# DISPOSITION OF PEER REVIEW COMMENTS FOR TOXICOLOGICAL PROFILE FOR DDT, DDE, and DDD

U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
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Agency for Toxic Substances and Disease Registry

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Peer reviewers for the third pre-public comment draft of the Toxicological Profile for DDT, DDE, and DDD were:

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## Comments provided by Peer Reviewer #1

## General Comments

**COMMENT:** I have always had a problem in all of the Tox Profiles where data is characterized as being "inconsistent" when not every publication quoted shows exactly the same effect. To my mind "inconsistent" means that some studies with any particular outcome show and increase, other show a decrease and still others show no effect. However the way it is used throughout is that is some studies show an increase but others show no effect, the result is labelled "inconsistent" with the implication that there is no effect of concern. This often is not the case. There are many reasons why a study would show no effect, such as the analytical method was not sufficiently sensitive, or the study population was too small. There is no attempt to apply the kinds of considerations that would constitute a meta-analysis, where studies are rated by quality and size. I realize that this has been a standard way in which the Tox Profiles have been done for years, and you probably don't want to change it now, but I urge you at least to explain that "inconsistent" in the ATSDR terminology does not mean there is no association. I have added some suggested language in the first chapter.

***RESPONSE:*** *As suggested in the Reviewer's annotated comments, the following discussion was added to Section 1.2:*

> Although the epidemiological studies provided consistent evidence of associations for some health effects, these studies are observational and do not establish causality; the observed statistical association may be due to effects of other factors, such as exposure to other pollutants. Likewise, the absence of an association does not necessarily imply the absence of causal relationship.

**COMMENT:** There is a major problem with analysis of DDT compounds, in that they migrate with other chlorinated pesticides, PCBs, dioxin/furans and other highly lipophilic chemicals. Often the health outcomes observed may show statistical significance with DDT concentration but in fact are a consequence of a different chemical that co-migrates with DDT. Several studies are already referenced in the document that show this being the case. But it needs to be clearly stated in Chapter 1 that "consistent" does not necessarily mean that the health outcome is caused by DDT/DDE. I have also added suggested language on this point in Chapter 1. This is the reserve side of "inconsistent", in that "consistent" should not be implied to mean "caused by" without adjustment for other lipophilic chemicals.

***RESPONSE:*** *The following discussion was added to Section 1.2:*

> Another limitation of the epidemiological database is that most studies lacked statistical control for exposure to other compounds, particularly highly lipophilic compounds such as polychlorinated biphenyls (PCBs), chlorodibenzo-*p*-dioxins (CDDs), and chlorodibenzofurans (CDFs), which may co-migrate with DDT and could be the causative agent.

**COMMENT:** In my judgment it is a mistake to dismiss inhalation as a route of exposure to DDT compounds. Grated there has been little study of inhalation as a route of exposure, in spite of a recent publication showing it to be important with indoor spraying of DDT. But there is increasingly strong evidence that inhalation is a major route of exposure for PCBs. It is true that there is very limited evidence that inhalation is a major route of exposure to DDT, but the air concentrations reported in section 5.5.1 are enormous – much greater than those documented to result in significant exposure to PCBs. I've added language that indicates that more research is needed here, and have recommended deletion of dogmatic statements that inhalation is totally unimportant. I realize that you may not want to

add PCB references to the DDT document, but I do think the principal is important and the PCB results are the best evidence that inhalation of lipophilic chemicals can be a significant route of exposure.

***RESPONSE:*** *The text in Sections 1.1 and 5.6 has been revised to eliminate the statement that intake via the inhalation route is negligible.*

*Section 1.1:* Although DDT and its metabolites are ubiquitous in the atmosphere, they have not been shown significantly to contribute to body burden; however, this has not been well studied.

*Section 5.6:* Although the contribution of inhaled DDT to the overall body burden is expected to be small, this has not been adequately investigated.

**COMMENT:** With regard to the specific questions regarding Chapter 1, the three points above are my major concerns in this chapter and throughout. I believe the animal data is extremely important, because only in animals can one control exposure only to DDT compounds. The animal result support the MRL values. I do agree that we do not have adequate inhalation data to derive MRLs for DDT, but given the air concentration reported in Chapter 5 inhalation should not be ignored.

***RESPONSE****: The statements in Intermediate-Duration MRL and Chronic-Duration MRL discussions in Section 6.2 were revised to indicate that studies evaluating inhalation toxicity are needed. Additionally, a data need was added to the discussion of Health Effects.*

*Intermediate MRL:* Additional inhalation data are needed to identify critical targets of toxicity and evaluate concentration-response relationships.

*Chronic MRL:* No chronic-duration inhalation toxicity studies in animals that could be used to establish concentration-response relationships were located; these data are needed for derivation of a chronic-duration inhalation MRL.

*Health Effects:* Air monitoring data suggest that DDT and its metabolites are still present decades after its use was banned. Health effect studies are needed to evaluate possible health effects associated with long-term exposure to low concentrations of DDT in air.

**COMMENT:** With regard to Chapter 2, most of my comments have been incorporated into the text. The issue of mechanisms of action is disappointing in that there really isn't much study in recent years. For other POPs there is a lot of study of gene induction and ROS generation, but there seems to be little with DDT compounds. Copies of references to be added are attached. However I have not added to any of the tables, and if the references are suggest are to be used some will need to be listed in the tables.

***RESPONSE:*** *See Responses to Annotated Comments.*

**COMMENT:** I don't really have more specific concerns about the substance of the other chapters, other than coming back to the same issues discussed above, which I have inserted at a number of places. The section on interactions with other chemicals in Chapter 3 is important and is well written, but there appears not to be a lot of information. I like the data needs identified in Chapter 6, but don't have much to add to that.

***RESPONSE:*** *No response needed.*

## Annotated Comments

The Reviewer suggested a number of editorial revisions, most of the suggested revisions were made to the profile. Some stylistic changes that were purely arbitrary were not incorporated. Responses to Reviewer comments that were not considered editorial or stylistic are presented below.

**COMMENT:** Regarding the following sentence in Section 1.1— Although DDT and its metabolites are ubiquitous in the atmosphere, they are present in such low concentrations that exposures through inhalation or dermal contact are considered to be negligible—the Reviewer comments "This is too strong a statement as elaborated in the health effects section."

***RESPONSE:*** *The Reviewer's suggested revision*—Although DDT and its metabolites are ubiquitous in the atmosphere, they have not been demonstrated to significantly contribute to body burden—*was made.*

**COMMENT:** The Reviewer suggested the following addition to Section 1.1:

*Huang et al. (2006) reported levels of total DDT-related compounds in farmed and wild salmon from around the world. Farmed salmon had concentrations as high as 28 ng/g wet weight, and total DDT was at detectable levels even in every wild Pacific salmon tested, but at lower concentrations.*

***RESPONSE:*** *The text in Section 1.1 discussing levels of DDT and metabolites in foodstuff was revised to include the Huang et al. (2006) reference:*

*p,p'*-DDE was the most frequently detected isomer in studies of U.S. food samples (FDA 2006; USDA 2016). Some of the foods with detectable levels of DDT and metabolites include American cheese, butter, catfish, carrots, summer squash, celery, and salmon (FDA 2006; Huang et al. 2006; USDA 2016).

*Additional information from the Huang et al. (2006) was added to Section 5.5.4:*

A comparison of ΣDDT concentrations between farmed raised salmon (from eight regions in Europe, North America, and South America) and wild Pacific salmon found significantly higher levels of total DDT in farm-raised salmon versus wild salmon (Huang et al. 2006). A comparison of total DDT levels across regions demonstrated significantly higher levels in Europe compared to North America and in North America when compared to South America.

**COMMENT:** The Reviewer suggested the addition of the following text to Section 1.2:

*It must be emphasized that "inconsistent evidence" does not mean "no effect". There are many reasons why a study may not show an effect, such as the analytical methods were not sensitive enough or the sample size was too small. In some cases there is true inconsistency, where some publications report increases in the outcome with DDT levels, whereas other report decreases. But in the case where some studies report increases in a specific outcome but other find no effect there is the possibility that there is a true association.*

*There is another major problem which complicates conclusions regarding causation of diseases by DDT and related compounds. DDT is monitored in adipose tissue or serum samples, but there are many other lipophilic chemicals that are also present in human fat. Furthermore it is common that samples elevated in one lipophilic chemical are also elevated in several others. Often these other chemicals are not measured, and even if measured there is no attempt to determine whether the*

*health outcome is due to DDT or rather to other chlorinated pesticides, dioxins/furans, polychlorinated biphenyls, brominated flame retardants or other persistent, lipophilic chemicals. Therefore even in the case of those health outcomes listed above where there are consistent associations with serum or adipose DDT, one must acknowledge that this does not prove that DDT caused the disease. This is why controlled exposure studies in animals are so important when considering causation.*

***RESPONSE:*** *The following discussion of causality was added to Section 1.2:*

Although the epidemiological studies provided consistent evidence of associations for some health effects, these studies are observational and do not establish causality; the observed statistical association may be due to effects of other factors, such as exposure to other pollutants. Likewise, the absence of an association does not necessarily imply the absence of a causal relationship. The available epidemiological studies do not provide sufficient data to describe exposure-response relationships for potential health outcomes associated with exposure to DDT, DDD, or DDE. Another limitation of the epidemiological database is that most studies lacked statistical control for exposure to other compounds, particularly highly lipophilic compounds such as polychlorinated biphenyls (PCBs), chlorodibenzo-*p*-dioxins (CDDs), and chlorodibenzofurans (CDFs), which may co-migrate with DDT and could be the causative agent.

**COMMENT:** The Reviewer suggested adding Cohn et al. 2003 to the string reference of studies examining time to pregnancy.

