thus providing evidence of concordance across species and strong scientific support for the accuracy of the findings.(PRA at 30)

The three studies are all a prospective epidemiology design, a robust scientific study design. Three groups of researchers, conducting three independent studies, including a total of nearly 800 children from New York and California, all found the same thing: maternal exposure to certain pesticides during pregnancy predicts neurological deficits in children during childhood. The scientists found that the children with higher exposure to these pesticides had lower IQ, poorer working memory, and diminished perceptual reasoning.

EPA indicates that it plans to conduct a "full weight of evidence evaluation integrating the epidemiology studies with the experimental toxicology studies" based on the neurodevelopmental endpoints. EPA indicates that such an assessment will consider criteria such as strength, consistency, specificity, dose response, temporal concordance and biological plausibility.

NRDC supports EPA's approach generally. NRDC also reminds EPA that both the EPA Cancer Guidelines (2005) and the Bradford-Hill Criteria for Causation (Hill Criteria) from which EPA's 'weight-of-evidence' approach is drawn emphasize that not all the criteria need to be met for a study's findings to be "true." In fact, epidemiology by design is biased towards the null

hypothesis (no effect) so that the chances of a Type I error (a false positive) is much less than the chances of a Type II error (a false negative.) In other words, when epidemiologic studies do find an effect, it is most likely real.

Proportion of risk attributable to chlorpyrifos versus other OPs – reason to ban all OPs, not excuse for inaction

All three epidemiologic studies report a "consistency of findings" including "behavioral delays in cognitive achievement, motor control, social behavior, and intelligence measures" across all three cohorts (PRA at 30). However, EPA states that, "the degree to which chlorpyrifos is implicated in these outcomes varies" (PRA at 30). In fact, while chlorpyrifos is certainly a contributor, it is not the only contributor to the harm. EPA acknowledges that other pesticide exposures are a risk contributor, including other OPs. (PRA at 32). However, EPA treats this as if it were a confounder that makes it difficult for EPA to rely on the epidemiological evidence – as if being unable to determine the attributable risk to one pesticide were an insurmountable problem. EPA writes, "All three cohort studies have limitations that include multiple chemical exposures and exposure to other organophosphates" (PRA at 32). In fact, when children are being poisoned by OP pesticides, then to the affected individuals it hardly matters what proportion of risk is attributable to chlorpyrifos versus the other OP pesticides. EPA should cancel chlorpyrifos because it contributes to serious irreversible harm. EPA should also cancel all OPs for the same reason.

EPA's own Scientific Advisory Panel (SAP) concluded that the results of the three cohort studies and the animal studies together indicate that "maternal chlorpyrifos exposure would likely be associated with adverse neurodevelopmental outcomes in humans" (PRA at 33). Importantly, the SAP also warned that "exposure to multiple cholinesterase-inhibiting pesticides or other neurotoxicants might result in additive or interactive effects" (PRA at 33). EPA reports that follow-up analyses by the Columbia University research team, the authors of one of the epidemiology studies (the Columbia study) reviewed by EPA, proved that the adverse impact of chlorpyrifos on cognitive development is not due to other anticholinesterase pesticides"(PRA at 33, footnote 4). All the science points to the fact that: 1) chlorpyrifos exposure during pre-birth development leads to adverse neurological damage, and 2) exposure to chlorpyrifos and other OP pesticides can be as bad or worse than exposure to chlorpyrifos alone. Therefore, EPA must not delay banning chlorpyrifos just because other OP pesticides are also harmful and the proportion of risk attributable to each OP pesticide may be unclear.

B. FQPA Safety Factor - *EPA's proposal to remove the 10X FQPA factor is unlawful and unscientific.*

The Food Quality Protection Act of 1996 (FQPA) requires EPA to give special consideration to the health of infants and children in regulating pesticides, and EPA must "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure

to the pesticide chemical residue."[1] In assessing the risks of pesticide exposure of infants and children, EPA must evaluate "available information concerning the cumulative effects on infants and children of such residues and other substances that have a common mechanism of toxicity."[2] It further mandates that EPA apply a 10X margin of safety to take into account potential pre- and postnatal toxicity and completeness of the toxicity and exposure databases.[3] EPA is to use 10X as the default safety factor; EPA can use a different margin of safety "only if, on the basis of reliable data, such margin will be safe for infants and children."[4]

In its previous 2000 assessment, in which the RfD was largely based on plasma and red blood cell (RBC) cholinesterase inhibition (ChEi) data from adult rodents, EPA retained the 10X FQPA factor (see Table 1 below). Now, EPA is proposing to remove the 10X FQPA factor in the 2011 assessment (by reducing it to 1X) for all oral exposures (acute and chronic) to chlorpyrifos and the chlorpyrifos oxon, because the risk estimates are now derived from juvenile and pregnant rats, and because it believes that it has supporting data from a developmental neurotoxicity study (DNT) in pregnant rats (MRID 44556901). (PRA at 9) We strongly oppose this argument and EPA's action to remove the protective FQPA factor, for the reasons described below.

Table 1 – Summary of dietary risk estimates and toxicological effects; comparison of 2000 and 2011 assessments

	Acute dietary risks	Chronic dietary risks
In 2000	Food and water PoD=0.5 mg/kg/day; 10XFQPA. **aPAD=0.0005 mg/kg/day** Based on plasma and RBC ChEi from adult rats	Food and water PoD=0.03 mg/kg/day 10XFQPA **cPAD=0.00003 mg/kg/day** Based on plasma and RBC ChEi from adult rats
In 2011 PRA	Food PoD=0.36 mg/kg/day; 1XFQPA **aPAD=0.0036 mg/kg/day** based on RBC ChEi in rat pups; Water PoD (oxon)=0.05 mg/kg/day 1XFQPA **aPAD=0.0005 mg/kg/day** based on CCA study (oxon) of RBC ChEi in male rat pups	Food PoD=0.03 mg/kg/day 1XFQPA **cPAD=0.0003 mg/kg/day** based on RBC ChEi in rat dams Water PoD (oxon)=0.011 mg/kg 1XFQPA **cPAD=0.00011 mg/kg/day** based on CCA study (oxon) of RBC ChEi in adult female rate

Note- information excerpted from PRA Table 8 (p. 47), Table 9 (p. 49), Table 17 (p. 66), and Table 18 (p. 67)

There are a number of very significant limitations to these data that obligate EPA to retain the 10X FQPA factor. First, both the 2000 and 2011 assessments rely on RBC ChEi measurements rather than brain measurements, which introduces a significant amount of uncertainty.

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[1] Federal Food, Drug and Cosmetic Act, FFDCA, § 408(b)(2)(C)(ii)(I)
[2] FFDCA § 346a(b)(2)(C)(i)(III)
[3] FFDCA § 408(b)(2)(C)
[4] FFDCA § 408(b)(2)(C)

Compared with RBC ChEi measurements, brain measurements are preferred because they have less variability (smaller confidence intervals), represent a direct measurement of the common mechanism of toxicity, and measure the endpoint of interest, which is central nervous system impacts.[5] (PRA at 39) In fact, the SAP expressed a preference for brain measurements over RBC or plasma measurements in its 2002 report.[6] Although we support EPA's use of measurements from juvenile animals, EPA has not reduced its uncertainty enough to justify eliminating the FQPA factor – the uncertainties that remain are significant.

Second, the DNT study (MRID 44556901) that EPA is using to support removal of the 10X FQPA factor is classified as "guideline-unacceptable" because it failed to identify either a NOAEL or LOAEL for the offspring. (PRA at 17) In other words, this study is completely unacceptable to support any reduction or removal of the 10X FQPA factor. In her review of the study, EPA scientist Sue Makris noted exposure-related effects on nervous system development including delayed sexual maturation, altered startle reflex, and statistically significant decreases in brain weight at postnatal day 11 through 62 across several brain regions. Dr. Makris suggested that the decreased brain weight possibly reflected a reduction in the number of neurons that is obscured by later additional brain mass through cell growth and other growth processes.[7] EPA is currently reviewing the DNT study and analyzing additional morphometric data (PRA at 17, 37). However, it is unclear how this analysis will help to make the study more useful. The SAP advised that "histological assessment and morphometric measurements used in the DNT have significant limitations and cannot detect changes in the network organization of the brain or other possible changes" that Dr. Makris and other EPA scientists were concerned about (PRA at 37). The SAP instead recommended unbiased stereology analysis to determine cell number and tissue volume changes. (PRA at 37.) NRDC would like EPA to address whether the Agency is following the SAP's recommendation. If EPA is not doing this, NRDC requests that EPA explain why not. NRDC also requests that EPA explain why it is analyzing more morphometric data, despite the SAP's determination that this would not be helpful. Finally, the DNT is both "unacceptable" and inadequately sensitive, and cannot be used to support reducing the FQPA factor.

The DNT study is faulty and inadequate – and its presence does not justify removing the 10X FQPA factor. In fact the study supports retaining the 10X factor because of the unique vulnerability of early life stages to permanent or lasting damage from early-life exposure to chlorpyrifos.

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[5] EPA report. Status of cumulative risk assessment for organophosphate pesticides. EPA OPP. January 15, 2002. (See relevant passages at 24, 37). http://www.epa.gov/oppsrrd1/cumulative/files/guidefinal_4-new.pdf

[6] EPA report. Status of cumulative risk assessment for organophosphate pesticides. EPA OPP. January 15, 2002. (See relevant passages at 24, 37). http://www.epa.gov/oppsrrd1/cumulative/files/guidefinal_4-new.pdf

[7] Makris et al, 11/12/1998. As reported in NRDC comments on chlorpyrifos 12/27/1999. http://ecologic-ipm.net/NRDC_chlorpyrifos.pdf

C. Dietary Assessment - *Exposures from food uses still exceeds safe levels*

In 2001 when EPA negotiated with the pesticide industry to allow most agricultural uses of OPs in exchange for the cancellation of household uses (except contained ant and roach baits), it seemed like a good first step, since household uses did account for major exposures and poisoning incidents in children. Since that time, numerous studies have shown that most of children's exposures to these chemicals now come from food and drinking water. Scientific researchers are finding that the amounts of these chemicals that many children get from food alone are significant – in the range of the levels that may affect brain development.[8] What's more, researchers have found that in families that switch temporarily to a diet containing organic fruits and vegetables, the levels of these chemicals in children's urine plummets to near-zero levels .[9] Malathion and chlorpyrifos, both OP pesticides, were particularly high in children eating conventional diets. [10] The study's findings support the National Research Council's 1993 report showing that dietary intake of pesticides represents a major source of exposure for infants and young children – this was the study that provided important foundational support for the passage of the FQPA.[11]

EPA's acute risk estimates from food alone (not drinking water) indicate that all populations are below the Agency's level of concern (aPAD), even for the most highly exposed population (children 1-2 yrs) (see Table 13, PRA at 63). Children 1-2 years old are at 9% of the aPAD (0.0036 mg/kg/day), presuming a 1X FQPA factor; this would be 90% if EPA had used the legally-required 10X FQPA where children are exposed, thus getting close to the level of concern. Note that the acute and chronic levels of concern (aPAD and cPAD values) for exposures from food have been weakened by 7-fold and 10-fold respectively from the old 2000 values to the 2011 values in the PRA (see Table 2 below)

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[8] Curl CL, Fenske RA, Elgethun K. Organophosphorus pesticide exposure of urban and suburban preschool children with organic and conventional diets. Environ Health Perspect. 2003 Mar;111(3):377-82. http://www.ncbi.nlm.nih.gov/pubmed/12611667

[9] Lu C, Barr DB, Pearson MA, Waller LA. Dietary intake and its contribution to longitudinal organophosphorus pesticide exposure in urban/suburban children. Environ Health Perspect. 2008 Apr;116(4):537-42. http://www.ncbi.nlm.nih.gov/pubmed/18414640

[10] Lu C, Barr DB, Pearson MA, Waller LA. Dietary intake and its contribution to longitudinal organophosphorus pesticide exposure in urban/suburban children. Environ Health Perspect. 2008 Apr;116(4):537-42. http://www.ncbi.nlm.nih.gov/pubmed/18414640

[11] NRC report. Pesticides in the diets of infants and children. National Research Council, National Academy Press. 1993

Table 2 – Comparison of 2000 and 2011 assessments

	Acute dietary risks	Chronic dietary risks
In 2000	10XFQPA **Food and water aPAD=0.0005 mg/kg/day**	10XFQPA **Food and water cPAD=0.00003 mg/kg/day**
In 2011 PRA	1XFQPA Food aPAD=0.0036 mg/kg/day **7-fold weaker than old value** 1XFQPA Water aPAD=0.0005 mg/kg/day **Same as old value**	1XFQPA Food cPAD=0.0003 mg/kg/day **10-fold weaker than old value** 1XFQPA Water cPAD=0.00011 mg/kg/day **3-fold more protective than old value**

Note- information excerpted from PRA Table 8 (p. 47), Table 9 (p. 49), Table 17 (p. 66), and Table 18 (p. 67). The water assessment in the 2011 PRA uses the oxon rather than the parent chlorpyrifos as the residue of concern. The water assessment presumes 100% conversion of parent to oxon, and the oxon is more toxic than the parent compound.

