# Report of the *ITER* Review Meeting on Literature Risk Values for Decabromodiphenyl Ether, TCDD, and Acrylamide

**February 15-16, 2011**

***ITER* Review Organized by**
**Toxicology Excellence for Risk Assessment**
**(http://www.tera.org/peer)**

## NOTE

This report was prepared by scientists of *TERA* and reviewed by the panel members. The members of the panel served as individuals on this panel, representing their own personal scientific opinions. They did not represent their companies, agencies, funding organizations, or other entities with which they are associated. Their opinions should not be construed to represent the opinions of their employers or those with whom they are affiliated.

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## Subject Publications and Participants

### 2,3,7,8-tetrachlorodibenzo(p)dioxin (TCDD) (CASRN 1746-01-6)

The manuscript by Ted Simon, Lesa L. Aylward, Christopher R. Kirman, J. Craig Rowlands, and Robert A. Budinsky (2009) entitled, "Estimates of Cancer Potency of 2,3,7,8- tetrachlorodibenzo(*p*)dioxin Using Linear and Nonlinear Dose-Response Modeling and Toxicokinetics," describes the derivation of a nonlinear reference dose for TCDD. Funding for the publication was supplied by the Dow Chemical Company.

The review panel members included Dr. Michael Dourson, Toxicology Excellence for Risk Assessment (TERA); Dr. Terresa Nusair, The Health & Environmental Safety Alliance, Inc. (HESA); Dr. Glenn Talaska, The University of Cincinnati; and Dr. John Christopher of CH2M/Hill, Inc.

### 2,2′,3,3′,4,4′,5,5′,6,6′-Decabromodiphenyl ether (BDE-209) (CASRN 1163-19-5)

The manuscript by Marcia L. Hardy, Marek Banasik, and Todd Stedeford (2009) entitled, "Toxicology and human health assessment of decabromodiphenyl ether," describes the derivation of an oral reference dose for BDE-209. Hardy and Stedeford are employed by Albemarle Corporation, a global specialty chemical manufacturer whose product line includes brominated flame retardants. The authors note in their publication that the views and opinions expressed in this article are those of the authors and not necessarily those of Albemarle Corporation or the Institute of Public Health and Environmental Protection (Netherlands).

The review panel members included Dr. Michael Dourson, Toxicology Excellence for Risk Assessment (TERA); Dr. Terresa Nusair, The Health & Environmental Safety Alliance, Inc. (HESA); Dr. Glenn Talaska, The University of Cincinnati; and Dr. John Christopher of CH2M/Hill, Inc.

### Acrylamide (CASRN 79-06-1)

The manuscript by Michael Dourson, Richard Hertzberg, Bruce Allen, Lynne Haber, Ann Parker, Oliver Kroner, Andy Maier, and Melissa Kohrman entitled (2008), "Evidence-based dose–response assessment for thyroid tumorigenesis from acrylamide," describes the derivation of a linear cancer slope factor as well as a reference dose. Funding for this publication came from Burger King Corporation, Frito-Lay, Inc., H.J. Heinz Company, KFC Corporation, McDonald's Corporation, The Proctor & Gamble Manufacturing Company, The Proctor & Gamble Distributing Company, and Wendy's International, Inc. for the investigation of issues related to the development of this article. The opinions of the authors do not necessarily reflect the opinions of the reviewers or sponsors.

The review panel members included Dr. Michael Dourson, Toxicology Excellence for Risk Assessment (TERA); Dr. Terresa Nusair, The Health & Environmental Safety Alliance, Inc. (HESA); Dr. Glenn Talaska, The University of Cincinnati; and Dr. John Christopher of CH2M/Hill, Inc.

## Background

The purpose of the International Toxicity Estimates for Risk *(ITER)* database is to provide risk assessors and managers with the latest human health risk values from organizations around the world. *ITER* includes chronic human health risk data from the Agency for Toxic Substances and Disease Registry (ATSDR), Health Canada, International Agency for Research on Cancer (IARC) (in progress), National Institute of Public Health and the Environment (RIVM) - The Netherlands, U.S. Environmental Protection Agency (U.S. EPA), and independent parties whose risk values have undergone peer review. However, the peer reviewed literature contains many more risk values that may be of value to risk practitioners. Therefore, TERA developed a process to include these peer-reviewed, "literature-based," values on the *ITER* database. In order to be considered for inclusion on *ITER*, "literature-based" values must meet the following criteria:

- Manuscript that includes derivation of a risk assessment value has been published in a peer-reviewed journal;
- Assessment follows an identified, commonly used methodology (e.g., U.S. EPA, IPCS, Health Canada); and
- The manuscript's acknowledgment clearly states the source of funding for the work, or the authors provide this source of funding at the review meeting for full disclosure to the panel and on *ITER*.