***RESPONSE:*** *The following revision was made to the discussion of Developmental and Reproductive Effects in Section 2.1:*

…(*time to pregnancy* [Axmon et al. 2006; Buck Louis et al. 2013, 2014; Cohn et al. 2003; Chevrier et al. 2013; Harley et al. 2008; Law et al. 2005]…

**COMMENT:** The Reviewer suggested adding the following text to the discussion of Metabolic Effects in Section 1.2:

*However some studies have shown that this association disappeared after adjusted for other lipophilic chemicals (Aminov et al., 2016; Codru et al., 2007; Everett and Matheson, 2010).*

***RESPONSE:*** *The intent of Section 1.2 is to provide an overview of the results of the epidemiological and laboratory animal studies. Given the number of epidemiological studies examining diabetes, it is not consistent with the toxicological profile guidance to cite all of the studies. Rather, several key studies are cited. The remaining studies finding associations with diabetes and those finding no associations are summarized in Section 2.18.*

**COMMENT:** The Reviewer suggested the following revisions in Section 1.2 (insertions marked in red and deletions in strikeout):

~~*Consistent evidence for no association was found in f*~~*Four case-control studies of pancreatic cancer (Gasull et al. 2010; Hardell et al. 2007; Hoppin et al. 2000) reported no association with DDT concentrations, but Garabrant et al. (1992)* ~~*and two*~~ *reported statistically significant association with both DDT and DDD and pancreatic cancer. No significant associations were found in two case-control studies of endometrial cancer (*Hardell et al. 2004; Sturgeon et al. 1998).

***RESPONSE:*** *The suggested revisions were not made to the discussion of Cancer Effects in Section 1.2. The focus of Section 1.2 is to provide the reader with an overview of the health effects database. Given*

*the amount of health effects data for DDT, the section focuses on endpoints in which there is consistent evidence of an association or no association. Within these discussions, citations are provided for studies that support these conclusions. A more thorough discussion of the available data is provided in Section 2.19.*

**COMMENT:** The Reviewer suggested the following addition to discussion of available epidemiological data in Section 2.:

> *However even in these studies there remains some uncertainly whether the associations observed are really due to DDT, DDE and/or DDD rather than other co-migrating lipophilic organohalogen chemicals.*

***RESPONSE:*** *The following text was added to this discussion in Section 2.1:*

> Although epidemiological studies provide consistent evidence of associations (or no associations) between DDT and some health outcomes, these data do not establish causality. Other factors, particularly co-exposure to other highly lipophilic compounds (e.g., PCBs, CDDs, CDFs), may have influenced the study results. Some of the epidemiological studies have statistically adjusted for exposure to one or more non-DDT compound to decrease the uncertainty; however, most studies did not include this adjustment.

**COMMENT:** Regarding the discussion of the deaths from pancreatic cancer reported in the Beard et al. 2003 study in Section 2.2, the Reviewer noted "This study is being given much too much weight. It is an occupational study of pesticide applicators where every individual was exposed to multiple pesticides. The DDT result is based only on exposure for the years after arsenic was used, and before modern pesticides were applied, but does not limit subjects to persons only exposed during that period of time. The pancreatic cancer was significant only for <fuve years exposure, and not for >five years exposure. I do not believe that one can make any conclusion regarding DDT exposure from this study, only pesticides in general. Furthermore you have stated previously that there was consistent evidence for no association with pancreatic cancer. That is not true, but the evidence is from the Garabrand study, not this one."

***RESPONSE:*** *The SMR of 5.27 was for workers only employed during the use of DDT and exposed for less than 3 years. The text was revised to indicate that an association was not found in workers exposed for ≥3 years:*

> This was not the case for pancreatic cancer; increases in deaths (SMR 5.27, 95% confidence interval [CI] 1.09–15.40) were found for workers with <3 years of DDT exposure, as compared the control population; no association was found in workers exposed to DDT for ≥3 years.

**COMMENT:** Regarding the discussion of the Cocco et al. (2005) study in Section 2.2, the Reviewer notes: "There is a major problem with all of these studies where serum DDE and/or DDT is monitored, in that without control for other organochoirnes that co-migrate with DDE it is not possible to conclude that the association is reallydue to DDE/DDT In this case they measured chlordane, but not other chlorinated pesticides, PCBs, dioxins/furan or brominated ompounds. Since this is the first place in the report that serum ample data is presented this may be the best play to expland and explain the problem.
I would suggest a new paragraph that says something like "Drawing conclusions based on serum DDE/DDt levels must be done with caution because of the fact that there are a number of lipophilic chemical that migrate with DDE/DDT, and often only a few are measured. In many cases indiviudls with elevated DDE/DDT will also have elevated levels of several other persistent chlorinated pesticides, PCBs, dioxins/furan and brominated flame retardants. Without adjustment for concentrations of other co-

migrating chemicals it is not certain that assoications, even if statistically significant, are really due to DDE/DDT."

***RESPONSE:*** *ATSDR agrees with the Reviewer that co-exposure to other persistent organic compounds limits the interpretation of the data. However, the wide range of populations examined, including DDT workers, does partially mitigate this uncertainty. To inform the reader of this uncertainty, the following text was added to Section 2.1:*

> Although epidemiological studies provide consistent evidence of associations (or no associations) between DDT and some health outcomes, these data do not establish causality. Other factors, particularly co-exposure to other highly lipophilic compounds (e.g., PCBs, CDDs, CDFs), may have influenced the study results. Some of the epidemiological studies have statistically adjusted for exposure to one or more non-DDT compound to decreases the uncertainty; however, most studies did not include this adjustment.

**COMMENT:** The Reviewer suggested the following revision (inserted text in red) in Section 2.3:

> *Consistent evidence for significant positive associations with BMI was found in most studies of adults ≥50 years of age with mean serum DDE levels (not adjusted for other lipophilic chemicals) lower than ~500 ng/g lipid (Arrebola et al. 2014; De Roos et al. 2012; Dhooge et al. 2010; Lee et al. 2012a; Schildkraut et al. 1999).*

***RESPONSE:*** *As noted in the Response to the previous comment, text was added to Section 2.1 noting that most studies did not statistically adjust for exposure to other compounds.*

**COMMENT:** Regarding the following sentence in Section 2.3—Lim et al. (2011) also showed an inverse relationship between weight change over a 10-year period and increased serum DDE, albeit at serum levels in the range of NHANES studies—the Reviewer commented: "This sentence is out of place. There is a lot of evidence that rapid weight loss resulted in elevated serum levels of persistent organics, but that is not what the rest of the paragraph relates to. The word "also" is not appropriate here"

***RESPONSE:*** *The sentence in question was revised:*

> Lim et al. (2011) found an inverse relationship between weight change over a 10-year period and increased serum DDE, albeit at serum levels in the range of NHANES studies.

**COMMENT:** The Reviewer suggested the addition of the Goncharov et al. 2011 study to Section 2.5.

***RESPONSE:*** *The Goncharov et al. 2011 study was added to the text of Section 2.5 and to Table 2-3.*

> A number of epidemiological studies have examined associations between serum or adipose levels of DDT, DDD, or DDE and cardiovascular outcomes, including general hypertension in adults (Arrebola et al. 2015b; Goncharov et al. 2011; Henriquez-Hernandez et al. 2014; Lee et al. 2007b; La Merrill et al. 2013; Lind et al. 2014; Park et al. 2016; Valera et al. 2013a, 2013b), gestational hypertension (Savitz et al. 2014), stroke (Lee et al. 2012a, 2012b), cardiovascular disease (Ha et al. 2007), and peripheral arterial disease (Min et al. 2011). The results provide inconsistent evidence for associations between serum or adipose levels of DDE or DDT and cardiovascular effects.
>
> In other epidemiological studies, no statistically significant positive associations with gestational hypertension (Savitz et al. 2014), cardiovascular disease (Ha et al. 2007), or systolic or diastolic blood pressure levels (Goncharov et al. 2011; Henriquez-Hernandez et al. 2014; Valera et al. 2013a, 2013b) were observed (**Error! Reference source not found.**).

**COMMENT:** The Reviewer suggested the addition of the following text to Section 2.5:

*In the study by Goncharov et al. (2011) significant association was found between blood pressure and hypertension for total PCBs, but not for any of nine chlorinated pesticides, including p,p' and o,p' DDT and DDE. This result suggests that some of the positive associations with blood pressure reported above may have been due to lipophilic chemicals other than DDT/DDE.*

***RESPONSE:*** *ATSDR agrees that the lack of control for exposure to other persistent organic compounds limits the interpretation of the results of DDT studies. A note added to the Introduction to Chapter 2 discusses this limitation:*

Although epidemiological studies provide consistent evidence of associations (or no associations) between DDT and some health outcomes, these data do not establish causality. Other factors, particularly co-exposure to other highly lipophilic compounds (e.g., PCBs, CDDs, CDFs), may have influenced the study results. Some of the epidemiological studies have statistically adjusted for exposure to one or more non-DDT compound to decreases the uncertainty; however, most studies did not include this adjustment.

**COMMENT:** The Reviewer suggested the following text be inserted into the discussion of hepatic mechanisms of action*: Rat hepatocytes exposed to 150 µM DDT showed changes in expression of genes related to estrogen, lipid and sugar metabolism (Jellali et al., 2018).*

***RESPONSE:*** *The following text was added to Section 2.9:*

*In vitro* studies in isolated hepatocytes also showed increases in expression of genes associated with hepatic estrogen, lipid, and sugar metabolism (Jellali et al. 2018).

**COMMENT:** The Reviewer suggested the addition of the following text to Section 2.13: "**…** (4) the lack of adjustment for other lipophilic chemicals that co-migrate in serum with DDT and DDE."