The published data show that chlorpyrifos contributes measurably to children's overall pesticide exposure from foods. EPA must not delay action to ban chlorpyrifos, just because other OP pesticides are also harmful and the proportion of risk attributable to each OP pesticide may be unclear.

D. Drinking water assessment – *exposures exceed level of concern for ALL infant scenarios*

In the 2000 assessment for drinking water, EPA assumed that parent chlorpyrifos was the residue of concern, whereas the 2011 PRA considers the oxon as the residue of concern, and assumes that 100% of chlorpyrifos is converted to oxon. The chlorpyrifos oxon is more toxic than the parent chlorpyrifos. NRDC supports this approach, which is based on reasonable assumptions that are health-protective and supported by science.

For drinking water alone (no food exposure), EPA's risk estimates indicate that infants (children under 1 year old) are ingesting chlorpyrifos at levels that exceed the Agency's level of concern (i.e. are likely to be unsafe) for all scenarios (see Table 3 below). EPA provided risk estimates using three crop-pesticide scenarios, a low-end scenario (sugar beets), a mid-range scenario (corn) and a high-end scenario (grapes). For each scenario, EPA calculated the exposure estimates for the average pesticide application rate and the maximum application rate allowed on the label. Risk estimates are presented for acute exposures at the 99.9th percentile population, and with no FQPA (1X) factor. As seen in the table below, ALL infant scenarios exceed EPA's level of concern (exposure as percent of aPAD), some by more than an order of magnitude. Grape crops grown with the maximum allowable application rate of chlorpyrifos exceed the aPAD by 27-fold (2700%), and corn grown with the maximum application rate exceeds the aPAD by 7.7-fold (770%) for infants. ALL populations (infants, children, and women of reproductive age) exceeded EPA's level of concern for grapes (the high-end scenario) grown with the maximum allowable application rate of chlorpyrifos. And women of reproductive age exceeded the aPAD at the maximum application rates for both corn (2-fold; 200% aPAD) and

grapes (7-fold; 710% aPAD). (PRA at 61) EPA selected these crop scenarios because "there is a large amount of chlorpyrifos applied to these crops per year" throughout the country, making them very realistic and common exposure scenarios.(PRA at 10.)

Table 3: Summary of Preliminary Acute Drinking Water Only Exposures and Risk (at the 99.9th percentile exposure for chlorpyrifos oxon; using 1X FQPA (Exposure is % aPAD).

	Low-end scenario (sugar beet)		**Mid-range scenario (corn)**		**High-end scenario (grape)**	
	Avg rate	Max rate	Avg rate	Max rate	Avg rate	Max rate
General population	99%	61%	38%	240%	19%	810%
Infants	340%	210%	120%	770%	59%	2700%
Children 1-2 yrs	140%	89%	54%	340%	26%	1200%
Children 3-5 yrs				310%	24%	1100%
Females 13-49 yrs	88%	54%	32%	200%	16%	710%

Note – above table is excerpted from Table 14 (PRA at 61). Highlighting indicates that exposure exceeds aPAD. Chlorpyrifos oxon aPAD (including 1X FQPA) is 0.0005 mg/kg/day.

If EPA had appropriately applied a 10X FQPA factor to the aPAD instead of 1X, all of these would have been 10 times higher, in which case ALL scenarios, for ALL population groups, at ALL application rates would have exceeded the aPAD. For example, for the maximum application rate grape scenario, infants would exceed the aPAD by 270-times (27,000%), children 1-2 yrs would exceed it by 120-times (12,000%), and children 3-5 yrs would exceed it by 110-times (11,000%). These outrageously high numbers are the estimates that EPA would have come up with had it applied the FQPA appropriately, as described in these comments. These values more accurately represent the true elevated risk that some families experience, including the hundreds of children with permanent neurological effects reported in the epidemiology studies that EPA has identified but failed to incorporate.

E. Residential bystander post-application inhalation exposures too high

We applaud EPA's efforts to develop a bystander exposure assessment for chlorpyrifos from field volatilization based on ambient and application site air monitoring data. EPA used a 10X FQPA factor for acute inhalation exposures (toxicity endpoint based on lung ChEi in a 6 hour study), but eliminated it (1X) for short- and intermediate-term inhalation scenarios. (PRA at 48)

The results of EPA's volatilization analysis for residential bystanders indicate that one-quarter (4 of 24) of the acute ambient air concentrations resulted in risk estimates that exceed the level of concern[12] (MOE<300). Over half (3 of 5) of the acute application site air concentrations resulted in risk estimates of concern (MOE<300), and almost all (4 of 5) of the short-term and

[12] Note: Residential risk is measured by a Margin of Exposure (MOE), which measures how close the residential exposure comes to the no-effect level, or NOAEL, from animal studies. Generally MOEs greater than 100 do not exceed the Agency's level of concern (this incorporates the standard uncertainty factors of 10X for interspecies variability and 10X for intraspecies variability).

intermediate-term application site air concentration assessments resulted in risk estimates of concern (MOE<30). (PRA at 72, 73.)

Some of the spray drift risk estimates calculated by EPA were very concerning. For example, acute adult and child inhalation (spray drift) exposure from ground mosquito treatments were 17, which poses an unacceptably high risk, 17-fold more risky than the target MOE of 300.

These data and assessments point strongly to both drift and volatilization as a significant pathway of exposure for people, including infants, children, and pregnant women. In some cases, EPA has shown that this pathway alone exceeds levels of concern and therefore presents an unacceptable risk. These risk estimates are derived from actual air monitoring data, rather than models, meaning that these risks are real and experienced by real people who are not being adequately protected under current pesticide practices.

Pesticide drift is a significant pathway of exposure as shown by a study published in Journal of Occupational and Environmental Medicine in 2011 by a collaborative team of researchers from Kaiser Permanente Center for Health Research, Fred Hutchinson Cancer Research Center, Rollins School of Public Health, and the University of Washington. The study examined the relationship between pesticide metabolites in urine and residential proximity to farmland. The authors found a positive correlation such that increasing pesticides in the bodies of the subjects was associated with living closer to farmland.[13]

Another study also published in 2011 by a research team from the University of California at Berkeley reports data showing that both dietary intake and proximity to farmland is correlated with measurable levels of OP pesticides including chlorpyrifos in infants and toddlers. The authors measured OP pesticide metabolites in the urine of about 400 children and found that for infants the levels were highest in the spring and summer when pesticides are being applied to farmland, whereas for the toddlers (1-2 yrs old) levels were highest when children lived less than 60 meters from an agriculture field.[14]

Approximately one million children of farm workers live in the United States. More than 320,000 children under the age of six live on farms in the United States.[15] Farm children,

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[13] Coronado GD, Holte S, Vigoren E, Griffith WC, Barr DB, Faustman E, Thompson B. Organophosphate pesticide exposure and residential proximity to nearby fields: evidence for the drift pathway. J Occup Environ Med. 2011 Aug;53(8):884-91. http://www.ncbi.nlm.nih.gov/pubmed/21775902

[14] Bradman A, Castorina R, Barr DB, Chevrier J, Harnly ME, Eisen EA, McKone TE, Harley K, Holland N, Eskenazi B. Determinants of organophosphorus pesticide urinary metabolite levels in young children living in an agricultural community. Int J Environ Res Public Health. 2011 Apr;8(4):1061-83. http://www.ncbi.nlm.nih.gov/pubmed/21695029

[15] Trouble on the Farm: Growing up with pesticides in agricultural communities. G. Solomon. Report of the Natural Resources Defense Council. 1998. Available at http://www.nrdc.org/health/kids/farm/farminx.asp

especially the children of farmworkers, come in contact with pesticides through residues from their parents' skin and clothing, soil and dust tracked into their homes, contaminated soil and other surfaces in areas where they play, food eaten directly from the fields, drift from agricultural pesticide applications, contaminated well water, and breast milk from exposed parents.[16] These exposure routes can be very significant, even dominant, for these children.

It is important that EPA incorporate these independent scientific studies, which provide real-world support for EPA's findings that drift and volatilization represent significant pathways of exposure for residential bystanders.

F. Occupational assessment – *workers unsafe*

Current occupational pesticide practices do not adequately protect workers. EPA reports that, of the 350 exposure scenarios assessed for the PRA, 62%, or 216 scenarios, had risk estimates that exceed the level of concern even with some level of personal protective equipment (PPE). A whopping 25%, or 80 scenarios, resulted in risk estimates of concern at ALL levels of PPE and engineering controls. (PRA at 14, 83). The risk estimates were even greater when EPA used real-world biomonitoring data instead of surrogate or modeled data to calculate exposures. (PRA at 15).

Importantly, unsafe occupational exposures to pesticides harm more than just adult workers: it also harms their families. The children of farmworkers are at greater risk than the general population for pesticide-related cancer from *in utero* and early life pesticide exposures through their parents. In a meta- analysis of 31 studies examining the link between parental occupational pesticide exposures and incidence of childhood leukemia, the authors found that risk of childhood leukemia was associated with prenatal maternal occupational pesticide exposure.[17] The authors found a statistically significant doubling in the risk of childhood leukemia if the child was exposed prenatally to pesticides through maternal occupational pesticide exposure (OR = 2.09; 95% CI, 1.51-2.88). The link was even stronger if the mother's exposure was high (OR = 2.45; 95% CI, 1.68-3.58), and for farm-related exposures (OR = 2.44; 95% CI, 1.53-3.89) as opposed to mixed or unknown pesticide exposure place. Childhood leukemia risk also was found to be significantly associated with prenatal maternal occupational exposure to insecticides (OR = 2.72; 95% CI, 1.47-5.04) and herbicides (OR = 3.62; 95% CI, 1.28-10.3). These studies and findings show the damaging and permanent risks that may be associated with early life exposures to pesticides through the mother's occupational exposure

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[16] Bradman A, Harnly ME, Draper W, Seidel S, Teran S, Wakeham D, Neutra R 1997. Pesticide exposure to children from California's Central Valley: results of a pilot study. J. Expo. Anal. Environ. Epidemiol. 7, 217-234

[17] Wigle DT, Turner MC, Krewski D. 2009 A systematic review and meta-analysis of childhood leukemia and parental occupational pesticide exposure. Environmental Health Perspectives. Oct. 117 (10): 1505-13

while pregnant. The risks to the fetus may be much more severe than the risks to the pregnant mother, even from the same exposure scenario.

A companion meta-analysis of childhood leukemia and home-pesticide use was published in 2010 by the same authors.[18] In that report, they found that fetal exposures to pesticides used in the home was associated with a 54% increase in risk of childhood leukemia (OR = 1.54; 95% CI, 1.13–2.11). The risk was even more strongly associated with pre-birth insecticide exposure (OR = 2.05; 95% CI, 1.80–2.32) and herbicide exposure (OR = 1.61; 95% CI, 1.20–2.16).

The 2004 National Institutes of Health (NIH) Agriculture Health Study, a robust prospective epidemiology study of pesticide applicators in the Midwest, reported increased childhood cancer risk associated with occupational exposure of the parents to pesticides (N=50 incident childhood cancer cases from a total of over 17,000 children of Iowa pesticide applicators).[19] In this study, the risk of all childhood cancers in farm children whose parent is a professional pesticide applicator increased by 36% compared to other similar children (SIR=1.36; 95% CI=1.03-1.79). Risk of all lymphomas combined was doubled (SIR=2.18; 95% CI =1.13-4.19) as was the increased risk of Hodgkin's lymphoma (SIR=2.56; 95% CI =1.06-6.14). Interestingly, an increased cancer risk was found in the children of fathers who did not use work gloves, compared with children whose fathers used the gloves when applying pesticides, (OR=1.98; CI=1.05-3.76). This study demonstrates that the children of pesticide applicators may be at greater risk than the general population for some kinds of cancers.