Authors of peer reviewed publications that meet these criteria submit their publications for an additional quality evaluation by a panel of risk experts. TERA staff screens each publication to determine: ( a) if each value was developed using a commonly accepted methodology, and (b) if the resulting risk value is consistent with the types of information *ITER* is designed to include (e.g., chronic human health risk values). The review panel then meets to discuss issues and address the charge questions. The values that the panel deems to be scientifically sound are then loaded on the *ITER* database to make these values more widely available.

## Panel Selection and Conflict of Interest Evaluation

TERA has developed an extensive list of expert scientists interested in serving on our peer review panels. For each *ITER* Review meeting, TERA sends an invitation to all potential panelists asking for volunteers willing to participate in the review meeting process on a pro-bono basis. TERA screens the panel volunteers to ensure that the resulting panel includes the necessary expertise to evaluate the risk values under review and to ensure there are no conflict of interest issues. In the instance that there are more volunteers than needed, TERA adjusts the panel membership and insures a proper balance of expertise. When a TERA value is being reviewed, an outside independent party reviews the panel membership and conflict of interest.

An essential part of an independent expert review is the identification of conflicts of interest and biases that would disqualify a candidate, as well as identification and disclosure of situations which may appear to be a conflict or bias. The purpose for evaluating conflict of interest is to ensure that the public and others can have confidence that the peer reviewers do not have financial or other interests that would

interfere with their ability to carry out their duties objectively. TERA follows the U.S. National Academy of Sciences (NAS) guidance on selection of panel members to create panels that have a balance of scientific viewpoints on the issues to be discussed. As a result, the expert panels have a broad and diverse range of knowledge, experience, and perspective, including diversity of scientific expertise and affiliation. Panel members serve as *individuals*, representing their own personal scientific opinions. They do not serve as representatives of their companies, agencies, funding organizations, or other entities with which they are associated. Their opinions should not be construed to represent the opinions of their employers or those with whom they are affiliated.

For the February 15-16, 2011 meeting, four experts volunteered to serve on the panel:

- Dr. Michael Dourson, Toxicology Excellence for Risk Assessment (TERA),
- Dr. Terresa Nusair, The Health & Environmental Safety Alliance, Inc. (HESA),
- Dr. Glenn Talaska, The University of Cincinnati, and
- Dr. John Christopher, CH2M/Hill, Inc.

TERA asked each candidate to report on his or her financial and other relationships with the authors and sponsors of each risk value by completing a questionnaire. The completed questionnaires were reviewed by TERA staff and discussed further with panel candidates as needed. (See www.tera.org/peer/COI.html for TERA conflict of interest and bias policy and procedures for panelist selection.) TERA determined that the selected panel members have no conflicts of interest and are able to objectively participate in this review. None of the panel members has a financial or other interest that would interfere with his or her abilities to objectively participate on the panel. None of the panel members is employed by the organizations that authored or sponsored the risk values. None of the panel members was involved in the preparation of the risk values. Dr. Dourson was not part of the panel reviewing the value for acrylamide, because that value had been developed by TERA and he is an author of the publication.

## Meeting Procedure

For the ITER Review meetings, the authors provide TERA with a documentation package, including supporting data and analyses, to support their risk value. TERA staff screens each package to ensure completeness. TERA has prepared a standard list of charge questions, which outlines the issues and questions to guide these reviews (see Appendix A). TERA forwards these charge questions to the panel, and panel members have the opportunity to add to the charge if additional questions are needed. TERA distributes the review materials and charge to panel members prior to the meeting. Panel members are given the opportunity to request additional literature as needed and to submit written pre-meeting comments as necessary.

For the February 15-16, 2011 meeting, TERA distributed the review materials and charge to panel members on January 18, 2011.

At the meeting, the authors briefly present their assessment, and then the panel members are given the opportunity to ask clarifying questions. The panel then conducts a thorough, systematic discussion of the

key data and decisions using the charge questions. The panel members are asked to indicate whether or not each risk value should be included on *ITER*. Panel members are also asked to note any substantive points or issues to include in the *ITER* file that they think would be helpful for the *ITER* user to be aware of when considering these values.

Panel comments and conclusions for the February 15-16, 2011 meeting are described for each manuscript in Appendices B, C and D.

## Meeting Report

After the meeting, the panel (assisted by a TERA scientist) compiles its recommendations and summarizes them for inclusion on *ITER*. Appendices B, C, and D provide the summaries of the panel's review and its comment on each of the subject publications that were reviewed on February 15-16, 2011. Also included is additional information that the panel asked the authors to provide. This meeting report serves as a record of the review; it has been reviewed by the panel members for accuracy before it is finalized. These comments are also available in the chemical entry in *ITER* in the quantitative estimate section.

# Appendix A

## Charge Questions for *ITER* Reviews

**1) METHODOLOGY**

- Was an appropriate risk assessment methodology used and applied correctly? Was the methodology applied correctly, and are the conclusions solid based on the work done? Other comments?

**2) ASSESSMENT QUALITY**

- Was a literature search done and fully explained/evaluated? Do the authors discuss alternative modes of action, viewpoints, or existing assessments? Other comments?