***RESPONSE:*** *Since the issue with co-exposure to other persistent organic compounds is a global issue, it was added to the introduction to Chapter 2:*

Although epidemiological studies provide consistent evidence of associations (or no associations) between DDT and some health outcomes, these data do not establish causality. Other factors, particularly co-exposure to other persistent organic compounds, may have influenced the study results. Some of the epidemiological studies have statistically adjusted for exposure to one or more non-DDT compound to decreases the uncertainty; however, most studies did not include this adjustment.

**COMMENT:** The Reviewer suggested the following addition to Section 2.14:

*In a study from South Africa where there is widespread indoor spraying of DDT, Huang et al. (2018) found elevated rates of childhood infections in relation to material DDT concentrations, and these associations were stronger among households below the poverty line and with poor maternal nutrition.*

***RESPONSE:*** *The Huang et al. (2018) study was added to Table 2-11 and the following text was added to Section 2.14:*

…elevated rates of persistent fever between 1 and 2 months were associated with p,p'-DDE, but not *o,p'*-DDT or *p,p'*-DDT and no associations were found for the number of ear infections or severe sore throats (Huang et al. 2018).

**COMMENT:** The Reviewer suggested the following revision to the discussion of mechanisms of action in Section 2.14: *alter expression of a variety of genes (Kiyosawa et al., 2008)*

***RESPONSE:*** *The Kiyosawa et al. (2008) study was not added since it did not discuss mechanisms of immunotoxicity.*

**COMMENT:** The Reviewer suggested the replacement of the Harada et al. (2016) with papers by Vijverberg et al. (1982) and Janicki and Kinter (1971) in the mechanisms of action discussion in Section 2.15.

***RESPONSE:*** *The suggested revision was made:*

> DDT has been shown to disrupt nerve membrane ion fluxes through induced closure of sodium channels (Vijverberg et al. 1982), inhibition of potassium transport, and by targeting $Na^{+}/K^{+}$ and $Ca^{2+}/Mg^{2+}$ ATPases (Janicki and Kinter 1971).

**COMMENT:** The Reviewer suggested the following addition to Section 2.16: *and in most case no attempt was made to determine whether effects seen were due to DDT or co-migrating organochlorine chemicals*

***RESPONSE:*** *Since the issue with co-exposure to other persistent organic compounds is a global issue, it was added to the introduction to Chapter 2:*

> Although epidemiological studies provide consistent evidence of associations (or no associations) between DDT and some health outcomes, these data do not establish causality. Other factors, particularly co-exposure to other highly lipophilic compounds (e.g., PCBs, CDDs, CDFs), may have influenced the study results. Some of the epidemiological studies have statistically adjusted for exposure to one or more non-DDT compound to decreases the uncertainty; however, most studies did not include this adjustment.

**COMMENT:** The Reviewer suggested the addition of the following sentence to the discussion of pancreatic cancer in Section 2.19: *However Garabrant et al. (1992) reported a RR of 4.8 (95% CI 1.3–17.6) in a case control study of DDT-exposed workers.*

***RESPONSE:*** *The following text was added to Section 2.19:*

> In contrast, there were associations between serum DDT and DDD levels and pancreatic cancer deaths in workers exposed at least 10 years prior to death (Garabrant et al. 1992). The RRs were 4.8 (95% CI 1.3–17.6) for DDT (6 cases, 7 controls) and 4.3 (95% CI 1.5–12.4) for DDD (9 cases, 12 controls).

**COMMENT:** The Reviewer suggested adding "Welfinger-Smith et al. 2011" to the statement in Section 5.1 regarding DDT levels in Artic marine life.

***RESPONSE:*** *The following revision was made:* Marine mammals in the Arctic often contain very high levels of DDT and DDE (Hargrave et al. 1992; Welfinger-Smith et al. 2011).

**COMMENT:** The Reviewer suggested the following revision in Section 5.1 (insertion marked in red):

*Human exposure to DDT is primarily through the diet. Exposure via inhalation at the ambient levels in air (Whitmore et al. 1994) is thought to be insignificant compared with dietary intake, although can be a significant exposure of people living in homes with indoor residual spraying in malaria endemic regions (Ritter et al., 2011).*

***RESPONSE:*** *The referenced statement was revised to clarify that it refers to U.S. exposures:*

Human exposure to DDT is primarily through the diet. In the United States, exposure via inhalation at the ambient levels in air (Whitmore et al. 1994) is thought to be insignificant compared with dietary intake.

**COMMENT:** The Reviewer suggested the following insertion to Section 5.1:

*While there has not been much study of inhalation as an important route of exposure, the concentrations of DDT/DDE in vapor phases in air are not insignificant (see section 5.5.1). Similar and even lower concentrations of other organochlorine compounds (PCBs) have been found to accumulate in bodies of teachers working in a school with contaminated air (Gabrio et al., 2000), and several have suggested that inhalation of PCBs may be more important than usually considered (Carpenter, 2015; Lehmann et al., 2015). Because humans breathe 23/7 even low concentrations of DDT/DDE in air may be absorbed and accumulate over time. Further study of inhalation as a route of exposure to DDT/DDE is necessary.*

***RESPONSE:*** *Inclusion of a discussion of inhalation exposure to PCB was not considered relevant for the Overview section of Chapter 5. A discussion of potential inhalation exposure via the inhalation route is discussed in Section 5.6.*

**COMMENT:** The Reviewer suggested the following addition (marked in red) to the first sentence in Section 5.6: *The general population is currently exposed to DDT and its metabolites primarily in food, although inhalation may contribute as well.*

***RESPONSE****: The referenced sentence was revised:* The general population is currently exposed to DDT and its metabolites primarily in food; with smaller amounts coming from inhalation exposure.

**COMMENT:** Regarding the following sentence in Section 5.6: DDT and its metabolites are ubiquitous in the atmosphere, but are present in such low concentrations that exposure via inhalation is negligible; the Reviewer commented "I strongly disagree with this conclusion. The concentrations of total DDT in air are not insignificant, as reported in Section 5.5.1. People breath 24/7 for life. As indicated earlier air concentrations of PCBs at comparable concentrations get absorbed and contributed significantly to body burden. It is true that this has not been demonstrated for DDT, but I think it is a great mistake to state that inhalation is not an important route of exposure." The Reviewer also suggested deleting the statement "Potential inhalation of relatively high levels of DDT should be possible only in areas of production or formulation."

***RESPONSE:*** *The discussion of potential inhalation exposure to DDT in Section 5.6 has been revised:*

DDT and its metabolites are ubiquitous in the atmosphere, but are typically present in in low concentrations. In 1986–1988, EPA collected data at two sites, Jacksonville, Florida and Springfield/Chicopee, Massachusetts, to assess the nonoccupational exposure to pesticides (NOPES) for residents of these cities (Whitmore et al. 1994). Indoor *p,p'*-DDE and *p,p'*-DDT levels in air were higher than outdoor levels in these communities, and the highest number of

indoor air samples with detectable DDT was observed in the spring season in Jacksonville (14%) and in the winter season in Springfield/Chicopee (20%), with estimated mean air DDE and DDT concentrations of ≤1.0 ng/$m^3$. Mean ΣDDT air exposures were estimated as 22 ng/day in Jacksonville and 94 ng/day in Springfield/Chicopee. For comparison, dietary exposures in these two communities for 1982–1984 were estimated to be around 1,900 ng/day. Nine of 11 carpets tested in Jacksonville contained ΣDDT with median and mean levels of 0.7 and 1.2 μg/g, respectively.

**COMMENT:** Regarding the statement in Section 6.2—No chronic-duration toxicity studies in which animals were exposed dermally or by inhalation were located. Additional information on possible toxicity from these exposure routes do not appear to be needed, because they represent minor routes of human exposure—the Reviewer commented: "Again I very much disagree. In my judgment studies on inhalation should be done and results extrapolated to air concentrations to which humans are exposed"

***RESPONSE:*** *The referenced sentence was revised:*

No chronic-duration inhalation toxicity studies in animals that could be used to establish concentration-response relationships were located.

*Additionally, the first sentence in the Health Effects subsection was revised to indicate the need for inhalation studies:*

Toxicity studies of laboratory animals exposed by the inhalation or dermal routes are very small in number. Air monitoring data suggest that DDT and its metabolites are still present decades after its use was banned. Health effect studies are needed to evaluate possible health effects associated with long-term exposure to low concentrations of DDT in air.

## Comments provided by Peer Reviewer #2

## General Comments

**COMMENT:** Summary report for reviewer #2: Overall, this Tox Profile is a very comprehensive resource for both health care providers and scientists. I have mentioned some issues, mainly in Chapter 2, that need to be addressed. However, some of these are studies which the authors of this profile may have deemed not to be applicable. If so, then I default to the authors' judgement but I felt they were worth mentioning. My comments are outlined below each question in Arial font.

***RESPONSE:*** *No response needed.*

## ATSDR Charge Questions and Responses

### *Chapter 1*

**QUESTION:** Do you agree with those effects known to occur in humans as reported in the text? If not, provide a copy of additional references you would cite and indicate where (in the text) these references should be included.

**COMMENT:** The effects known to occur in humans, those where positive associations have been found (diabetes), and those where epidemiological data are inconclusive (neurological disorders such as Alzheimers) were covered well.

***RESPONSE:*** *No response needed.*

**QUESTION:** Are the effects only observed in animals likely to be of concern to humans? Why or why not? If you do not agree, please explain.

**COMMENT:** This is a challenging question. The acute effects of high dose exposure are highly unlikely to occur in the US given the DDT ban. However, some of the transgenerational effects that have occurred in animal studies (i.e. obesogenic effect in generational studies) are difficult to extrapolate to humans given the longevity needed to study this in humans and multitude of extraneous factors that humans are constantly exposed to which may alter disease outcome.

***RESPONSE:*** *No response needed.*

**QUESTION:** Have exposure conditions been adequately described? If you disagree, please explain.

**COMMENT:** Current exposure conditions both in the US and abroad have been well described.

***RESPONSE:*** *No response needed.*

**QUESTION:** Do you believe the derived acute, intermediate, and chronic oral MRLs values are justifiable? If you disagree, please explain. (see also Appendix A).