These data together provide strong support for EPA to address the risks posed to fetuses, infants, and young children from parental or direct occupational exposure to pesticides.

EPA must finalize its policy to provide a measure of protection to the children of farm workers

On December 9, 2009 EPA announced in the Federal Register that it would accept public comments on its *Policy Paper on Revised Risk Assessment Methods for Workers, Children of Workers in Agricultural Fields, and Pesticides with No Food Uses*. The comment period was extended to April 12, 2010 (75 Fed Reg 6031 (Feb 5, 2010)). To date, the policy still has not been finalized. This long overdue policy addresses the risks to agriculture workers and their children who have been shown to experience significantly higher pesticide exposures than the average child. The policy extends the now-established methods of conducting an aggregate and cumulative risk assessment from the general population to agriculture workers and their children. Specifically, the policy will require that when EPA assesses risks from pesticide

[18] Turner MC, Wigle DT, Krewski D. Residential pesticides and childhood leukemia: a systematic review and meta-analysis. Environ Health Perspect. 2010 Jan;118(1):33-41. PubMed PMID: 20056585; PubMed Central PMCID: PMC2831964.

[19] Flower KB, Hoppin JA, Lynch CF, Blair A, Knott C, Shore DL, Sandler D. 2004 Cancer risk and parental pesticide application in children of agricultural health study participants. Environmental Health Perspectives. April; 112(5) 631-5

exposure to agriculture workers and their children, EPA will now quantitatively include a calculation of the risks from aggregate exposures to the same pesticide used in multiple settings, the risks from cumulative exposures to multiple pesticides that share a common toxic mechanism, and the unique risks posed to infants and children due to their potentially increased sensitivity to pesticides. These factors (aggregate and cumulative exposure, and sensitive subpopulations) are already required by the FQPA to be included in EPA's pesticide risk assessments, and should include the children of farmworkers.

We encourage EPA to finalize this important policy as soon as possible, to extend a measure of protection to the most highly pesticide exposed children in the country.

CONCLUSION

Weight of evidence is based on quality, not quantity

The "weight of evidence" should not be measured in pounds of paper, i.e. number of studies that do find an effect versus those that don't. In fact, many studies sponsored by the chemical manufacturer fail to find an effect when independent and government-funded studies actually do show harmful effects. For detailed documentation proving a long history of industry's attempts to misrepresent or suppress scientific evidence that its products are harmful, see the following books:

> Doubt is Their Product: How Industry's Assault on Science Threatens Your Health (Oxford University Press, 2008) by David Michaels, PhD, MPH, now Assistant Secretary of Labor for Occupational Safety and Health.
>
> Poisoned Profits: The Toxic Assault on Our Children (Random House, 2008) by Philip Shabecoff and Alice Shabecoff.
>
> Secret History of the War on Cancer (Basic Books, 2007) by Devra Davis, PhD.
>
> Deceit and Denial: The Deadly Politics of Industrial Pollution (University of California Press, 2002) by Gerald Markowitz and David Rosner.

These books use as examples many industrial chemicals and pesticides, including: arsenic, asbestos, atrazine, bendectin, benzene, beryllium, chromium VI, cigarette smoke, dioxin, fen-phen, food additives, formaldehyde, lead, mercury, PFOA, popcorn lung (diacetyl), and vinyl chloride.

EPA must not let chlorpyrifos be added to the list of chemicals for which regulatory agencies ignored important data showing harm.

Therefore, EPA should revoke all tolerances and cancel all remaining uses of chlorpyrifos.

Thank you for your consideration of these comments.

Respectfully,

Jennifer Sass *Mae Wu*

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ATTACHMENT 4

EARTHJUSTICE

ALASKA CALIFORNIA FLORIDA MID-PACIFIC NORTHEAST NORTHERN ROCKIES
NORTHWEST ROCKY MOUNTAIN WASHINGTON, DC INTERNATIONAL

May 7, 2013

Via Electronic Filing

Mr. Richard P. Keigwin, Jr., Director
Pesticide Reevaluation Division
Office of Pesticide Programs
Environmental Protection Agency
1200 Pennsylvania Avenue, NW
Washington, D.C. 20460-0001

Docket ID # EPA–HQ–OPP–2008–0850

Re: Chlorpyrifos Registration Review; Preliminary Evaluation of the Potential Risk From Volatilization

Dear Mr. Keigwin:

On behalf of United Farm Workers, Pineros y Campesinos Unidos del Noroeste, Farmworker Justice, California Rural Legal Assistance Foundation, Farmworker Association of Florida, Pesticide Action Network of North America, and Natural Resources Defense Council, we submit the following comments on EPA's Preliminary Evaluation of the Potential Risk from Volatilization of Chlorpyrifos (Preliminary Volatilization Evaluation), first noticed in the Federal Register on February 6, 2013. *See* 78 Fed. Reg. 8522 (Feb. 6, 2013).

Chlorpyrifos is a highly toxic organophosphate pesticide used widely in agriculture to protect food and feed crops from insect pests. In 1996, Congress adopted the Food Quality Protection Act (FQPA) tc strengthen protections for children in the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and the Federal Food, Drug, and Cosmetic Act (FFDCA), requiring that by 2006 EPA review its pesticide registration decisions to ensure that "no harm" will result to infants and children from "aggregate exposure" to pesticides. 21 U.S.C. § 346a(b)(2)(C)(ii)(I), (II); *see also id.* § 346a(q)(1)(C); 7 U.S.C. § 136(bb).

In 2000, recognizing that chlorpyrifos poses unacceptable risks to children, EPA entered into an agreement with chlorpyrifos manufacturers to eliminate virtually all residential uses of the pesticide. Unfortunately, EPA allowed continued use of chlorpyrifos in agricultural settings. As a result, workers and their families, including possibly thousands of vulnerable children, continue to be exposed to this toxic chemical—not only in fields but also in nearby homes, backyards, and schools.

Our clients have long advocated that EPA must close this glaring loophole. In 2007, Natural Resources Defense Council and PANNA petitioned EPA to revoke all tolerances and cancel all registrations for chlorpyrifos, citing among other problems EPA's failure to assess drift (whereby sprayed pesticide particles drift to nearby areas) and volatilization (whereby pesticide particles vaporize and are transported away from fields where they are applied) and their risks to

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B. The FQPA Amended FIFRA and the FFDCA to Require Pesticide Residues On Food To Be Safe.

Congress adopted the FQPA in 1996, amending FIFRA and the FFDCA in several key ways designed to strengthen protections against exposures to pesticide residues on food, particularly for infants and children.

First, the FQPA requires EPA to ensure that there is a reasonable certainty that "no harm" will result from "aggregate exposure" to the pesticide, including all anticipated dietary exposures and "all other exposures for which there is reliable information." 21 U.S.C. § 346a(b)(2)(A)(ii). Second, the FQPA requires EPA to assess "cumulative effects" of a pesticide residue in combination with "other substances that have a common mechanism of toxicity." *Id.* § 346a(b)(2)(D)(v), (b)(2)(C)i)(III).

Third, the FQPA requires EPA to scrutinize carefully risks to infants and children from exposure to pesticides. In addition to ensuring the safety of the general population, EPA must "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure" to pesticides. 21 U.S.C. § 346a(b)(2)(C)(ii)(I), (II). The FQPA adopts a presumptive tenfold margin of safety "for the pesticide chemical residue *and other sources of exposure*," in order to "take into account potential pre- and post-natal toxicity…" *Id.* § 346a(b)(2)(C)(ii) (emphasis added). EPA may use a different margin of safety only if reliable data shows that such margin will be "safe" for infants and children. *Id.*

Finally, the FPQA integrated FIFRA and the FFDCA by requiring EPA to use the FQPA's safety standard as a criterion in FIFRA registration actions as to pesticide uses that result in dietary risks from residues in or on food. 7 U.S.C. § 136(bb)(2). Thus, in order to show that a pesticide has no "unreasonable adverse effects" and register a pesticide under FIFRA, a registrant must also show that the pesticide and its prescribed tolerance are "safe" under the FQPA. 7 U.S.C. § 136(bb); 21 U.S.C. § 346a(b)(2)(A)(ii). Congress gave EPA until 2006 to bring pesticide registrations into compliance with the FQPA's standards. 7 U.S.C. § 136a-1(g)(2); 21 U.S.C. § 346a(q)(1)(C).

II. ANALYSIS

A. The Preliminary Volatilization Evaluation Supports Canceling Chlorpyrifos Registrations and Revoking Chlorpyrifos Tolerances.

In the Preliminary Volatilization Evaluation, EPA found that chlorpyrifos applied to fields can volatilize and harm people nearly a mile away (and likely farther)[1] depending on several factors, including application rate, field size, and percentage of the population exposed to harmful levels. Specifically, EPA concluded that "[g]iven the current available information and the state of the science concerning volatilization of pesticides, this preliminary risk assessment

[1] The Preliminary Volatilization Evaluation uses a model known as "PERFUM" to estimate buffer distances that would reduce exposure levels to below levels of concern based on different application rates. Preliminary Volatilization Evaluation at 34, Table 10. However, PERFUM does not produce buffer zones greater than 4,724 feet. *Id.* Thus, "buffer zones for cases where the 4724 ft (1440 m) limit is reached may be very large." *Id.*

04, 305 n.38; *Envtl. Defense Fund, Inc. v. EPA*, 510 F.2d 1292, 1297 (D.C. Cir. 1975) (FIFRA "does not require [EPA] to establish that the product is unsafe," but rather places the burden of establishing the safety of a product "at all times on the applicant and registrant"); *see also Envtl. Defense Fund, Inc. v. EPA*, 465 F.2d 528, 532 (D.C. Cir. 1972) (burden of proof rests on applicant for pesticide registration); *Envtl. Defense Fund, Inc. v. Ruckelshaus*, 439 F.2d 584, 593 (D.C. Cir. 1971) (in cancellation proceedings, "manufacturer [has] the burden of proving the safety of his product"). Thus, the fact that a pesticide has an active registration is not a defense to cancellation. 7 U.S.C. § 136a(f)(2); *Reckitt,* 762 F.Supp.2d at 45.

Here, EPA has found that risks of concern are exceeded for bystanders exposed to chlorpyrifos volatilization. Prelim. Vol. Eval. at 55. As explained below, EPA's findings with respect to volatilization, in conjunction with other harms identified in the Preliminary Risk Assessment, provide indisputable evidence that chlorpyrifos *is* harmful; therefore, EPA must cancel chlorpyrifos registrations and tolerances because chlorpyrifos use is not "safe."

2. The Preliminary Volatilization Evaluation Undermines EPA's Prior Determination That Cumulative Exposures To Chlorpyrifos Are Safe.

In 2006, EPA approved continued use of chlorpyrifos in agriculture based on a cumulative risk assessment that concluded that organophosphate use cumulatively would result in "no harm" to the public. *See* Letter from Debra Edwards, EPA, Director of Special Review and Registration Division, to Jim Jones, EPA, Office of Pesticide Programs (Jul. 31, 2006); *see also* EPA, *Organophosphorous Cumulative Risk Assessment – 2006 Update* (Jul. 31, 2006) [hereinafter, *2006 Cumulative Risk Assessment*]. However, EPA's cumulative risk assessment failed to account for important ways in which members of the public are exposed to chlorpyrifos, including pesticide drift and volatilization. Thus, EPA did not consider the very real harm faced by farmworker communities who live near agricultural fields and routinely are exposed to chlorpyrifos when it is applied to fields.

Now, as part of EPA's review of chlorpyrifos registrations, EPA is finally taking steps to comply with its obligation to assess all of the risks of chlorpyrifos exposure, including pesticide drift and volatilization. Mounting evidence assessed by EPA contradicts EPA's prior determination that chlorpyrifos can be used safely. As discussed, EPA's Preliminary Volatilization Evaluation discloses that risks of concern are exceeded for bystanders exposed to chlorpyrifos volatilization. Prelim. Vol. Eval. at 55. The assessment also shows that for many crops extremely large (and therefore not feasible) buffers would be required to ensure that risks of concern are not exceeded for bystanders, depending on the size of the field and the application rate. For example, for oranges, the average application rate is so high (greater than 2 pounds of active ingredient/acre ("Lbs. a.i./A")) that maximum buffers would need to be between 1,476 feet and 4,724 feet (if not greater), and whole field buffers would need to be between 623 feet and 2,838 feet, to ensure that bystander risks do not exceed the level of concern for 99 percent of the population.