**COMMENT:** Yes, the oral MRL values appear to be justifiable given the current exposure data.

***RESPONSE:*** *No response needed.*

**QUESTION:** Do you agree that the data do not support derivation of acute, intermediate, and chronic inhalation MRLs?

**COMMENT:** Yes, I agree that the data do not support derivation of inhalation MRLs given that oral exposure is the primary route of exposure in the US. Derivation of inhalation MRLs would be less reliable than oral MRLs given the lack of data regarding inhalation exposure.

***RESPONSE:*** *No response needed.*

## *Chapter 2*

**QUESTION:** Do the health effect conclusions made in Chapter 2 adequately reflect the findings in the published literature for DDT, DDE, and DDD?

**COMMENT:** Yes, the major health effect conclusions are clearly defined and delineated for consistent associations between reproductive effects, asthmatic wheeze, type 2 diabetes (T2D), and liver carcinogenicity. These are reflected in the included literature reports.

***RESPONSE:*** *No response needed.*

**QUESTION:** Were adequately designed human studies identified in the text (i.e., good exposure data, sufficiently long period of exposure to account for observed health effects, adequate control for confounding factors)? Were the major study limitations sufficiently described in the text without going into lengthy discussions? If study limitations were not adequately addressed, please suggest appropriate changes.

**COMMENT:** The section did a very good job of highlighting the major human studies which drive each health effect and highlighting the strengths and weaknesses of the literature base. I have made minor comments noting mention of potential mixture effects earlier in the discussion prefacing studies examining DDT and T2D.

***RESPONSE:*** *See Responses in the Annotated Comments section.*

**QUESTION:** Were adequately designed animal studies identified in the text (i.e., adequate number of animals, good animal care, accounting for competing causes of death, sufficient number of dose groups, and sufficient magnitude of dose levels)? If not, does the inadequate design negate the utility of the study? Please explain.

**COMMENT:** Yes, this section highlighted the multitude of animal studies examining the major endpoints of DDT or metabolite toxicities. With regard to dose, it should be mentioned in the introduction to the animal study sections that the doses which are utilized in a majority of studies are in extreme excess of which would be routinely encountered via modern oral exposure in the US.

***RESPONSE:*** *The following statement was added to Section 1.2:*

In most cases, the doses associated with adverse effects in laboratory animals are higher than could be reasonable anticipated from background exposure in the United States.

**QUESTION:** Were the animal species appropriate for the most significant toxicological endpoint of the study? If not, which animal species would be more appropriate and why?

**COMMENT:** The clear majority of animal studies utilized rodent models (mouse or rat) which are the most routinely used in toxicity testing. However, there did appear to be a species related sensitivity to the carcinogenic effect of DDT with mice being more susceptible than rats and non-human primates. This should be mentioned in that section.

***RESPONSE:*** *Species differences in the carcinogenicity of DDT are noted in Section 2.19:*

Intermediate exposures, in which animals were exposed to DDT in food, caused cancer increases in mice, but not in rats or hamsters. Mice that were observed for 50–105 weeks after cessation of treatment developed liver hepatomas following dietary exposure to 42.8 mg *p,p'*-DDT/kg/day for 15–30 weeks (Tomatis et al. 1974b). DDT did not produce increases in the tumor incidence in rats exposed to 10–20 mg/kg/day in the food for up to 45 weeks (*p,p'*-DDT: Kimbrough et al. 1964; technical DDT: Laug et al. 1950; DDT(NS): Numoto et al. 1985) or in hamsters fed 50 mg *p,p'*-DDT/kg/day for 30 weeks (Tanaka et al. 1987).

Chronic exposure (>1 year) to technical DDT, *p,p'*-DDT, or DDT(NS) caused cancer in multiple strains of mice and in some rat studies, but not in dogs; most studies in nonhuman primates have also been negative.

**QUESTION:** Are you aware of any studies that are not included in the profile that may be important in evaluating the toxicity of DDT, DDE, and DDD? Please provide a copy of each study and indicate where in the text each study should be included.

**COMMENT:** Mangum et al (2016) Exposure to p,p'-DDE Alters Macrophage Reactivity and Increases Macrophage Numbers in Adipose Stromal Vascular Fraction. Tox Sci. PMID: 26748080. This study examine the effects of DDE exposure on macrophage reactivity highlighting a pro-inflammatory phenotype following in vitro exposure. Additionally, this study employs a 5 day 2.0 mg/kg oral dosing then demonstrates increased macrophage recruitment to the epididymal adipose tissue which would be characterized as a potential in vivo immunological effect. I have made comments in the profile document regarding potential insertion of this study in both immunological effects and potential mechanisms sections.

Mangum et al (2015) Exposure to p,p'-DDE enhances differentiation of 3T3-L1 preadipocytes in a model of sub-optimal differentiation. Tox Letters. PMID: 26200599. This study demonstrates increased adipogenesis following DDE exposure in a murine preadipocyte model which supports studies already cited in "Mechanisms of Body Weight Effects" section as noted in the profile document.

***RESPONSE:*** *The Mangum et al. (2016) paper was added to Section 2.14:*

Disruptions to humoral and cell-mediated immune responses could be due to a variety of cellular and system responses that have been observed *in vitro*. Exposure to DDT or to related compounds has been shown to increase ROS, nitric oxide (NO), or TNF-α production (Perez-Moldonado et al. 2005; Dutta et al. 2008); induce pro-inflammatory responses (Gaspar-Ramirez et al. 2015c; Kim et

al. 2004); alter inflammatory mediator production (Mangum et al. 2016) and apoptotic pathways (Alegria-Torres et al. 2009; Perez-Moldonado et al. 2004, 2005); alter immune cell morphologies and activity (Dutta et al. 2008; Reed et al. 2004; Udoji et al. 2010); and lead to aberrant cytokine production (Alegria-Torres et al. 2009; Kim et al. 2004; Quaranta et al. 2006) and alterations in the complement system (Dutta et al. 2008).

*The Mangum et al. (2015) paper was added to Section 2.18:*

Several studies demonstrate the ability of DDT to disrupt lipid homeostasis. *In vitro* studies with *p,p'*-DDT or *p,p'*-DDE suggest there may be AhR-independent effects causing increased adipogenesis (Kim et al. 2016; Mangum et al. 2015; Moreno-Aliaga and Matsumuru 2002), adipocyte fatty acid uptake (Howell and Mangum 2011), and dipokine (adiponectin, leptin, resistin) levels (Howell and Mangum 2011).

**QUESTION:** Are you aware of any studies that are not included in the profile that may be relevant to deriving MRLs for any of the DDT isomers?

**COMMENT:** No, I am not aware of any additional studies that are not included that may be relevant to deriving MRLs for the isomers.

***RESPONSE:*** *No response needed.*

**QUESTION:** Were all appropriate NOAELs and/or LOAELs identified for each study (both in the text and the Levels of Significant Exposure (LSE) tables and figures)? If not, did the text provide adequate justification for excluding NOAELs/LOAELs including, but not limited to, citing study limitations? Please suggest appropriate changes.

**COMMENT:** Yes, all appropriate NOAELs and LOAELs are identified for each study. However, there are a few points listed below for consideration which may alter these values.

In the acute DDE exposure by Howell et al (2015) the raw body weights are significantly greater in DDE exposed animals compared to vehicle but the percent changes are not significantly greater. Thus, this needs to be specified as to which measurement the LOAEL for body weight was determined. This may alter the less serious LOAEL for acute DDE with regards to body weight.

In table 2-1, figure keys 129 and 130 which are studies by LaMerrill et al. these are just classified as developmental effects whereas the endpoints that were measured were metabolic (figure key 129) and cardiovascular (figure key 130). While the DDT exposure was gestational/developmental, the endpoints were measured later on in life. Should there be additional classifications to the findings in this table? The findings from figure key 130 are discussed in the "Evidence of Cardiovascular Effects of DDT, DDD, or DDE in Animals" section and findings from figure key 129 in the "Studies of Diabetic Outcomes in Laboratory Animals" section. If so, these studies would need to be put into the appropriate LOAEL and NOAEL classifications.

***RESPONSE:*** *The results of the Howell et al. (2015) study were corrected; the 2 mg/kg/day dose was identified as a NOAEL for body weight effects in Table 2-1. The effects reported in the La Merrill et al. 2014a, 2014b, 2016) studies were considered developmental effects since they involved perinatal exposure. A statement was added to the Developmental section (2.17) pointing the reader to Sections 2.5, 2.10, and 2.18 for discussions of the cardiovascular, renal, and diabetic outcome results reported in the La Merrill et al. (2014a, 2014b, 2016) studies:*

Numerous adverse developmental outcomes have been reported in the offspring of rodents exposed to DDT/DDE during gestation and/or lactation. Effects include fetotoxicity, alterations in growth, neurodevelopmental toxicity, and impaired development of the reproductive system; these effects are discussed in greater detail below. Some developmental studies have also evaluated cardiovascular (La Merrill et al. 2016), renal (La Merrill et al. 2016), and diabetic (La Merrill et al. 2014a, 2014b) outcomes in the offspring of rats perinatally exposed and evaluated as adults; these effects are considered developmental toxicity and are discussed in Sections 2.5, 2.10, and 2.18, respectively.

**QUESTION:** Do you agree with the categorization of "less serious" or "serious" for the effects cited in the LSE tables?

**COMMENT:** Yes, most of the characterizations seem appropriate with a majority of the serious effects being exacerbation of the less serious in a dose dependent manner which is rational when both “less serious” and “serious” effects were noted.

***RESPONSE:*** *No response needed.*

**QUESTION:** Have all possible mechanisms of action been discussed within their relevant health effect section? If not, please explain.