In addition, our October 6, 2011 comments pointed out that EPA's Preliminary Risk Assessment determined that several subpopulations, especially infants, are exposed to levels of chlorpyrifos in drinking water that are not safe. *See* Letter from Patti Goldman, Earthjustice, *et*

particular chemical.[3] In the 2006 Cumulative Risk Assessment, EPA explained that for "highly refined exposure assessments," such as for dietary exposures, EPA has used the 99.9th percentile as a starting point for assessing safety. *2006 Cumulative Risk Assessment* at 13 (citing EPA, *Choosing a Percentile of Acute Dietary Exposures as a Threshold of Regulatory Concern* (Mar. 16, 2000)). For exposure assessments that rely on highly conservative estimates of pesticide residues, however, "EPA has generally based its safety determinations on the estimated exposure of the 95th percentile of the population." *Id.* at 12.

For the 2006 Cumulative Risk Assessment, used to support EPA's reregistration of chlorpyrifos in 2006, EPA ultimately applied a hybrid approach in which children nationally ages 3-5 were protected at the 99.89th percentile, and children in Florida ages 1-5 were protected at between 99.5th and 99.75th percentiles. *Id.* Thus, even though the cumulative risk assessment relied on a mix of refined and unrefined exposure data, all of the exposure percentiles EPA used were above 99.5 percent, meaning that less than 0.5% of the population could be exposed to harmful levels of organophosphate pesticides.

Here, EPA may not rely on an even lower 95th percentile to fulfill its duty to ensure no harm under the FQPA. Such an approach would mean that hundreds or even thousands of people could be harmed by chlorpyrifos volatilization contrary to the FQPA's no-harm mandate. Indeed, in California, heavy pesticide use is not confined to isolated rural areas where few people are exposed. To the contrary, *over one million people* in the San Joaquin Valley live, work or go to school in close proximity to agricultural areas where pesticides are applied.[4] Many of these residents are children—in just three California counties (Kern, Fresno and Tulare), 22,000 children attend schools near sites of heavy pesticide use. *Id.* EPA may not simply write-off 5% of these residents and children and justifiably conclude that chlorpyrifos causes "no harm."

Numerous uncertainties in the Preliminary Volatilization Evaluation further weigh in favor of a more health-protective approach, Prelim. Vol. Eval. at 47-54, including:

1) the PERFUM model EPA used to estimate potential buffers to minimize volatilization health risks has not been validated for non-fumigant pesticides, including chlorpyrifos;

2) the key studies relied on to estimate volatilization risks used chlorpyrifos application rates that are much lower than the maximum allowed for several crops, Prelim. Vol. Eval. at 49;

3) the Preliminary Volatilization Evaluation, Prelim. Vol. Eval. at 48, relies on an acute inhalation study involving testing of only adult female rats, when EPA's 2011 Preliminary Risk Assessment found that juvenile rats at post-natal day 11 represented

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[3] EPA, 2001 GENERAL PRINCIPLES FOR PERFORMING AGGREGATE EXPOSURE AND RISK ASSESSMENTS 10 (Nov. 28, 2001).

[4] Environmental Working Group, *Every Breath You Take: Airborne Pesticides in the San Joaquin Valley* (Jan. 2001) (EWG Pesticides Report) 1 (attached hereto as Exhibit A).

ancestry—approximately 83% of farmworkers nationwide identify themselves as Latino.[5] The alfalfa study referenced in the Preliminary Volatilization Evaluation occurred in Los Banos, CA, in Merced County, where census data from 2011 shows that 55.7% of residents are of Hispanic or Latino origin.[6] The impacts of chlorpyrifos volatilization also will disproportionately harm minority children, who are more vulnerable to pesticide poisonings. In California, for example, over 73 percent of children attending schools within 1.5 miles of sites where at least 10,000 pounds of pesticides were applied in 1998 were non-white.[7]

In addition, chlorpyrifos volatilization will disproportionally affect low-income households. Nationwide, between 2001 and 2002 30% of farmworkers had a total family income below the federal poverty guidelines.[8] In California's San Joaquin Valley, in 2009 46.2% of households had incomes less than 2 times the federal poverty threshold, and 8.4% of households were living in severe poverty, with incomes less than 1/2 the federal poverty threshold.[9]

While it may be easy to waive off 5% in the abstract, these are real people, families and children who will suffer the consequences. Neither the FQPA, nor Executive Order 12898, permit such an approach.

b. EPA Must Account for Air Monitoring Studies Showing That Rural Areas Have Unsafe Levels of Chlorpyrifos.

Fifteen air monitoring studies presented to EPA during registration review confirm that harm to rural communities from chlorpyrifos drift and volatilization is not just an abstract proposition. As we noted in our October 2011 comments, the majority of these air monitoring studies were conducted in Tulare County, California and Yakima County, Washington, two rural counties that are predominantly low-income and communities of color. EJ Comments at 6. EPA's Preliminary Risk Assessment reviewed these fifteen studies and, although it discounted them for various reasons, determined that many of the air samples in those studies exceed EPA's levels of concern. *Preliminary Risk Assessment* at 71.

The air monitoring studies demonstrate that low-income and communities of color are bearing the brunt of chlorpyrifos use in agricultural areas as a result of drift and volatilization. The studies provide concrete proof that an EPA decision to allow 5% of rural communities to

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[5] U.S. DEP'T OF LABOR, FINDINGS FROM THE NATIONAL AGRICULTURAL WORKERS SURVEY (NAWS) 2001-2002: A DEMOGRAPHIC AND EMPLOYMENT PROFILE OF UNITED STATES FARM WORKERS, RESEARCH REPORT NO. 9 (March 2009) ("NAWS") 4 (attached hereto as Exhibit B).

[6] *See* United States Census Bureau, State and County QuickFacts, Merced County, California, *available at* http://quickfacts.census.gov/qfd/states/06/06047.html.

[7] EWG Pesticides Report at 11.

[8] NAWS at 47.

[9] JOINT CENTER FOR POLITICAL AND ECONOMIC STUDIES, PLACE MATTERS FOR HEALTH IN THE SAN JOAQUIN VALLEY: ENSURING OPPORTUNITIES FOR GOOD HEALTH FOR ALL 8 (March 2012) (attached hereto as Exhibit C).

adult rats, but there is no basis for assuming that the sensitivity in rats for oral exposures is identical to the sensitivity in rats for inhalation exposures.

In addition, we agree with the comment of the California Department of Pesticide Regulation (DPR), submitted to EPA on September 30, 2011 in regards to EPA's Preliminary Risk Assessment, that recent data suggest that developmental neurotoxicity may be a more sensitive endpoint then cholinesterase inhibition and that "[p]rotection against brain cholinesterase inhibition alone may be insufficient to protect against such effects." Letter from Gary Patterson, Ph.D., Dep't of Pesticide Regulation, *et al.* to Tom Myers, EPA, Office of Pesticide Programs 3 (Sep. 30, 2011) (attached hereto as Exhibit D). The recent data includes *in vitro* studies, as well as a Columbia University study that revealed an association between chlorpyrifos blood levels of >6.17 pg/ml during the third trimester and neurobehavioral deficits in children exposed *in utero*, where blood levels were less than the concentration expected to result in brain cholinesterase inhibition. *Id.* Based on the "unique susceptibility of infants and children," DPR recommended that EPA retain the FQPA's 10X safety factor to protect against developmental neurotoxicity. *Id.* For the same reasons, EPA should retain the FQPA's 10X safety factor when assessing risks of chlorpyrifos volatilization.

In short, EPA has not established "on the basis of reliable data" that no margin of safety is "safe" for infants and children. EPA must apply the FQPA's 10X safety factor for infants and children to its assessment of chlorpyrifos volatilization risk, in addition to any other uncertainty factors that apply.

III. CONCLUSION

EPA's Preliminary Volatilization Evaluation confirms that rural communities are being harmed by chlorpyrifos use in agricultural areas. These harms are borne disproportionately by low-income and communities of color that are exposed to chlorpyrifos as a result of volatilization. To comply with the FQPA and EPA's environmental justice duties under Executive Order 12898, EPA must revoke all chlorpyrifos tolerances and cancel all registrations for this dangerous pesticide.

Sincerely,

Erin M. Tobin

Erin Tobin
Irene Gutierrez
Counsel for United Farm Workers, *et al.*

cc: Steven Bradbury, Director, Office of Pesticide Programs (bradbury.steven@epa.gov)

No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

DECLARATION OF MARGARET REEVES IN SUPPORT OF SECOND PETITION FOR A WRIT OF MANDAMUS

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DECLARATION OF MARGARET REEVES

I, MARGARET REEVES, declare and state as follows:

1. I am a senior scientist at Pesticide Action Network of North America ("PANNA").

2. On April 12, 2012, I filed with this Court my declaration in an earlier petition for writ of mandamus challenging the U.S. Environmental Protection Agency's ("EPA") unreasonable delay in responding to the petition National Resource Defense Council ("NRDC") and PANNA submitted to EPA, which requested that EPA ban the use of chlorpyrifos and revoke all tolerances (allowable residue limits on food) for chlorpyrifos. That mandamus action was *In re Pesticide Action Network North America and Natural Resources Defense Council*, No. 12-71125, and I have attached my declaration in that case to this declaration as Attachment 1.

3. I have reviewed my previous statements to the Court in that matter and reaffirm them here. All information in my prior declaration remains true and accurate, and I incorporate all of that declaration as if set forth fully here. My prior statements apply with equal force to this renewed petition for writ of mandamus.

4. Since filing that declaration, I have remained in my capacity as a senior scientist for PANNA and have closely followed EPA's work in responding

to the 2007 Petition, along with ongoing research in the greater scientific community related to the study of chlorpyrifos and its harms.

5. PANNA filed comments on the chlorpyrifos interim re-registration eligibility decision on October 2, 2006. It was our hope that EPA would address these comments when it completed its cumulative risk assessment for the organophosphates, but EPA did not do so. PANNA and NRDC then filed this petition (and pursued a direct challenge to the re-registration decision) to compel EPA to address its serious omissions in its re-registration decision.

6. EPA released a preliminary human health risk assessment for chlorpyrifos in 2011, which acknowledges the need to address spray drift and volatilization drift and that studies show widespread effects resulting from chlorpyrifos exposure. PANNA filed comments on this assessment providing additional evidence and showing why EPA's assessment understates the risks to children from chlorpyrifos. Our comments are attached as Exhibit 1.

7. In my previous declaration at ¶¶ 7-11, I discussed the danger and frequency of chlorpyrifos poisonings, including a 2002 PANNA report I co-authored. Chlorpyrifos has continued to be associated with acute pesticide poisonings, and data on chlorpyrifos poisonings collected and released by California's Department of Pesticide Regulation show that chlorpyrifos poisonings remain unacceptably high.

a. The 2002[1] PANNA report showed that California's Pesticide Illness Surveillance Program ("PISP") had reported 156 chlorpyrifos poisoning cases between 1998 and 2000. We also noted that the reported poisonings likely represented only the tip of the iceberg, as many, probably most cases go unreported for myriad reasons including lack of familiarity among workers, residents, and physicians with signs and symptoms of pesticide-related illnesses and/or fear of retaliation among workers for reporting job-related incidents. We also pointed out that about half of all drift cases occurred when investigations determined that there had been no violations of pesticide use or worker safety regulations. In other words, the results demonstrated that the regulations themselves were inadequate to protect workers, and others, from pesticide exposure and associated poisonings.

b. More recent PISP data suggest that poisonings by agricultural use of chlorpyrifos continue albeit at apparently lower rates.

c. While most PISP cases are reported for workers, reports of direct acute poisonings among children exposed at school are not uncommon, with 34 cases reported (for all pesticides) between 2008 and 2011. The PISP reports of chlorpyrifos cases among workers in that time period totaled 62 with 49 attributed to drift exposure. There is a lag period of at least two or three years between incident and public reporting of PISP data, so while we believe incidents continue to occur, these are the most up-to-date data available.

d. A recent report of agricultural pesticides used near California schools showed that chlorpyrifos was the 8th most common highly hazardous pesticide applied within ¼ miles of public schools.[2]

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[1] Reeves, M., A. Katten and M. Guzmán, *Fields of Poison 2002: California farmworkers and pesticides*, Pesticide Action Network (2002).

[2] California Environmental Health Tracking Program, *Agricultural Pesticide Use Near Public Schools in California* (2014).

8. Organophosphate pesticides pose a high risk to people, and especially to fetuses, infants, and young children, but EPA's action to date demonstrates a double standard that results in unacceptable neglect of rural and farm children while suburban and urban children receive some necessary protections against exposure to chlorpyrifos. EPA took effective measures to cancel almost all residential uses of organophosphate pesticides, which have resulted in significant and measurable reduction in poisonings to children from roach baits, residential foggers or "bug bombs," and other homeowner uses. These protections, while necessary, do not address dangerous forms of exposure to chlorpyrifos and other organophosphate pesticides from spray drift and volatilization drift, which primarily affect children living in rural and farming communities. Often, the children affected are the children of farmworkers, meaning that the harm EPA allows falls disproportionately on children in low-income and minority communities. Any continued poisonings in light of the science underlying chlorpyrifos would be unacceptable, but this double standard is especially alarming because of the disproportionate nature of the harm on already overburdened communities. Rural and farm children should be accorded the same protections as other children from this dangerous category of pesticides.

9. Because of EPA's now-longer failure to act on the pending petition, PANNA members and their children are still being exposed to unsafe levels of chlorpyrifos and will continue to be as long as the chlorpyrifos registrations and food tolerances challenged in the petition remain in effect.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct to the best of my knowledge.

Executed this 27 day of August, 2014, at San Francisco, CA.

Margaret Reeves

MARGARET REEVES

ATTACHMENT 1

No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners.

DECLARATION OF MARGARET REEVES, PH.D. IN SUPPORT OF PETITION FOR A WRIT OF MANDAMUS

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Attorneys for Petitioners

I, MARGARET REEVES, declare and state as follows:

1. I am a senior scientist at Pesticide Action Network of North America ("PANNA").

2. I have a Ph.D. in Agricultural Ecology from the University of Michigan (1991), and I spent two years of post-doctoral research in Agronomy at Ohio State University (1991-1993). Before joining PANNA in 1996, I spent most of nine years in Central America, teaching and conducting research in tropical agricultural ecology. I worked with university colleagues and Non-Governmental Organizations to improve productivity of low-input, ecologically sound agricultural methods. I have published articles, in both Spanish and English, in professional and popular/educational journals.

3. My current job responsibilities at PANNA include managing information and outreach regarding acute pesticide poisonings and long-term human health impacts of exposure to pesticides, especially among farmworkers, their families, and community members. I use information collected (1) directly from pesticide-affected communities and (2) from the peer-reviewed literature, to support advocacy campaigns to eliminate the use of the most hazardous pesticides, reduce the use of all synthetic or hazardous pesticides, support the use of least toxic pest management methods, and to advocate for the promulgation and effective enforcement of adequate safety regulations for farmworkers.

4. PANNA is a non-profit advocacy and education organization that was founded in 1982 and is dedicated to preventing harm to the public from pesticides. PANNA works to replace pesticide use with ecologically sound and socially just alternatives. As one of five Pesticide Action Network (PAN) regional centers worldwide, we link local and international consumer, labor, health, environment, and agriculture groups into an international citizens' action network. This network challenges the global proliferation of pesticides, defends basic rights to health and environmental quality, and works to ensure the transition to a just and viable society.

5. PANNA has over 70,000 members nationally. A number of our members have expressed concerns about the serious human health and environmental effects of organophosphate pesticides in general, and chlorpyrifos in particular. We have conducted both air monitoring and biomonitoring for chlorpyrifos in Tulare County, California and found levels of chlorpyrifos that are of serious concern. Our members who participated in those studies are looking to PANNA to help them eliminate this avoidable source of contamination in their communities and in their bodies. They are especially concerned about exposure of their children to pesticides.

6. In addition to working for PANNA, I have been a member of PANNA since I joined PANNA in 1996. I am continuously grateful for the excellent work

that my colleagues do, on behalf of myself and all its members, to rid our collective local, national and international communities of hazardous pesticides, and to support farmers around the globe who are successfully producing healthy food without dependence on hazardous pesticides.

7. PANNA and its members, are very concerned that state and federal regulatory systems for pesticides are failing farmworkers in the United States, including PANNA members. Prominent among them is chlorpyrifos and other organophosphate nerve toxin insecticides. Acute pesticide poisoning refers to adverse health effects associated with exposure to pesticides that occur immediately or shortly following the exposure. They may be of short duration, last days or weeks, and may, in some cases, lead to long-term effects such as chronic neurological problems. Acute effects often lead to temporary job loss and loss of income. Acute effects include irritation of eyes, nose and throat; skin irritation; respiratory difficulty; headache; exhaustion; blurred vision; stomach cramps and vomiting; excessive salivation; tremors, staggering gait and dizziness; numbness; chest tightness; and excessive sweating.

8. The California Department of Pesticide Regulation's (DPR) reported number of acute poisonings is likely a serious underestimate since it is probable that many, if not most, acute agricultural poisonings never get reported. Furthermore, the DPR data show that poisonings frequently occur in the absence of

violations of pesticide use and worker safety laws. In other words, current regulations and laws governing pesticides fail to protect farmworkers from acute poisoning incidents even when they are followed.

9. Of the reported poisonings in California, fifty-one percent from 1998 to 2006 occurred when pesticides drifted from the site of application onto workers. Another 25% resulted from dermal contact with pesticide residues in fields. See Reeves, M., A. Katten and M. Guzmán. 2002. Fields of Poison 2002: California farmworkers and pesticides. Pesticide Action Network, San Francisco, CA. The report is available online at: http://www.panna.org/sites/default/files/FieldsofPoison2002Eng.pdf. Chlorpyrifos was among the top five chemicals in the reported poisonings.

10. These data only address the most serious short-term worker poisoning incidents. There are ample data elsewhere that show that pesticides have long-term, chronic adverse health effects on farmworkers. Those effects include nervous system damage, development problems, hormone disruption, immune system damage, cancer, reproductive effects, and birth defects.

11. Extensive discussion of these issues is provided in a PANNA report that I co-authored. Fields of Poison 2002: California farmworkers and pesticides. That report was made in collaboration with the Californians for Pesticide Reform, United Farm Workers (UFW), and California Legal Rural Assistance Foundation

(CRLAF). It revealed that pesticide safety laws fail to protect many of the state's 700,000 farmworkers from poisonings even when the laws are followed. For that reason, PANNA believes that human pesticide exposures need to be reduced, in some cases, dramatically so.

12. In addition to acute poisoning data, a continuously growing body of data demonstrates that both workers and consumers, including children, are regularly exposed to chlorpyrifos and other organophosphate pesticides. To complement these data, PANNA has conducted numerous field studies in California's Central Valley and elsewhere documenting the presence of chlorpyrifos (exceeding EPA's level of concern) in the air in communities located near citrus orchards where use of the pesticide is common during the summer months. We have also demonstrated the presence of chlorpyrifos in the bodies of people, including pregnant women, in these same communities, in some cases at levels substantially above "average" population levels as determined by the U.S. Centers for Disease Control.

13. PANNA partners and members of the statewide coalition Californians for Pesticide Reform (of which PANNA is an active, founding member) successfully used these data to help win increased county-level protections from pesticide drift by establishing buffer zones for aerial applications of Restricted Use Pesticides (RUP). Unfortunately, California regulations do not recognize

chlorpyrifos as subject to this restriction despite the listing of chlorpyrifos as a RUP by U.S. Environmental Protection Agency.

14. At PANNA, we have been engaged in efforts to ban the use of chlorpyrifos since before the successful ban on domestic use went into effect in 2001. Since then, our efforts have focused on protecting farmworkers, agricultural communities, and consumers from the continued use of chlorpyrifos in agriculture. That work has included: (1) providing detailed technical comments within the process of EPA's re-registration review of chlorpyrifos and of all organophosphate pesticides, (2) detailed analysis of chlorpyrifos body burden data from the Center for Disease Control biannual NHANES study and publication of a report, (3) extensive attempts to attain information about Dow Chemical Company's influence in EPA's chlorpyrifos registration decisions through the Freedom of Information Act, (4) conducting the air monitoring and biomonitoring of chlorpyrifos in CA communities, (5) participation in CA's Proposition 65 hearings urging the Office of Environmental Health Hazard Assessment to list chlorpyrifos as a reproductive or developmental toxicant, and (6) participation as in the 2007 petition to EPA to ban all remaining uses of chlorpyrifos that is the subject of the unreasonable delay case at hand.

15. I am aware that on approximately September 12, 2007, PANNA and the Natural Resources Defense Council submitted a document to EPA entitled

"Petition to Revoke All Tolerances and Cancel All Registrations for the Pesticide Chlorpyrifos" (the "2007 Petition"). I am familiar with the contents of the 2007 Petition, which essentially requested that EPA cancel all registrations and revoke all tolerances for chlorpyrifos. It is my understanding that EPA has not formally responded to that petition.

16. By failing to respond to the 2007 Petition, EPA has harmed PANNA and its members, such as me, by failing to address the human health concerns related to pesticides that are of concern to PANNA and its members. If EPA were to respond to the 2007 Petition, it would help address the concerns of many PANNA members, such as me, by either modifying the use of chlorpyrifos, or allowing PANNA to challenge EPA's decision for failure to do so.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct. Executed this 12th day of April, 2012, at San Francisco, California.

Margaret Reeves

MARGARET REEVES

EXHIBIT 1

October 6, 2011

Pesticide Re-evaluation Division
Office of Pesticide Programs
Environmental Protection Agency
1200 Pennsylvania Ave., NW
Washington, DC 20460-0001

Re: Preliminary Human Health Risk Assessment for Chlorpyrifos Registration Review, Docket ID EPA-HQ-OPP-2008-0850

On behalf of Farmworker Justice, Pesticide Action Network, Pineros y Campesinos Unidos del Noroeste, Sea Mar Community Health Center, and United Farm Workers, we submit the following comments on EPA's preliminary human health risk assessment for chlorpyrifos. We incorporate by reference the comments of the Natural Resources Defense Council, the Farm Worker Pesticide Project, and Physicians for Social Responsibility.

Chlorpyrifos is one of the organophosphate pesticides, which EPA has long recognized "pose the greatest risk to public health." 65 Fed. Reg. 42,021 (Aug. 4, 1997). The preliminary human health risk assessment reviews convincing evidence that chlorpyrifos is even more dangerous than the agency previously believed. The revised drinking water assessment shows that many subpopulations are exposed to unsafe levels of chlorpyrifos oxon in drinking water; new air monitoring studies reveal that chlorpyrifos is present in the air at dangerous levels at many rural locations; for many uses, no amount of protective clothing or engineering controls can adequately protect workers from unsafe exposures; and recent epidemiological studies confirm that *in utero* exposure to chlorpyrifos and organophosphates have harmed the development of children. Taken together, these new lines of evidence directly contradict EPA's prior assertions that aggregate exposures to chlorpyrifos, and cumulative exposures to organophosphates, are safe.

For far too long, chlorpyrifos has poisoned workers, sickened residents, and harmed the development of our children. EPA should, without delay, initiate proceedings to revoke the tolerances and cancel the registrations for chlorpyrifos.

I. EPA Should Not Delay Issuance of a Final, Lawful Reassessment of Tolerances and Re-Registration Review Decision for Chlorpyrifos.

In 1996, Congress passed the Food Quality Protection Act ("FQPA"), which amended the Federal Food, Drug, and Cosmetic Act and the Federal Insecticide, Fungicide, and Rodenticide Act ("FIFRA"). Pub. L. No. 104-170, 110 Stat. 1489 (1996). Under the FQPA, EPA can establish a tolerance only if the agency has determined that "there is a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue." 21 U.S.C. § 346a(b)(2)(A)(ii). To ensure that then-existing pesticides would comply with the new safety standard, Congress instructed EPA to reassess the tolerances and review the registrations for all pesticides. *Id*. § 346a(q)(1); 7 U.S.C. § 136a(g)(1)(A). Congress required EPA to complete all of the tolerance reassessments by 2006. 21 U.S.C. § 346a(q)(1).

EPA reviewed the registrations and reassessed the tolerances for chlorpyrifos in 2006. EPA, Reregistration Eligibility Decision for Chlorpyrifos (2006). EPA's reregistration decision violated the FQPA's requirement to consider aggregate exposure of infants and children to pesticide residues by failing to consider exposure to pesticide drift and volatilization. *See generally* Petition from United Farm Workers et al. to EPA, *Pesticides in the Air -- Kids at Risk: Petition to EPA to Protect Children from Pesticide Drift* (2009). Thus, despite the 2006 deadline, EPA has still not ensured that there is a reasonable certainty that no harm will result from aggregate exposure of infants and children to chlorpyrifos.