**COMMENT:** Yes, it appears that the major mechanisms of action (if known) are discussed within the relevant health effect section.

***RESPONSE:*** *No response needed.*

## *Chapter 3*

### *Toxicokinetics*

**QUESTION:** Is there adequate discussion of absorption, distribution, metabolism, and excretion of the substance? If not, suggest ways to improve the text.

**COMMENT:** Overall, the discussion of ADME highlighted the major known aspects of DDT/DDE toxicokinetics. There have not been many new studies examining this aspect of DDT/DDE which is actually needed given the positive associations between DDT/DDE exposure and diabetes and weight gain which may alter ADME.

One recent study by Howell et al (2014) PMID: 24582731 examined DDE levels in serum, adipose, and liver at 7, 14, and 21 days following 5 day acute exposure to DDE. They noted an increase in adipose DDE from 7 to 21 days with a corresponding decrease in both liver and serum levels which is most likely due to distribution.

***RESPONSE:*** *The Howell et al. (2014) study was added to Section 3.1.2:*

Another study of rats administered DDE for 5 days showed that serum and liver DDE levels significantly decreased between 7 and 21 days post-exposure; in contrast, adipose levels increased (although the change between days 7 and 21 was not statistically significant) (Howell et al. 2014).

**QUESTION:** Have all available pharmacokinetic/pharmacodynamic models and supporting data been presented? If not, please explain.

**COMMENT:** Yes, there have not been any new models published to my knowledge.

***RESPONSE:*** *No response needed.*

**QUESTION:** Is there adequate discussion of the differences in toxicokinetics between humans and animals? Is there adequate discussion of the relevance of animal toxicokinetic information for humans?

**COMMENT:** Yes, given the lack of toxicokinetic data in humans there is relatively little information to compare. However, the known differences have been noted.

***RESPONSE:*** *No response needed.*

***Children and Other Populations that are Unusually Susceptible***

**QUESTION:** Are there any data relevant to child health and developmental effects that have not been discussed in the profile and should be? Please provide any relevant references.

**COMMENT:** No, I am not aware of any studies which were not already discussed in the profile.

***RESPONSE:*** *No response needed.*

**QUESTION:** Is there a discussion of populations at higher risk of susceptibility? Do you agree with the choice of populations? Please explain and provide any additional relevant references.

**COMMENT:** Yes, there is a discussion of high risk populations with developmental exposures and elderly populations being highlighted. These are appropriate populations which may be at increased risk of susceptibility. Of note, there is an increase in interest of the menopausal transition being a potential window of susceptibility.

***RESPONSE:*** *No response needed.*

***Biomarkers of Exposure and Effect***

**QUESTION:** Are the biomarkers of exposure specific for the substance? Please explain.

**COMMENT:** Yes, the primary biomarker of DDT exposure that is routinely used is the primary bioaccumulative metabolite, DDE. Additionally, DDT has been measured as an index of exposure. The caveat is also presented that serum or adipose DDT/DDE levels can not predict dosing levels of previous exposure, just bioaccumulation.

***RESPONSE:*** *No response needed.*

**QUESTION:** Are the biomarkers of effect specific for the substance? Please explain.

**COMMENT:** There are no known or proven biomarkers of effect for DDT/DDE which is an area warranting future investigation.

***RESPONSE:*** *No response needed.*

***Interactions with Other Chemicals***

**QUESTION:** Is there adequate discussion of the interactive effects with other substances? Does the discussion concentrate on those effects that might occur at hazardous waste sites? Please explain and provide any additional references.

**COMMENT:** Yes, there is discussion of the interaction with other substances especially with regard to the ability of DDT/DDE to induce cytochrome P450s and potentially bioactivate or detoxify other xenobiotics. Both procarcinogenic and anticarcinogenic effects of other chemicals when given with DDT/DDE were discussed. Additionally, potentiation or inhibition of neuroactive chemicals was addressed.

***RESPONSE:*** *No response needed.*

**QUESTION:** If interactive effects with other substances are known, does the text discuss the mechanisms of these interactions? Please explain and provide any additional references.

**COMMENT:** Yes, the major mechanisms that were mentioned were metabolic activation or inactivation as well as potentiation of effects on neuronal sodium channels.

***RESPONSE:*** *No response needed.*

## *Chapter 4*

**QUESTION:** Are any of the values or information provided in the chemical and physical properties tables wrong or missing? Please explain and provide any additional references.

**COMMENT:** These appear to be complete.

***RESPONSE:*** *No response needed.*

**QUESTION:** Is information provided on the various forms of the substance? Please explain.

**COMMENT:** Yes, information is given on DDT and its metabolites and impurities.

***RESPONSE:*** *No response needed.*

*Chapter 5*

**QUESTION:** Is the information on production, import/export, use, and disposal of the substance complete? Please explain and provide any additional relevant references.

**COMMENT:** Yes, the information is as complete as possible with all facets being addressed.

***RESPONSE:*** *No response needed.*

**QUESTION:** Has the text appropriately traced the substance from its point of release to the environment until it reaches the receptor population? Does the text provide sufficient and technically sound information regarding the extent of occurrence at NPL sites? Do you know of other relevant information? Please provide references for added information.

**COMMENT:** Yes, the text addresses production and how the DDT compounds travel through the environment including air and water. The text clearly delineates how many NPL sites have tested positive for DDT compound contamination.

***RESPONSE:*** *No response needed.*

**QUESTION:** Does the text cover pertinent information relative to transport, partitioning, transformation, and degradation of the substance in all media? Do you know of other relevant information? Please provide references for added information.

**COMMENT:** Transport, partitioning, transformation, and degradation are well addressed in all media that DDT contamination would be likely to occur.

***RESPONSE:*** *No response needed.*

**QUESTION:** Does the text provide information on levels monitored or estimated in the environment, including background levels? Are proper units used for each medium? Does the information include the form of the substance measured? Is there an adequate discussion of the quality of the information? Do you know of other relevant information? Please provide references for added information.

**COMMENT:** This is an extensive section where environmental levels, including major food stuffs, are measured and provided.

***RESPONSE:*** *No response needed.*

**QUESTION:** Does the text describe sources and pathways of exposure for the general population and occupations involved in the handling of the substance, as well as populations with potentially high exposures? Do you agree with the selection of these populations? If not, why? Which additional populations should be included in this section?

**COMMENT:** Yes, the major route of exposure, ingestion of contaminated food, is well addressed and data is provided demonstrating that children < 2 years of age would be the most likely or highly exposed due to milk consumption. This section covers all major populations/ages with current exposure levels.

Additionally, this section makes if very clear that exposure via DDT/DDE measurement can not predict isolated exposures but is reflective of cumulative body burden.

***RESPONSE:*** *No response needed.*

## *Chapter 6*

**QUESTION:** Do you know of other studies that may fill a data gap? Please provide any relevant references.

**COMMENT:** Toxicokinetic studies in normal and obese models are lacking which may provide novel information regarding compound fate in a physiological state that may predispose toward compound accumulation (obesity). I see this as a data need.

***RESPONSE:*** *The following statement was added to Section 6.2 (Absorption, Distribution, Metabolism, Excretion subsection):*

> However, there are limited data to evaluate potential toxicokinetic differences associated with obesity or diabetes. These data would be useful given the associations between DDT/DDE exposure and obesity and DMT2.

**QUESTION:** Do you agree with the identified data needs? Please explain.

**COMMENT:** Yes, for the most part more developmental exposure data is needed given this is a likely time of higher exposure and a known window of susceptibility for other environmental contaminants.

***RESPONSE:*** *No response needed.*

**QUESTION:** Are the data needs presented in a neutral, non-judgemental fashion? Please note any bias in the text.

**COMMENT:** I did not note any significant bias.

***RESPONSE:*** *No response needed.*

## *Chapter 7*

**QUESTION:** Are you aware of any additional regulations or guidelines that we should add? Please provide citations.

**COMMENT:** The regulations and guidelines appear to be complete.

***RESPONSE:*** *No response needed.*

**QUESTION:** Are there any that should be removed? Please explain.

**COMMENT:** No

***RESPONSE:*** *No response needed.*

## *Appendix A*

**QUESTION:** Do you agree or disagree with the proposed acute, intermediate, and chronic oral MRL values? Explain. If you disagree, please specify the MRL value that you propose.

**COMMENT:** I agree with the proposed acute, intermediate, and chronic MRL values based on the currently available data.

***RESPONSE:*** *No response needed.*

**QUESTION:** Do you agree/disagree with each component of the total uncertainty factor? Explain. If you disagree, please specify the uncertainty factor(s) that you propose.

**COMMENT:** The uncertainty factor components appear rationale for each MRL.

***RESPONSE:*** *No response needed.*

**QUESTION:** Please comment on any aspect of our MRL database assessment that you feel should be addressed.

**COMMENT:** The database assessment appears to be comprehensive.

***RESPONSE:*** *No response needed.*

**QUESTION:** Do you agree or disagree with proposed no MRL for any inhalation duration?

**COMMENT:** Yes, I agree with the no MRL for inhalation. There is not a sufficient data set to allow for an accurate calculation of this value.

***RESPONSE:*** *No response needed.*

## *Appendix B*

**QUESTION:** Does Appendix B provide a sufficiently clear documentation of ATSDR's health effects literature search strategy and inclusion/exclusion criteria?

**COMMENT:** Tables B2 and B3 are very cumbersome and difficult to interpret in a rapid manner.

***RESPONSE:*** *Tables B-2 and B-3 list the query strings used for the literature search and they are intended to be detailed documentation of the literature search strategy. For most users of the toxicological profile, the discussion of the literature search and screen provided in Section B.1, as well as the inclusion criteria in Table B-1, provides an adequate high level summary of the literature search strategy.*

**QUESTION:** Does it provide enough transparency regarding ATSDR's implementation of its inclusion and exclusion criteria (e.g., how ATSDR chose the studies it included in the health effects chapter)?