Figure 1 shows the percent of the Population Adjusted Dose ("PAD") accounted for by inhalation exposure for rural residents compared to dietary exposure (food and drinking water) using the 2006 PAD of 0.0005 mg/kg-day. These data, most of it collected and publicly available prior to 2006, indicate that exposure through drift and volatilization constitutes a substantially greater fraction of total exposure than dietary exposure.

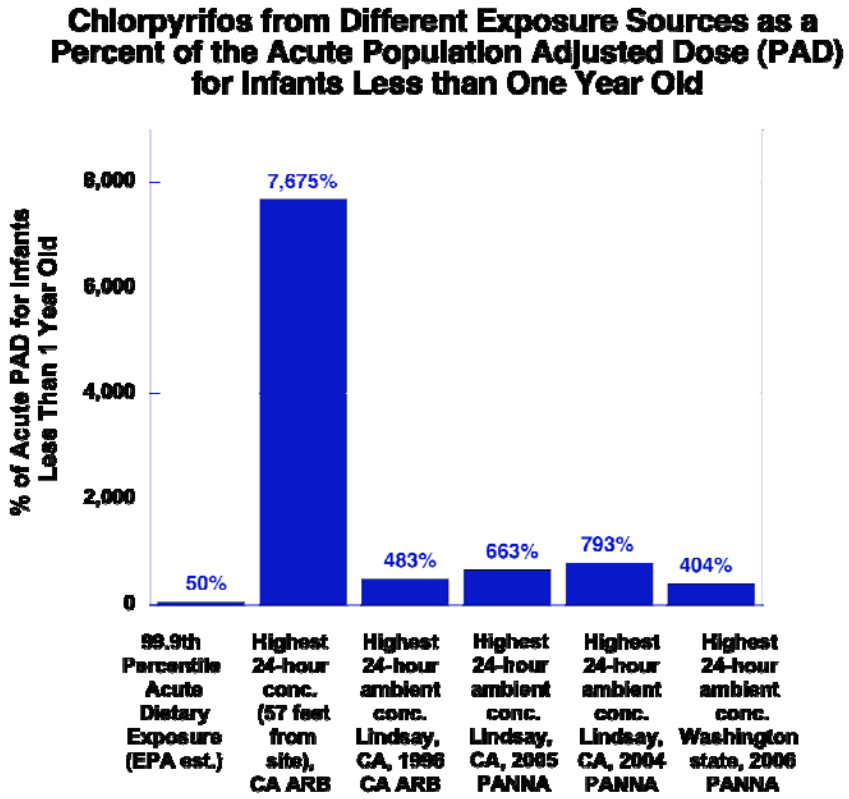

Figure 1: Exposure to chlorpyrifos from the inhalation route is very high for people living in areas of high chlorpyrifos use.

Five years have passed since the FQPA deadline for properly reassessing the tolerances for chlorpyrifos. EPA must avoid any further delay in complying with the law, especially in light of the overwhelming evidence that the current tolerances for chlorpyrifos are not safe.

II. The FQPA Requires EPA to Reassess Tolerances Based on Actual Monitoring Data Showing Unsafe Atmospheric Concentrations of Chlorpyrifos

In recent years, there have been a number of air monitoring studies for chlorpyrifos. E.g., Mills, Katherine et al., *Air Monitoring for Chlorpyrifos in Lindsay, California, June-July 2004 and July-August 2005* (2006); Fenske, Richard et al., *Organophosphorus Pesticide Air Monitoring Project* (2009). The Preliminary Human Health Risk Assessment reviews 15 air monitoring studies for chlorpyrifos in California and Washington. EPA, *Chlorpyrifos: Preliminary Human Health Risk Assessment for Registration Review* at 71 (2011) [hereinafter, *Preliminary Risk Assessment*]. After comparing the data to EPA's Levels of Concern, the preliminary risk assessment concludes that the concentration of chlorpyrifos in many of the air samples[1] exceeds EPA's levels of concern. *Id*.

[1] Four out of twenty-four of the acute ambient air concentrations exceeded the level of concern, three out of five of the acute application site air concentrations exceeded the level of concern, and four out of five of the short and intermediate term application site air concentrations exceeded the level of concern. *Preliminary Risk Assessment* at 71.

The risk assessment points to purported limitations in the air monitoring studies, suggesting that the results might be discounted as a result. For example, the risk assessment notes that individuals do not stay in the same place for 24 hours, and therefore an individual may not be exposed to the concentration of chlorpyrifos measured in a 24-hour sample. *Id*. at 74. It would be unconscionable to discount the air monitoring results on this basis; indeed, they reflect the real world for rural children. Infants and young children, people who work out of the home, and older people with restricted mobility may very well spend 24-hour periods in one location, such as their homes. Most of the studies evaluated by EPA measured pesticide concentrations at residential locations, where these vulnerable people and others like them may indeed be exposed to the 24-hour concentrations measured in the studies.

Moreover, while infants and children may move from their homes to their schools, the air monitoring studies have detected high levels of chlorpyrifos at both schools and private residences. PANNA data from Lindsay, CA air monitoring studies, *Preliminary Risk Assessment* at 73. Based on the air monitoring results, EPA should assume that rural children are in harm's way where they live, go to school, and play. Moreover, it would not be credible to assume that indoor locations are safe given the likelihood that windows will be open during seasons when chlorpyrifos is applied.

In addition, it is worth noting that the HEC process does not necessarily produce reliable 24-hour reference concentrations ("RfCs") because the test animal exposures do not match anticipated human exposures. Most inhalation exposures for laboratory animals are set at a constant concentration for six hours per day, five days per week, providing time for the animals' repair systems to respond to the chemical insult during the "rest" periods (see Figure 2). For both the acute (one-day exposure) and short-term (90-day exposure) chlorpyrifos studies, dosing occurred for six hours per day, five days per week. The rest periods during these studies provide an opportunity for the laboratory animals to replenish depleted cholinesterase and begin repairing damaged tissues.

Figure 2: Exposure pattern for laboratory animals exposed to methyl iodide via inhalation for a typical six hours per day, five days per week study.

Exposure patterns for people living near fumigant application sites are substantially different, with an exposure spike that may cause acute effects during the first day or two after the application, followed by a decreasing concentration over the next week or two (see Figure 3).

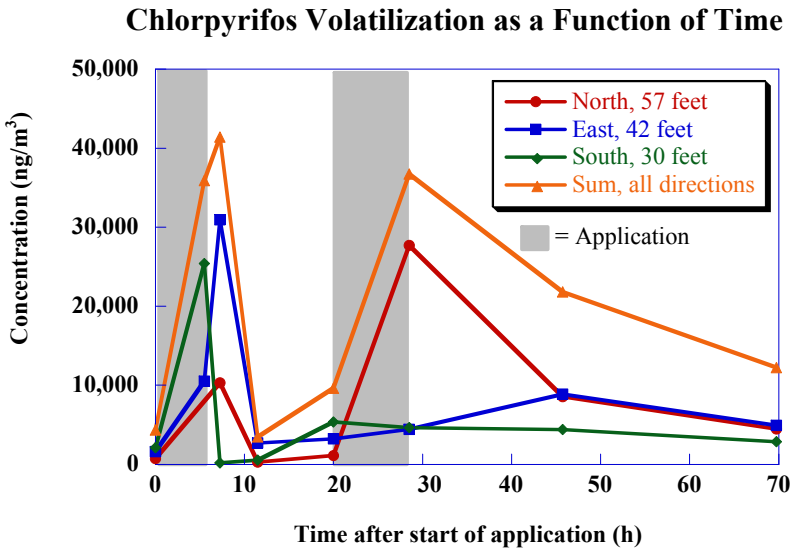

Figure 3: Exposure pattern for an actual application of chlorpyrifos (data from the California Air Resources Board monitoring study[2]) showing a spike in concentration after the application.

Real-world exposure can be continuous (assuming one stays at home and the wind direction is constant), with no opportunity for recovery. The high spike in concentration is likely to have a significantly different toxic effect than the constant exposure experienced by laboratory animals.

Because of the possibility of mixed acute and sub-chronic effects, this failure in inhalation exposure dosing is likely to be one of the most significant flaws in current reference concentration methodology that leads to an underestimation of the actual HEC, especially for toxicity arising from cholinesterase inhibition. Because the time course and duration of animal inhalation studies do not effectively mimic human exposures, the selected endpoints may not be protective of real-world exposures.

The risk assessment also states that data from California and Washington may not be representative of atmospheric concentrations in other areas of the country. Unfortunately, EPA has neither conducted air monitoring itself nor required registrants to conduct such air monitoring for chlorpyrifos, and thus there are no air monitoring data outside of California and Washington. Nonetheless, EPA's obligation under the FQPA is to conduct risk assessments based on "available information," 21 U.S.C. § 346a(b)(2)(C)(i)(I) – (III), and "EPA cannot reject the best available evidence simply because of the possibility of contradiction in the future by evidence unavailable at the time of action -- a possibility that will *always* be present." *Chlorine Chemistry Council v. EPA*, 206 F.3d 1286, 1290-91 (D.C. Cir. 2000). Until EPA has evidence that children's exposure to chlorpyrifos in some parts of the country are lower than the exposures in the California and Washington air monitoring studies, EPA must act based on the data it has. Accordingly, it must use the air monitoring studies to reflect children's exposures and lower tolerances for chlorpyrifos to prevent the unacceptable aggregate exposures that result.

[2] CARB, *Report for the Application and Ambient Air Monitoring of Chlorpyrifos (and the Oxon Analogue) in Tulare County during Spring/Summer 1996,* Test Report #C96-040 and # C96-041 (1998), *available at* http://www.cdpr.ca.gov/docs/empm/pubs/tac/chlrpfs.htm

A. EPA's Consideration of Air Monitoring Data Should Further the Environmental Justice Goals Expressed in Executive Order 12898.

Under the terms of Executive Order 12898, each federal agency must pursue environmental justice "by identifying and addressing, as appropriate, disproportionately high and adverse human health or environmental effects of its programs, policies, and activities on minority populations and low-income populations in the United States." Executive Order 12898, § 1-101 (Feb. 11, 1994). In 2011, federal agencies, including EPA, signed a Memorandum of Understanding to implement the environmental justice goals expressed in Executive Order 12898.[3]

The majority of the air monitoring studies were conducted in Tulare County, California and Yakima County, Washington, and both counties have percentages of people of color and people living below the poverty level that exceed the national average.[4] Air monitoring studies indicate that poor people and/or people of color in rural communities where chlorpyrifos is sprayed are exposed to atmospheric concentrations of chlorpyrifos that exceed the atmospheric concentrations to which non-rural populations are exposed. Some of these concentrations exceed EPA's levels of concern. *Preliminary Risk Assessment* at 72. As a result, the effects of harmful atmospheric concentrations of chlorpyrifos are being borne by rural populations that are predominantly people of color and/or low income. In furtherance of the goals of the recent Memorandum of Understanding and Executive Order 12898, EPA must address these disproportionate health impacts by setting tolerances and imposing registration restrictions such that exposure to chlorpyrifos is limited to levels that are safe for all populations and do not leave people of color and low-income children disproportionately burdened by pesticide pollution.

B. Air Monitoring Studies Show That Some Rural Sites Have Levels of Atmospheric Chlorpyrifos That Are Not Safe.

EPA may establish a residue tolerance only if EPA establishes that a tolerance is safe, and must modify or revoke a tolerance if EPA determines the tolerance is not safe. 21 U.S.C. § 346a(b)(2)(A)(i). A tolerance is safe if EPA has "determined there is a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue, including all anticipated dietary exposures and all other exposures for which there is reliable information." *Id.* § 346a(b)(2)(A)(ii).

As EPA recognizes, non-occupational exposure to atmospheric concentrations of a pesticide must be considered in the aggregate exposure analysis. For chlorpyrifos, there is reliable information, consisting of 15 air monitoring studies, indicating that applications of chlorpyrifos on many crops result in drift and/or field volatilization that create unsafe

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[3] Memorandum of Understanding on Environmental Justice in Executive Order 12898 (2011), *available at* http://epa.gov/environmentaljustice/resources/publications/interagency/ej-mou-2011-08.pdf.

[4] According to the United States Census Bureau, in 2010, the percentage of people who identified as a race other than "White, non-Hispanic" in Yakima County was 61.5% and in Tulare County was 66.3%. These percentages exceed the nationwide percentage of 34.8%. Similarly, 22% and 23% of persons in Yakima and Tulare counties, respectively, were below the poverty level in 2009, compared to 14.3% nationwide.

atmospheric concentrations of chlorpyrifos. Therefore, EPA must modify chlorpyrifos use patterns or revoke residue tolerances for chlorpyrifos to reduce exposures to, or below, acceptable levels.

III. EPA Should Retain the 10X FQPA Safety Factor for Infants and Children in Light of Uncertainty Regarding the Effects of Chlorpyrifos on Endocrine Systems and Neurological Development.

The FQPA specifies that in the case of threshold effects, an additional tenfold margin of safety for the residue and other sources of exposure shall be applied for infants and children. 21 U.S.C. § 346a(b)(2)(C)(ii)(I). EPA can apply a different margin only if "on the basis of reliable data, such margin will be safe for infants and children." *Id*.

Congress intended "that EPA interpret the language of this section in furtherance of the . . . recommendations of the National Research Council's Study, Pesticides in the Diets of Infants and Children." H.R. Rep. No. 104-669 at 43 (1996). The National Research Council study recommended that EPA apply a tenfold uncertainty factor "when there is evidence of postnatal developmental toxicity and when data from toxicity testing relative to children are incomplete." *Id*.

Since the purpose of the FQPA safety factor is to account for uncertainty regarding the special vulnerability of infants and children to pesticides, Congress specified that EPA could apply a lower uncertainty factor only if EPA has reliable data showing that the alternative margin is safe. With respect to endocrine effects, EPA lacks reliable data that a 1X safety factor is safe for infants and children.

EPA recently placed chlorpyrifos on the first list of chemicals to undergo tier 1 screening in the endocrine disruptor screening program, and issued test orders requiring such screening. 74 Fed. Reg. 17,579 (Apr. 15, 2009). Under EPA's guidelines for the endocrine disruptor screening program, a chemical undergoes tier 1 screening only if there is uncertainty as to whether the chemical is capable of disrupting the endocrine system; if there is already data on this issue, then a chemical proceeds directly to tier 2 testing or to hazard assessment. 63 Fed. Reg. 71,542 (Dec. 28, 1998).

By issuing tier 1 screening orders for chlorpyrifos, EPA has acknowledged that the agency does not have adequate data to satisfy the tier 1 screening requirements, and that there is uncertainty regarding the endocrine disruption effects of chlorpyrifos. As a result, a 1X safety factor would not be based on reliable data indicating that the margin is safe for infants and children. If EPA lacks reliable data regarding the effects of chlorpyrifos on the endocrine systems of infants and children, then EPA is precluded from deviating from the 10X FQPA safety factor.

In addition, both the toxicity data and the epidemiological data indicate that the effects of chlorpyrifos on neurodevelopment both prenatally and in infant and juvenile animals are substantially greater than in adults. We refer EPA to the NRDC comment letter for a detailed analysis of these concerns, and note that in the absence of a no observed adverse effect level

("NOAEL"), the developmental neurotoxicity study provides no assurance that children will be protected if the FQPA 10X factor is not retained.

IV. In Its Final Tolerance and Registration Decisions, EPA Must Consider Data Showing That Cumulative Exposures to Chlorpyrifos and Other Organophosphates Are Not Safe.

Two subsections of the FQPA require EPA to consider cumulative effects when establishing a tolerance. In establishing, modifying, leaving in effect, or revoking a tolerance, EPA must assess the risk of a pesticide chemical based on "available information concerning the cumulative effects on infants and children of such residues and other substances that have a common mechanism of toxicity." 21 U.S.C. § 346a(b)(2)(C)(i)(III). Similarly, for populations other than infants and children, EPA must consider "available information concerning the cumulative effects of such residues and other substances that have a common mechanism of toxicity." *Id*. § 346a(b)(2)(D)(v).

Organophosphates were the first chemicals EPA identified as having a common mechanism of toxicity, based on their "ability to bind to and phosphorylate the enzyme acetylcholinesterase in both the central (brain) and peripheral nervous systems." EPA, *Organophosphorus Cumulative Risk Assessment -- 2006 Update* at 3 (2006) [hereinafter *Cumulative Risk Assessment*]. EPA interprets the FQPA to require the agency to find that the cumulative effects of exposures to organophosphates from all pathways are safe. *Id*. at 15.

A. Recent Epidemiology Studies Confirm Earlier Studies Indicating That Cumulative Exposures to Organophosphates are Associated with Neurodevelopmental Deficits.

Since the 2006 Cumulative Risk Assessment for organophosphates and associated re-registration determinations, at least three major epidemiology studies on chlorpyrifos and/or organophosphates have been published. The Columbia University studies have found an association between levels of chlorpyrifos in umbilical cord blood and negative neurological and behavioral outcomes in children at 3 and 7 years of age.[5] Statistical analyses confirm that the negative effects of chlorpyrifos are statistically significant and persist after controlling for other chemical exposures.

Two other epidemiology studies, conducted by researchers at the Mt. Sinai School of Medicine and the University of California at Berkeley, found that increased levels of urinary organophosphate metabolites are associated with certain negative neurodevelopment outcomes in children.[6] Unlike the Columbia study, the Mt. Sinai and UC Berkeley studies did not attempt to

[5] Rauh, V., et al., *Impact of Prenatal Chlorpyrifos Exposure on Neurodevelopment in the First Three Years of Life among Inner-City Children*, 118 Pediatrics 6 (2006); Rauh, V. et al., *Seven-Year Neurodevelopmental Scores and Prenatal Exposure To Chlorpyrifos, a Common Agricultural Pesticide*, Environmental Health Perspectives 119 (8): 1196-01 (2011).

[6] Engel, S. et al., *Prenatal Exposure to Organophosphates, Paraoxonase 1, and Cognitive Development in Childhood*, Environmental Health Perspectives 119 (8): 1182-88 (2011) ("We found that prenatal maternal urinary

correlate outcomes with exposure to chlorpyrifos alone but instead correlated outcomes with exposure to organophosphate pesticides. EPA has stated that because the Mt. Sinai and UC Berkeley studies did not specifically measure exposure to chlorpyrifos, they are of limited use in the risk assessment for chlorpyrifos. *Preliminary Risk Assessment* at 31-32.

While the UC Berkeley and Mt. Sinai studies may not attribute the observed outcomes solely to exposure to chlorpyrifos, that does not mean the studies can be cast aside. Under the FQPA, EPA's risk assessment cannot be limited to aggregate exposures to chlorpyrifos. Instead, EPA must consider as well the cumulative effects of exposure to chlorpyrifos and other chemicals with a common mechanism of toxicity. 21 U.S.C. § 346a(b)(2)(C)(i)(III), 346a(b)(2)(D)(v). The UC Berkeley and Mt. Sinai studies are credible evidence that must be used in assessing the cumulative risk from organophosphates.

The UC Berkeley and Mt. Sinai studies indicate that from 1997 through 2001, children developing in the womb were exposed to actual levels of organophosphates that resulted in later developmental and behavioral harm. FIFRA Scientific Advisory Panel, *Meeting Minutes of the FIFRA Scientific Advisory Panel Meeting Held September 16-18, 2008 on the Agency's Evaluation of the Toxicity Profile of Chlorpyrifos* at 37-38 (2008) [hereinafter *SAP Report*]. These studies show that, at a minimum, for the years 1997 through 2001, cumulative exposures to organophosphates were not safe. EPA's interpretation of these epidemiological studies must conform to the FQPA's mandate that EPA assess not just aggregate exposure to chlorpyrifos but cumulative exposures to organophosphates.

B. EPA's Cumulative Effects Analysis Should Account for Additive or Interactive Effects between Organophosphate Pesticides.

At the September 2008 meeting of the FIFRA Scientific Advisory Panel, the SAP suggested, after reviewing recent epidemiology studies on chlorpyrifos and other organophosphates, that the agency consider potential additive and synergistic effects of chlorpyrifos and other organophosphates. The SAP "supported the statement that exposures to all three AChE -inhibiting insecticides may act in combination to produce the observed effects. The Panel agreed that there may, in fact, be additive effects or effects generated by a mixture of the agents." *SAP Report* at 43; *see also id*. at 13.

In interpreting the Columbia studies, the SAP noted that diazinon, an organophosphate, and propoxur, a carbamate, were present along with chlorpyrifos. If the data are used to show the combined effect of diazinon and chlorpyrifos, "there is a slightly greater reduction in birth weight" than the effects of chlorpyrifos alone. "This may indicate that the effect of the combined chemicals is slightly greater than the individual chemicals alone and that there could

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dialkylphosphate metabolite concentrations were negatively associated with aspects of neurodevelopment at 12 and 24 months, and also at 6-9 years of age, in an urban, inner-city population."); Eskenazi, B. et al., *Organophosphate Pesticide Exposure and Neurodevelopment in Young Mexican-American Children*, Environmental Health Perspectives 115 (5): 792-98 (2007) ("[W]e report an adverse association of prenatal organophosphate pesticide exposure as measured by DAPs with mental development and pervasive developmental problems at 24 months of age."); Bouchard, M. et al., *Prenatal Exposure to Organophosphate Pesticides and IQ in 7-Year-Old Children*, Environmental Health Perspectives 119 (8): 1189-95 (2011).

be potential interaction between the two chemicals with respect to the association." *Id*. at 41. Indeed, the SAP notes that Rauh found that "the combination of chlorpyrifos and diazinon produced slightly greater effects for MDI than were seen for chlorpyrifos alone." *Id*. at 42.

The available evidence, including epidemiological studies and the recommendations of the SAP, suggest that there may be additive and/or synergistic effects from exposure to chlorpyrifos and other organophosphates. The agency should consider these potential additive and/or synergistic effects in assessing cumulative effects.

C. The Preliminary Risk Assessment Undermines Key Conclusions in the 2006 Organophosphate Cumulative Risk Assessment.

EPA completed the most recent cumulative risk assessment for organophosphates in 2006. If EPA relies on the 2006 Cumulative Risk Assessment in its forthcoming final decision on chlorpyrifos tolerances and registrations, EPA must account for more recent analyses that undermine key conclusions in the 2006 Cumulative Risk Assessment.

For example, in the preliminary risk assessment, EPA calculates that several subpopulations -- especially infants -- are exposed to levels of chlorpyrifos in drinking water that exceed levels of concern *Preliminary Risk Assessment* at 61. This directly contradicts the 2006 Cumulative Risk Assessment, which found that, individually and cumulatively, the levels of organophosphates in drinking water were safe. *Cumulative Risk Assessment* at 15 ("[T]he results of the OP CRA [cumulative risk assessment] support a reasonable certainty of no harm finding as required by FQPA and therefore EPA has completed reassessment of the OP tolerances."). EPA's Cumulative Risk Assessment conclusion is no longer tenable, in light of the preliminary risk assessment's calculation that levels of chlorpyrifos in drinking water are not safe.

Second, in the 2006 Cumulative Risk Assessment, EPA did not consider bystander exposures to chlorpyrifos. Recent air monitoring studies reveal harmful levels of chlorpyrifos in the air at many rural sites. Given that the air monitoring data show that some rural subpopulations are being exposed to harmful levels of chlorpyrifos through drift and field volatilization, the air monitoring data call into question the overall conclusion that cumulative exposures to organophosphates are safe.

A number of other currently registered organophosphate pesticides are also subject to spray drift and/or field volatilization. The California Air Resources Board has acquired air monitoring data for acephate, azinphos-methyl, DEF, diazinon, ethoprop, malathion, methamidophos, methidathion, methyl parathion, naled and phorate.[7] In all cases, measurable levels of the pesticide were found in air near application sites and in ambient air in areas of high use. EPA should account for these exposures when evaluating the filling of the "risk cup" and the cumulative risks associated with use of organophosphates.

[7] CARB, Toxic Air Contaminant Program Monitoring Reports (2011), *available at* http://www.cdpr.ca.gov/docs/emon/pubs/tac/tacstdys.htm.

The 2006 Cumulative Risk Assessment for organophosphates fails to account for unsafe drinking water and air exposures. EPA's revised drinking water assessment for chlorpyrifos, and recent data on atmospheric concentrations of chlorpyrifos, contradict EPA's 2006 conclusion that cumulative exposures to organophosphates are safe. Moreover, EPA has announced no plans to undertake a new cumulative risk assessment for organophosphates. At a minimum, to meet the FQPA safety standard, EPA should revoke all tolerances for chlorpyrifos to eliminate the unacceptable risks posed by chlorpyrifos specifically and to reduce the overall exposure of infants and children to organophosphates.

D. The Preliminary Human Health Risk Assessment Supports Canceling the Uses of Chlorpyrifos That Result in Unreasonable Risks to Farmworkers.