**COMMENT:** Again, the above mentioned tables are cumbersome. However, Section B.1.2. Literature Screening does a good job of explaining the inclusion and exclusion criteria. The flow chart (Figure B.1) is easy to follow.

***RESPONSE:*** *No response needed.*

### *Overall Usability of the Profile*

**QUESTION:** Does the new chapter organization make it easier for you to find the information you need? For example, are you satisfied with the organization of the health effects chapter by organ system rather than exposure route?

**COMMENT:** Yes, I like the new organization. Given that there is one major exposure route for DDT/DDE since its ban, the organ system effects are better than exposure route.

***RESPONSE:*** *No response needed.*

**QUESTION:** Does the profile contain all of the information you need? Is there information you would like to see that is not currently included?

**COMMENT:** The profile is a very comprehensive resource on DDT/DDE. It contains all of the major information that a health care provider or a scientist would need.

***RESPONSE:*** *No response needed.*

**QUESTION:** If you have used the Toxicological Profiles before, which chapter(s) have you used the most and for what purpose?

**COMMENT:** I have used the DDT/DDE Tox Profile before and I've found the exposure and health effects chapters to be the most useful. I've used these for both grant proposal generation and manuscript preparation.

***RESPONSE:*** *No response needed.*

**QUESTION:** Are the new tables and figures clear and useful? Do they make the toxicological profile easier to read?

**COMMENT:** The tables and figures are very helpful because this is a large profile that is very data rich.

***RESPONSE:*** *No response needed.*

## Annotated Comments

**COMMENT:** Regarding the sentence in Section 1.2— Elevated fasting blood glucose levels were observed in adult mice 3 weeks following 5-day oral exposure to 2 mg/kg/day *p,p'*-DDE, but not 0.4 mg/kg/day, but fasting blood glucose levels were not elevated after 4, 8, or 13 weeks of exposure (Howell et al. 2014, 2015).—the Reviewer commented "There was a significant dietary interaction where DDE + HFD elevated FBG at 4, 8, and 12 weeks."

***RESPONSE:*** *The discussion of the dietary interactions between DDE and fat content of the diet are discussed in Section 2.18.*

**COMMENT:** The Reviewer made the following comment in Section 1.3: "Oral is primary route of exposure in US given the ban so this is appropriate."

***RESPONSE:*** *No response needed.*

**COMMENT:** The Reviewer made the following comment on the Nims et al. (1998) study in Table 2-1: "Mangum et al 2016 shows increase epididymal adipose macrophage infiltration"

***RESPONSE:*** *The Mangum et al. (2016) was added to the discussion of Mechanisms of Action in Section 2.14:*

> Exposure to DDT or to related compounds has been shown to increase ROS, nitric oxide (NO), or TNF-α production (Perez-Moldonado et al. 2005; Dutta et al. 2008); induce pro-inflammatory responses (Gaspar-Ramirez et al. 2015c; Kim et al. 2004); alter inflammatory mediator production (Mangum et al. 2016) and apoptotic pathways (Alegria-Torres et al. 2009; Perez-Moldonado et al. 2004, 2005); alter immune cell morphologies and activity (Dutta et al. 2008; Reed et al. 2004; Udoji et al. 2010); and lead to aberrant cytokine production (Alegria-Torres et al. 2009; Kim et al. 2004; Quaranta et al. 2006) and alterations in the complement system (Dutta et al. 2008).

**COMMENT:** The Reviewer made the following comment on the Howell et al. (2014) study in Table 2-1: "Once this was normalized to predose BW, the % change was not statistically significant although raw body weights were significant."

***RESPONSE:*** *Table 2-1 was corrected to indicate that the 2 mg/kg/day was a NOAEL for body weight effects.*

**COMMENT:** The Reviewer made the following comment on the Howell et al. (2015) study in Table 2-1: "Dietary interaction between DDE and HFD to produce transient hyperglycemia, decreased HFD-induced hyperphagia"

***RESPONSE:*** *The interaction between DDE and dietary fat levels are discussed in the text in Section 2.18.*

**COMMENT:** The Reviewer made the following comment on the LaMerrill et al. (2014a, 2014b) study in Table 2-1: "Should multiple endpoints be considered?"

***RESPONSE:*** *ATSDR considers all of the observed effects in this study to be developmental since exposure occurred between gestation day 12 and postnatal day 5.*

**COMMENT:** The Reviewer made the following comment on the LaMerrill et al. (2014a, 2014b) study in Table 2-1: "While this is classified as developmental exposure, should the endpoints be separated as metabolic and hematological?"

***RESPONSE:*** *ATSDR considers these effects to be developmental since exposure occurred between gestation day 12 and postnatal day 5.*

**COMMENT:** The Reviewer made the following comment on the LaMerrill et al. (2016) study in Table 2-1: "Consideration of multiple endpoints?"

***RESPONSE:*** *ATSDR considers all of the observed effects in this study to be developmental since exposure occurred between gestation day 12 and postnatal day 5.*

**COMMENT:** The Reviewer made the following comment on the LaMerrill et al. (2016) study in Table 2-1: "While this is classified as developmental exposure, should the endpoint be cardiovascular?"

***RESPONSE:*** *ATSDR considers the effects observed in the offspring to be developmental effects.*

**COMMENT:** The Reviewer made the following comment on the Howell et al. (2015) study in Figure 2-2: "Since percent change in body weight in #62 were not significant but raw body weights were then this does not suggest a significant obesogenic effect. However, there was a trending increase in percent change to accompany the raw body weight increase."

***RESPONSE:*** *ATSDR revised Figure 2-2 to indicate that the 2 mg/kg/day dose is a NOAEL for body weight effects.*

**COMMENT:** The Reviewer made the following comment on Figure 2-2: "If the developmental endpoint in studies 129 and 130 are expanded to cardiovascular, endocrine, and hematological then this will need to be revised."

***RESPONSE:*** *As noted in the response to previous comments on the La Merrill et al. (2014a, 2014b, 2016) studies, the cardiovascular, endocrine, and hematological effects observed in the offspring were considered developmental effects.*

**COMMENT:** Regarding the statement— male mice exposed to up to 2 mg *p,p'*-DDE/kg/day for 5 days (Howell et al. 2014)—in Section 2.3, the Reviewer commented: ". Does this conflict with table on page 30-31 and study #62? % change was not significant but raw body weight increase was significant."

***RESPONSE****: Since no significant alterations in body weight were observed after corrected for pre-dosing weight, the 2 mg/kg/day dose was considered a NOAEL for body weight effects.*

**COMMENT:** Regarding the statement— The human epidemiological studies are consistent with the hypothesis that endocrine disrupting compounds (EDCs), including DDT, may act as obesogens that display non-monotonic dose-response relationships, leading to weight gain at lower levels, but to growth restriction or weight loss at higher doses (Tang-Peronard et al. 2011)—in Section 2.3, the Reviewer commented: "GOOD!"

***RESPONSE:*** *No response needed.*

**COMMENT:** Regarding the statement—Further studies may increase understanding of the complexities between the timing of DDT exposure, differences between DDT metabolites, dose, and gender, as well as the influences of initial weight status and significant weight change on serum levels of DDT and DDT toxicity (La Merrill et al. 2013).—in Section 2.3, the Reviewer commented "Mangum et al 2015 demonstrated increased adipogenesis with DDE exposure with suboptimal differentiation conditions with corresponding increases in adipogenic marker genes. Additionally, Kim et al 2016 reinforced these studies with adipogenic effects of DDT and DDE."

***RESPONSE:*** *The Mangum et al. (2015) and Kim et al. (2016) studies were added to the Mechanisms of Action discussion in Section 2.18:*

> Several studies demonstrate the ability of DDT to disrupt lipid homeostasis. *In vitro* studies with *p,p'*-DDT or *p,p'*-DDE suggest there may be AhR-independent effects causing increased adipogenesis (Kim et al. 2016; Mangum et al. 2015; Moreno-Aliaga and Matsumuru 2002), adipocyte fatty acid uptake (Howell and Mangum 2011), and dipokine (adiponectin, leptin, resistin) levels (Howell and Mangum 2011).

**COMMENT:** Regarding the discussion of the La Merrill studies in Section 2.5, the Reviewer commented "Classified as cardiovascular here but developmental in table."

***RESPONSE:*** *The effects reported in the La Merrill et al. 2016) studies were considered developmental effects since they involved perinatal exposure. A statement was added to the Developmental section (2.17) pointing the reader to Sections 2.5, 2.10, and 2.18 for discussions of the cardiovascular, renal, and diabetic outcome results reported in the La Merrill et al. (2014a, 2014b, 2016) studies:*

> Numerous adverse developmental outcomes have been reported in the offspring of rodents exposed to DDT/DDE during gestation and/or lactation. Effects include fetotoxicity, alterations in growth, neurodevelopmental toxicity, and impaired development of the reproductive system; these effects are discussed in greater detail below. Some developmental studies have also evaluated cardiovascular (La Merrill et al. 2016), renal (La Merrill et al. 2016), and diabetic (La Merrill et al. 2014a, 2014b) outcomes in the offspring of rats perinatally exposed and evaluated as adults; these effects are considered developmental toxicity and are discussed in Sections 2.5, 2.10, and 2.18, respectively.

**COMMENT:** Regarding the statement—. Information on possible immunological effects in laboratory animals after acute oral exposure to DDT and related compounds is restricted to a report that dietary exposure of New Zealand rabbits to 4.3 mg DDT(NS)/kg/day for 10 days produced no effects on serum antibody titers to *Salmonella typhi* infection (Shiplov et al. 1972) in Section 2.14, the Reviewer commented "It's not restricted if there's another report."