The preliminary human health risk assessment analyzes 305 occupational exposure scenarios. Eighty scenarios "resulted in risk estimates of concern . . . at all levels of personal protection and engineering controls considered." *Preliminary Risk Assessment* at 14. EPA should initiate proceedings to cancel the uses of chlorpyrifos that fit these 80 scenarios, unless EPA can substantiate formal findings that these occupational risks are not unreasonable adverse effects. To do so, EPA would have to find that the benefits of such uses outweigh the risks such that they are not unreasonable adverse effects within the meaning of FIFRA.

For other occupational exposure scenarios, EPA relies on personal protective equipment to reduce exposure below the level of concern. Personal protective equipment is often ineffective, for a variety of reasons, ranging from faulty equipment, to the failure of employers to provide equipment, to weather conditions that make wearing protective equipment not feasible. *See, e.g.*, Washington State Pesticide Incident Reporting and Tracking Review Panel, *2009 Annual Report* at 61-64 (2009). Risk estimates based on the use of protective equipment should incorporate real-world data on the adequacy of such equipment, or, in the absence of such data, should rely on realistic assumptions about the effectiveness of such equipment in reducing exposures.

Sincerely,

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No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

DECLARATION OF SATTIE CLARK IN SUPPORT OF SECOND PETITION FOR A WRIT OF MANDAMUS

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Attorneys for Petitioners Pesticide Action Network North America and Natural Resources Defense Council, Inc.

DECLARATION OF SATTIE CLARK

I, SATTIE CLARK, declare and state as follows:

1. I have been a member of the Natural Resources Defense Council (NRDC) since 2002, and I support NRDC's efforts to protect the public from exposure to harmful pesticides.

2. I am 49 years old and live in Redwood Valley, California with my family.

3. I have a son who is nine years old.

4. I am aware that organophosphate pesticides including chlorpyrifos are widely used in agricultural areas and pose significant risks to human health. I am concerned about the health impacts directly attributable to chlorpyrifos, as well as the aggregate impacts of exposure to chlorpyrifos and chemicals like it along with other hazardous environmental contaminants.

5. I am very concerned about my son's potential exposure to chlorpyrifos and chemicals like it, and the immediate and long-term consequences to his health. He has celiac disease and suffers from poor digestion and chronic stomach inflammation. Accordingly, both my son and I are particularly vulnerable to the effects of toxins because our immune and detoxification systems are compromised. We have been diagnosed with a genetic mutation of methylenetetrahydrofolate reductase (MTHFR), which prevents our bodies from being able to properly break down toxins. I am concerned that exposure to chlorpyrifos and chemicals like it could make us highly susceptible to cancer and other chronic diseases due to our bodies' vulnerability to toxic buildup. I believe it is important that the Environmental Protection Agency (EPA) consider the impacts of pesticides on vulnerable populations, including children and those with chronic illnesses, in its evaluation of their safety.

6. I am worried about the chronic effects of long-term exposure to chlorpyrifos and pesticides like it. I spent several years of my childhood in Sutter, California surrounded by nut

and fruit orchards that were frequently sprayed for pests. I am concerned that exposure to pesticides like chlorpyrifos during that time has had lasting impacts on my health.

7. Between 2001 and 2006, I resided in Corbett, Oregon on property downwind from a working raspberry farm. I am concerned that exposure to chlorpyrifos and pesticides like it during that time caused long-term damage to my health. I am also concerned that, because I lived in Corbett during my pregnancy and after the birth of my son, my exposure to these chemicals has adversely affected his health and development.

8. The decision to move my family and business to Redwood Valley, California in March of 2013 was primarily informed by my concern for exposure to chemicals like chlorpyrifos. We decided to move to Mendocino County due to our belief that its pesticide regulations for vineyards are more stringent than that of its neighboring counties, such as Sonoma or Napa. Because of my concern that exposure to pesticides like chlorpyrifos might worsen the health conditions that my family and I suffer from, we were not able to pursue purchasing properties in other parts of California that we would have otherwise considered.

9. I am particularly worried about the neurotransmitter impacts of chlorpyrifos exposure. Both my son and I require medical treatment for neurotransmitter support, and I am concerned that our exposure to chlorpyrifos and chemicals like it contributed to our respective neurotransmitter deficiencies.

10. I am also concerned about the endocrine disrupting impacts of chlorpyrifos and chemicals like it. Following the birth of my son, I was diagnosed with post-partum hypothyroidism, and more recently I have experienced pituitary dysfunction. I am concerned that exposure to chlorpyrifos and chemicals like it contributed to these issues, and that ongoing exposure could cause more severe endocrine effects.

11. Members of my family swim in fresh water near agricultural regions in California and Oregon. My sister frequently swims in fresh water in the Sacramento Valley, and she has experienced impaired neurotransmitter function. I am concerned that exposure to agricultural runoff containing pesticides like chlorpyrifos has contributed to her condition.

12. I am concerned about pesticide residues, including chlorpyrifos, on the produce that my family consumes. Because of this concern, I purchase organic produce at a substantially higher cost than that of conventional produce. I believe that purchasing organic produce is necessary to protect my son from the adverse health impacts associated with chlorpyrifos and pesticides like it.

13. Although I have not used chlorpyrifos or chemicals like it around my home, and try to avoid exposure by consuming only organic produce and meat, I am alarmed that my family may be exposed to this pesticide through pathways that I cannot control.

14. I am aware that in 2007 NRDC and the Pesticide Action Network of North America submitted to EPA a petition to revoke all tolerances and cancel all registrations for chlorpyrifos. I understand that EPA has not formally responded to that petition.

15. In 2012, I submitted a declaration in an earlier lawsuit in an attempt to compel EPA to respond to the 2007 Petition. That earlier lawsuit was *In re Pesticide Action Network North America and Natural Resources Defense Council*, No. 12-71125, and I have attached my declaration in that case to this declaration as Attachment 1. I have reviewed my previous statements to the Court in that matter and reaffirm them here. All information in my prior declaration remains true and accurate with the updates provided in this declaration. My prior statements apply with equal force to this renewed petition for writ of mandamus.

16. EPA's failure to respond to NRDC's petition has harmed NRDC and its members, such as myself, by leaving unaddressed the adverse health impacts associated with chlorpyrifos. If the court compels EPA to respond to the petition, it will address my concerns, and those of other NRDC members, by restricting the use of chlorpyrifos or allowing NRDC to challenge EPA's decision to maintain current tolerances and registrations for the pesticide.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct, to the best of my knowledge, information, and belief.

Executed this 13th day of August, 2014 at 4:22 pm.

SATTIE CLARK

No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
NATURAL RESOURCES DEFENSE COUNCIL, INC.,

Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

DECLARATION OF SHARON BOLTON IN SUPPORT OF SECOND PETITION FOR A WRIT OF MANDAMUS

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Attorneys for Petitioners Pesticide Action Network North America and Natural Resources Defense Council, Inc.

DECLARATION OF SHARON BOLTON

I, SHARON BOLTON, declare and state as follows:

1. I have been a member of the Natural Resources Defense Council (NRDC) since 2009, and I support NRDC's efforts to compel the Environmental Protection Agency (EPA) to protect the public from hazardous chemicals.

2. I am 63 years old and I live in Tyler, Texas.

3. I have three children and three young grandchildren, all of whom reside in Tyler.

4. I am aware that organophosphate pesticides including chlorpyrifos are commonly applied to a variety of crops in the United States and represent a hazard to human health. I initially learned about the risks of pesticide exposure over forty years ago, and I am deeply concerned about the consequences of chlorpyrifos use to public health and the environment.

5. I am concerned about my family's exposure to chlorpyrifos and chemicals like it. I live near peach orchards, and I am worried that I may be exposed to chlorpyrifos and other organophosphate pesticides that are applied to those orchards. My children and grandchildren also live in this area, and I worry that exposure to these chemicals will adversely affect their health.

6. I am worried about chronic effects of long-term exposure to chlorpyrifos and pesticides like it. I was raised in Muncie, Indiana, and resided there until I was thirty years old. During that time, I lived in an agricultural area on property neighboring working farms. I am concerned that exposure to chlorpyrifos and pesticides like it during that time caused damage to my health.

7. I am concerned about the impacts of chlorpyrifos and chemicals like it on my children. I lived in Indiana during my pregnancies and after the birth of my children, and I worry

that my exposure to these chemicals adversely affected their health. In addition, my children played outdoors on my property in Indiana when they were young. I regularly observed aerial applications of chemicals to the corn fields across the street from my home, which sometimes took place while my children were playing outdoors. Since then, I have been concerned about exposure resulting from drift and its long-term effect on my children.

8. I have three young grandchildren, a 10-month-old, a five-year-old and a seven-year-old. They frequently play outdoors, and I am concerned that exposure to chlorpyrifos and chemicals like it may adversely affect their health and development.

9. I am particularly worried about the hormone disrupting effects of chlorpyrifos. My niece resides in Elwood, Indiana, within twenty miles of a number of working farms. She experienced a hormone abnormality that required consistent medical treatment for the entirety of her childhood. I am concerned that exposure to chlorpyrifos and pesticides like it interfered with her hormone functions.

10. I am concerned about pesticide residues, including chlorpyrifos, on the fruit and vegetables that I consume. Because of this concern, I purchase organic produce whenever possible.

11. I drink tap water and believe that it should be free of hazardous concentrations of chlorpyrifos and other toxic chemicals.

12. Although I do not use chlorpyrifos or chemicals like it at home, I am concerned about exposure that is not within my control.

13. I am aware that in 2007 NRDC and the Pesticide Action Network of North America petitioned EPA to revoke all tolerances and cancel all registrations for chlorpyrifos. I understand that EPA has not formally responded to that petition.

14. In 2012, I submitted a declaration in an earlier lawsuit in an attempt to compel EPA to respond to the 2007 Petition. That earlier lawsuit was *In re Pesticide Action Network North America and Natural Resources Defense Council*, No. 12-71125, and I have attached my declaration in that case to this declaration as Attachment 1. I have reviewed my previous statements to the Court in that matter and reaffirm them here. All information in my prior declaration remains true and accurate with the updates provided in this declaration. My prior statements apply with equal force to this renewed petition for writ of mandamus.

15. EPA's failure to address NRDC's petition has harmed NRDC and its members, such as myself, by permitting ongoing harm to public health. If the court compels EPA to respond to the petition, it will address my concerns, and those of other NRDC members, by restricting the use of chlorpyrifos or allowing NRDC to challenge EPA's decision to maintain current tolerances and registrations for the pesticide.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct, to the best of my knowledge, information, and belief.

Executed this 13 day of August, 2014 at 8:00 am.

Sharon Bolton

SHARON BOLTON

No. _____

UNITED STATES COURT OF APPEALS
FOR THE NINTH CIRCUIT

IN RE PESTICIDE ACTION NETWORK NORTH AMERICA
AND
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Petitioners,

v.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY,

Respondent.

DECLARATION OF GINA TRUJILLO IN SUPPORT OF SECOND PETITION FOR A WRIT OF MANDAMUS

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Attorneys for Petitioners Pesticide Action Network North America and Natural Resources Defense Council, Inc.

DECLARATION OF GINA TRUJILLO

I, Gina Trujillo, declare as follows:

1. I am the Director of Membership Development and Membership Services at the Natural Resources Defense Council, Inc. ("NRDC"). I have held this position for nine years. Before that, I was the Associate Director of Membership. I have worked in NRDC's Membership Department for twenty-one years.

2. My duties include supervising the preparation of materials that NRDC distributes to members and prospective members. Those materials describe NRDC and identify its mission.

3. NRDC is a membership organization incorporated under the laws of the State of New York. It is recognized as a not-for-profit corporation under section 501(c)(3) of the United States Internal Revenue Code.

4. NRDC currently has approximately 300,461 members. There are NRDC members residing in each of the fifty United States and in the District of Columbia.

5. NRDC's mission statement declares that "The Natural Resources Defense Council's purpose is to safeguard the Earth: its people, its plants and animals, and the natural systems on which all life depends." Among NRDC's six priorities is "stemming the tide of toxic chemicals."

6. Protecting the public from the substantial adverse health effects caused by exposure to toxic chemicals, including chlorpyrifos, is central to NRDC's purpose.

Pursuant to 28 U.S.C. § 1746, I declare under penalty of perjury that the foregoing is true and correct, to the best of my knowledge, information, and belief.

Executed this 6th day of August, 2014, at New York, NY.

GINA TRUJILLO