***RESPONSE:*** *The statement is referring to acute oral exposure studies; the second study discussed in this paragraph (Cetkovic-Cvrlje et al. 2016) is an intraperitoneal injection study.*

**COMMENT:** Regarding the discussion of acute data in Section 2.14, the Reviewer commented "Mangum et al 2015 showed increase macrophage infiltration of epididymal adipose but no significant alterations in reactivity."

***RESPONSE:*** *The Mangum et al. (2015) study was added to the Mechanisms of Action discussion in Section 2.18:*

> Several studies demonstrate the ability of DDT to disrupt lipid homeostasis. *In vitro* studies with *p,p'*-DDT or *p,p'*-DDE suggest there may be AhR-independent effects causing increased adipogenesis (Kim et al. 2016; Mangum et al. 2015; Moreno-Aliaga and Matsumuru 2002), adipocyte fatty acid uptake (Howell and Mangum 2011), and dipokine (adiponectin, leptin, resistin) levels (Howell and Mangum 2011).

**COMMENT:** Regarding the discussion of the mechanisms of immunotoxicity in Section 2.14, the Reviewer commented "Magnum et al. 2016 should be included."

***RESPONSE:*** *The Magnum et al. (2016) paper was added to Section 2.14:*

> Disruptions to humoral and cell-mediated immune responses could be due to a variety of cellular and system responses that have been observed *in vitro*. Exposure to DDT or to related compounds has been shown to increase ROS, nitric oxide (NO), or TNF-α production (Perez-Moldonado et al. 2005; Dutta et al. 2008); induce pro-inflammatory responses (Gaspar-Ramirez et al. 2015c; Kim et al. 2004); alter inflammatory mediator production (Mangum et al. 2016) and apoptotic pathways (Alegria-Torres et al. 2009; Perez-Moldonado et al. 2004, 2005); alter immune cell morphologies and activity (Dutta et al. 2008; Reed et al. 2004; Udoji et al. 2010); and lead to aberrant cytokine production (Alegria-Torres et al. 2009; Kim et al. 2004; Quaranta et al. 2006) and alterations in the complement system (Dutta et al. 2008).

**COMMENT:** Regarding the statement—Clear explanations of the inconsistency across the studies are not available, but possible contributors could be small case sample sizes and different distributions of age, BMI, gender, and exposure levels—in Section 2.18, the Reviewer commented "It may be worth mentioning that the human exposures to DDT and related compound occur as part of a larger exposure to organochlorine or persistent organic pollutant (POP) mixtures which may alter biological affect."

***RESPONSE:*** *The suggested revision was made:*

> Clear explanations of the inconsistency across the studies are not available, but possible contributors could be small case sample sizes and different distributions of age, BMI, gender, and exposure levels; exposure to varying levels of other persistent organic pollutants (e.g., PCBs, CDDs, etc.) may also influence the study results.

**COMMENT:** Regarding the discussion of Studies of Diabetic Outcomes in Laboratory Animals in Section 2.18, the Reviewer commented "Very good job of succinctly summarizing recent in vivo studies."

***RESPONSE:*** *No response needed.*

**COMMENT:** Regarding the discussion of the La Merrill et al. (2014a, 2014b) study in Section 2.18, the Reviewer commented "Classified as developmental effects in table 2-1 but looks like more than developmental."

***RESPONSE:*** *The effects reported in the La Merrill et al. (2014, 2014b) study were considered developmental effects since they involved perinatal exposure. A statement was added to the Developmental section (2.17) pointing the reader to Sections 2.5 and 2.10 for discussions of the cardiovascular and renal outcome results reported in the La Merrill et al. (2014a, 2014b) study:*

> Numerous adverse developmental outcomes have been reported in the offspring of rodents exposed to DDT/DDE during gestation and/or lactation. Effects include fetotoxicity, alterations in growth, neurodevelopmental toxicity, and impaired development of the reproductive system; these effects are discussed in greater detail below. Some developmental studies have also evaluated cardiovascular (La Merrill et al. 2016), renal (La Merrill et al. 2016), and diabetic (La Merrill et al. 2014a, 2014b) outcomes in the offspring of rats perinatally exposed and evaluated as adults; these effects are considered developmental toxicity and are discussed in Sections 2.5, 2.10, and 2.18, respectively.

**COMMENT:** Regarding the discussion of Mechanistic Information on DDT Influence on Diabetic Outcome in Section 2.18, the Reviewer commented "This section does an excellent job of summarizing the major mechanistic studies."

***RESPONSE:*** *No response needed.*

**COMMENT:** Regarding the statement in Section 2.19 that breast cancer meta-analyses noted that exposure was measured in mature adults and may not reflected earlier exposure, the Reviewer commented "Good this is acknowledged."

***RESPONSE:*** *No response needed.*

**COMMENT:** Regarding the discussion of chronic-duration cancer studies in laboratory animals, the Reviewer commented "Noted species differences."

***RESPONSE:*** *The species differences in carcinogenicity are discussed in in Section 2.19.*

**COMMENT:** Regarding the discussion in Section 2.19 of liver tumors observed in rats, the Reviewer commented "Majority of chronic rat studies show pro-carcinogenic effect in terms of hepatic carcinogenicity. Exposure levels are in excess of anticipated human exposures."

***RESPONSE:*** *A statement was added to the Relevance to Public Health discussion (Section 1.2) to note that doses exceeded anticipated human exposure levels:*

> The liver and lung appear to the primary cancer targets for isomers of DDT, DDE, and DDD in laboratory animals orally exposed to doses which exceed anticipated human exposures.

**COMMENT:** Regarding the statement— Between 4 and 14 days after exposure, the tissue/blood concentration ratio was about 6 for liver and muscle, 35 for skin, and 400 for adipose tissue (Mühlebach

et al. 1991)—in Section 3.1.2, the Reviewer commented "Howell et al 2015 showed extensive partitioning in adipose as opposed to liver and serum up to 21 days following 5 day exposure to DDE with levels increasing in the adipose from day 7 to day 14 and dropping in the serum and liver during this time suggesting distribution to the adipose."

***RESPONSE:*** *The Howell et al. (2015) study was added to this discussion:*

> Similar results were found in a study designed to induce diabetes in high saturated fat-fed mice, administered DDE for 5 days followed by weekly gavage doses of DDE for 13 weeks; the adipose/serum and liver/serum concentration ratios were approximately 950 and 70, respectively (Howell et al. 2015).

**COMMENT:** Regarding the statement—Of most interest would be studies on the isomer, *p,p'*-DDE, detected at the highest concentrations in environmental media, human tissues and fluids, and foods—in Section 6.2 (Acute-Duration MRLs), the Reviewer commented "It stands to reason that this is needed BUT most DDE exposures would most likely be chronic in nature."

***RESPONSE:*** *No response needed.*

**COMMENT:** Regarding the Section 6.2 subsections Absorption, Distribution, Metabolism and Elimination and Comparative Toxicokinetics, the Reviewer commented "Given the associations between DDT/DDE and obesity/type 2 diabetes, toxicokinetic studies in both normal and obese, diabetic models may be advantageous."

***RESPONSE:*** *The following sentence was added to the Absorption, Distribution, Metabolism and Elimination subsection:*

> However, there are limited data to evaluate potential toxicokinetic differences associated with obesity or diabetes. These data would be useful given the associations between DDT/DDE exposure and obesity and DMT2.

**COMMENT:** Regarding the statement— Additional acute oral exposure studies during critical windows of embryonic, fetal, or neonatal neurodevelopment using different isomers (e.g., *p,p'*-DDE) or mixtures or species are needed to better understand the effects of DDT, DDE, and DDD on early life neurodevelopment—in the Children's Susceptibility subsection of Section 6.1, the Reviewer commented "Totally agree."

***RESPONSE:*** *No response needed.*

**COMMENT:** Regarding the Howell on-going study listed in Table 6-1, the Reviewer commented "This study has been concluded."

***RESPONSE:*** *The study was deleted from Table 6-1.*

## Comments provided by Peer Reviewer #3

## General Comments

**COMMENT:** This toxicological profile is thorough, well written, and brings the evidence up to date. I actually regretted not reading Section 6.2 "Identification of Data Needs" first. Doing that would have focused my attention while reading Chapter 2 "Health Effects." I am more familiar with the epidemiological literature than the toxicological literature, so I was impressed by Section 3.1.5 "Physiologically Based Pharmacokinetic (PBPK)/Pharmacodynamic (PD) Models." Clearly, progress is being made to draw information together and make predictions using a modeling approach.

While it is not evident from the long history of epidemiological literature on DDT and DDE, lipid adjustment of Persistent Organic Pollutants in blood is being discontinued. The reason is the publication of an influential article in 2005 by Schisterman et al., which concluded results based on lipid-adjusted concentrations are biased. The toxicological profile tends to rely on lipid-adjusted findings whenever reporting new results such as NHANES data. Certainly the older DDT and DDE epidemiological literature was lipid adjusted, but the authors should note the movement of the field is away from lipid adjustment.

Schisterman, E.F., Whitcomb, B.W., Louis, G.M., Louis, T.A., 2005. Lipid adjustment in the analysis of environmental contaminants and human health risks. Environmental Health Perspectives 113, 853-857.

***RESPONSE:*** *ATSDR disagrees with the Reviewer and notes that Schisterman and colleagues support the use of serum lipid measurements for the determination of reference values for persistent organic chemicals, like DDT/DDE, and in models that use these chemicals to assess causation. To allow for comparisons between monitoring data presented as unadjusted serum concentrations and U.S. background levels, tables with NHANES serum data expressed as the whole weight were added to Section 5.6*

**COMMENT:** Section 1.1 "Overview and U.S. Exposures" – Perhaps the single statement in the toxicological profile that I most object to is on page 1, lines 8-10, "Three countries have reported uses since 2003…" A little bit of research would have shown the Stockholm Convention currently approves DDT for use in 17 countries. These exemptions are primarily for malaria control. The countries exempted are: Botswana, Eritrea, Ethiopia, India, Madagascar, Marshall Islands, Mauritius, Mozambique, Namibia, Senegal, South Africa, Swaziland, Uganda, Venezuela, Yemen, Zambia, and Zimbabwe.

Stockholm Convention, 2018. Acceptable Purposes: DDT (Register of DDT pursuant to paragraph I of part II of annex B of the Stockholm Convention). Available at: http://chm.pops.int/Implementation/Exemptionsandacceptablepurposes/RegistersofAcceptablePurposes/AcceptablePurposesDDT/tabid/456/Default.aspx

That DDT is used in more than three countries is evident in Section 5.2.2 "Import/Export" where it is stated that India is the only exporter of DDT in world and in 2013/2014 exported "…primarily to the nations of Botswana, Mynmar, Namibia, South Africa, and Zimbabwe (UNEP 2015)." While the toxicological profile is primarily intended for US readers, it should where possible address the world-wide public health problem of DDT use as it exists in 2018.

Of importance to the US is the fact DDT was not banned in Mexico until the year 2000. There are many places in the toxicological profile where noting this should be important. On page 321, lines 23-26, it is noted that "exposure to DDT, DDE, and DDD in imported foods is minimized due to FDA enforcement programs. FDA randomly collects and analyzes a wide variety of imported commodities (e.g., coffee, tropical fruits) to determine if pesticide residues are above EPA tolerances. Pesticide tolerances established by EPA apply equally to domestic and imported food (Wessel and Yess 1991)." The authors should try to determine what the EPA pesticide tolerances were for DDT, DDE, and DDD in 1973-2000 that applied to imported food from Mexico. Dietary intake of DDT and its metabolites in the United States during those years could have been higher than they would have been otherwise due to contaminated food from Mexico.

The effect of DDT use in Mexico is also evident in the 1999-2004 NHANES data. In Tables 5-10 and 5-11, *p,p'*-DDT and *p,p'*-DDE concentrations were reported for race/ethnicity categories (Note: Non-Hispanic Whites are missing from Table 5-10). Mexican Americans were summarized as one group, when in fact they should be considered as two: Mexican Americans Born in Mexico, and Mexican Americans Born in the US. A comparison of these two groups is included in Table 2 of Everett et al. (2017a). Those Born in the US had 11.6% with *p,p'*-DDT >14.50 ng/g serum lipids, and those Born in Mexico had 34.4% with *p,p'*-DDT >14.50 ng/g serum lipids. The *p,p'*-DDE data were skewed, therefore both means and the interquartile range (IQR) were reported. The *p,p'*-DDE IQR for Mexican Americans Born in the US was 196.1-784.0 ng/g serum lipids, and the *p,p'*-DDE IQR for Mexican Americans Born in Mexico was 351.2-2050.1 ng/g serum lipids. Statistics for *p,p'*-DDT and *p,p'*-DDE concentrations, that were not lipid adjusted, were also reported in this article.

Everett CJ, Thompson OM, Dismuke CE., 2017a. Exposure to DDT and diabetic nephropathy among Mexican Americans in the 1999-2004 National Health and Nutrition Examination Survey. Environmental Pollution 222:132-137.

A simple, yet informative, summary of the *p,p'*-DDT and *p,p'*-DDE data for Mexican Americans Born in Mexico, that were included in the 1999-2004 NHANES, was reported in Everett et al. (2017b). In a multiple regression analysis (Table 3), immigrants from Mexico who had been in the US for less than 5 years had 1.007 ng/g (not lipid adjusted) more *p,p'*-DDT than immigrants that had been in the US for greater than 30 years. This article also shows that *p,p'*-DDT concentrations in blood declined after DDT use was banned in Mexico. The proportion of the sample elevated (*p,p'*-DDT >0.086 ng/g), and the *p,p'*-DDT concentrations from the multiple regression analysis, were lower in 2003-2004, 3-4 years after DDT use was banned in Mexico, than in 1999-2000.

Everett CJ, Thompson OM, Dismuke CE., 2017b. DDT and DDE concentrations in blood of Mexican Americans exposed to DDT in Mexico: The 1999-2004 National Health and Nutrition Examination Survey. International Journal of Agricultural and Environmental Research 3(4):370-376. Available at: https://www.ijaaer.com

***RESPONSE**: The text in Section 1.1 was revised to delete the statement that three countries reported uses since 2003; the revised statement indicates that a small number of countries continue to use DDT:*

> Under the Stockholm Convention, use of DDT is primarily restricted to controlling vector-borne diseases, such as malaria and leishmaniasis; a small number of countries continue to use DDT for these purposes.

## Specific Comments

**COMMENT:** Page 179, line 30 – While the study by Grindler et al. (2015) covers the period 1999–2008, no DDE measurements on individuals were made in the NHANES 2005-2008 years.

***RESPONSE:*** *The Grindler et al. (2015) paper does not report that there are no NHANES data for the 2005–2008 sampling periods.*

**COMMENT:** Pages 213-223, (Section 2.18 Other Noncancer) "Epidemiology Studies of Diabetic Outcomes in Humans" and Table 2-20. – This section is an excellent review of the literature on associations of DDT, DDE and DDD with diabetes. The authors are to be commended for their attention to detail and their succinct summary.

***RESPONSE:*** *No response needed.*

**COMMENT:** Page 234, lines 13-18 – These equivocal statements about DDT being a human carcinogen can be expanded to include the 2009 "Pine River statement" which concluded DDT and DDE may be associated with breast cancer (Eskenazi et al. 2009).

Eskenazi, B., Chevrier, J., Rosas, L.G., Anderson, H.A., Bornman, M.S., Bouwman, H., Chen, A., Cohn, B.A., de Jager, C., Henshel, D.S., Leipzig, F., Leipzig, J.S., Lorenz, E.C., Snedeker, S.M., Stapleton, D., 2009. The Pine River statement: Human health consequencies of DDT use. Environmental Health Perspectives 117, 1359-1367.

***RESPONSE:*** *Regarding the summary of the cancer classifications presented in Section 2.19, it is ATSDR practice to only report HHS (NTP), EPA, and IARC cancer classifications.*

**COMMENT:** Page 261 (Section 3.2 Children and Other Populations that are Unusually Susceptible) and page 324, lines 14-15. – Evidence of "increasing body burden with increasing age" can be expanded by referring to Tables 2 and 4 in Everett et al. (2017b), (reference given above). The proportion of Mexican Americans Born in Mexico with *p,p'*-DDT in blood >0.086 ng/g was 25.8%, 31.4%, 46.6% and 66.4% in 12-19 year olds, 20-39 year olds, 40-64 year olds, and ≥65 year olds, respectively ($p<0.0001$). In a multiple regression analysis, *p,p'*-DDE in blood increased 0.486 ng/g per year of age ($p<0.0001$).

***RESPONSE:*** *The results of the Everett et al. (2017b) paper was added to Section 5.7. The study was not added to Section 3.2 because this section does not discuss the potential for higher exposure. The following text was added to Section 5.7:*

> A study of Mexican Americans, born in Mexico, found elevated serum levels of *p,p'*-DDT that declined with increasing years in the United States and increased with age (Everett et al. 2017). Significantly higher serum levels were found in Mexican Americans living in the United States for <5 years, as compared to those living in the United States for >30 years. The percentage of individuals with serum *p,p'*-DDT levels >0.086 ng/g were 10.1, 55.8, 29.5, and 4.6% for 12–19, 20–39, 40–64, and ≥65 years of age, respectively. The study also found a decline in serum *p,p'*-DDT serum levels 3–4 years after Mexico banned the use of DDT in 2000.

**COMMENT:** Page 276, line 29 – Is this suppose to read "ΣDDT levels in sea lions…"? Please correct if so.

***RESPONSE:*** *The typographically error in Section 5.1 was corrected to ΣDDT.*

**COMMENT:** Page 279, lines 29-30 – A zero is missing. The statement should read "…at a temperature range of 930-1,210 °C, a residence time of…"

***RESPONSE:*** *The typographically error in Section 5.2.4 was corrected to 930–1,210°C.*

**COMMENT:** Page 284, line 13 – "Latosol soil" is not a term used in the US Soil Taxonomy. Please add the name of the country this study (Sjoeib et al. 1994) was conducted in.

***RESPONSE:*** *The text in Section 5.4.1 was revised to indicate that the studies were conducted in Indonesia:*

> Other studies conducted in Indonesia using a latosol soil (pH 5.7) found that 5.9% of the radioactivity was lost through volatilization during a 6-week incubation at 45°C (Sjoeib et al. 1994).

**COMMENT:** Page 310, lines 1-5 – I believe this statement definitely applies to food from Mexico: "However, with continued use of DDT in other countries, imported foods may continue to contribute small amounts of DDT and DDE to the daily diet of consumers. From 1981 to 1986, the FDA analyzed 13,283 imported agricultural commodities for pesticide residues. No commodities exceeded the EPA tolerance levels for DDT or DDE; however, 3.1% of the samples had detectable levels of DDT or DDE (Hundley et al. 1988)." What the EPA tolerance levels were at that time should be discussed in the toxicological profile.

***RESPONSE:*** *The referenced statement in Section 5.5.4 was deleted from the profile since it is based on analysis of food samples collected over 25 years ago.*

**COMMENT:** While I have not made specific comments about the animal studies presented in Chapter 2. "Health Effects", I have read all the information and concur with the conclusions in Section 6.2 "Identification of Data Needs." I would only add that knowledge of health effects of DDE is of great importance now due to the quantities in the environment and that found in many humans around the world. We are entering an era where body burden of DDT is less and that of DDE is high. We need to know the health effects of DDE in the absence of significant quantities of DDT.

***RESPONSE:*** *No response needed.*