**3) CONCLUSIONS**

- Are the publication's conclusions scientifically sound and supported by the data? Do the authors fully explain and support the choice of critical effect, point of departure, and dose-response? Other comments?

**4) VALUE**

- Is this publication of sufficient value to include on *ITER*? Who are the intended users of the derived value, and how do they benefit from this information on *ITER*? Other comments?

**5) OTHER**

- Are there additional issues or comments relevant to the publication's risk value and its conclusions?

# Appendix B

## Decabromodiphenyl Ether (DecaBDE)

CAS 1163-19-5

*ITER* PR-February 2011

**Source Document**

Hardy M.L., Banasik M., and Stedeford T. 2009. Toxicology and Human Health Assessment of Decabromodiphenyl Ether. *Crit. Rev. Toxicol.* 39(S3): 1-44. Available at http://www.ncbi.nlm.nih.gov/pubmed/19874087

**Data Summary**

| | **Key Information/Data** | |
|---|---|---|
| **Chemical Name** | 2,2′,3,3′,4,4′,5,5′,6,6′-decabromodiphenyl ether (BDE-209) | |
| **Risk Value** | 4 mg/kg-day | |
| **Year of Publication** | 2009 | |
| **Point of Departure (POD) (Experimental)** | $BMDL_{10}$ 419 mg/kg-day | |
| **POD (Adjusted)** | $BMDL_{10[HEC]}$ 113 mg/kg-day | |
| **Uncertainty Factors** | $UF_A$ - 10<br>$UF_H$ - 3<br>$UF_S$ - 1<br>$UF_L$ - 1<br>$UF_D$ - 1<br>Composite: 30 | $UF_H$ – 3 for toxicodynamics<br>× 1 for toxicokinetics |
| **Target Organ** | Liver (Hepatocellular degeneration) | |
| **Species** | Rat | |
| **Study** | NTP, 1986 | |
| **Methodology** | U.S. Environmental Protection Agency (US EPA). (2000). Benchmark Dose Technical Guidance Document Draft EPA/630/R-00/001. October 2000. (http://www.epa.gov/nceawww1/pdfs/bmds/BMD-External_10_13_2000.pdf) | |

**Overview of Approach** We evaluated the available pharmacokinetic data and human and animal toxicity data for 2,2′,3,3′,4,4′,5,5′,6,6′- decabromodiphenyl ether (BDE-209) (CASRN 1163-19-5) with the objective of deriving a reference dose (RfD) based on the best available science. The available studies for deriving an RfD were first screened using the Klimisch criteria and further evaluated using the United

States Environmental Protection Agency's general assessment factors for data quality and relevance (i.e., soundness, applicability and utility, clarity and completeness, uncertainty and variability, and evaluation and review). The chronic 2-year dietary feeding study conducted by the United States National Toxicology Program (NTP, 1986, Technical Report Series No. 309) was selected for RfD derivation.

Hepatocellular degeneration in male rats was chosen as the critical endpoint in the development of an RfD. For dose-response characterization, we applied benchmark-dose modeling to animal data and determined a point of departure (the 95% lower confidence limit for a 10% increase in hepatocellular degeneration) of 419 mg/kg-day for oral exposures. Based on the similar pharmacokinetic characteristics of BDE-209 across species, this value was converted to a human equivalence dose of 113 mg/kg-day by applying a dosimetric adjustment factor based on body weight scaling to the ¾ power. An oral RfD of 4 mg/kg-day was calculated by using a composite uncertainty factor of 30, which consisted of 10 for intraspecies uncertainty, 3 for interspecies uncertainty (i.e., 3 for toxicodynamics × 1 for toxicokinetics), and 1 for deficiencies with the database. We consider the RfD to be adequately protective of sensitive subpopulations, including women, their fetuses, children, and people with hepatocellular diseases.

## Results of Review

**Overall assessment:**

Based on the reading and analysis of the information provided, the panel identified their overall recommendation for the proposed *ITER* materials they reviewed as:

☑ Acceptable with comments (as indicated)

**Panel Conclusions and Recommendations**

The panel determined on February 15, 2011 that the risk value derived in *Toxicology and Human Health Assessment of Decabromodiphenyl Ether* by Hardy et al. (2009) should be included on the *ITER* database.

The panel thought the literature was reviewed well and that the publication contains a thorough discussion of alternative viewpoints. All panelists agreed with the authors that the Viberg et al. (2003, 2007) and Rice et al. (2007) studies should not be used for a point of departure, even though these studies raised some uncertainty in some panelists' minds regarding the validity of potential developmental neurotoxicity endpoints. The validity of the developmental

neurotoxicity effects noted in these studies was lessened by the negative findings in a subsequent guideline study by Biesemeier et al. (2010). One panel member felt that given the uncertainty associated with the developmental neurotoxicity issues, an additional uncertainty factor for database might be considered, but others thought an explanation was warranted without the additional uncertainty factor.

After further discussion, the panel's unanimous consensus around the issue of whether developmental effects are the critical effect is that the Viberg et al. (2003, 2007) studies are considered to be hypothesis generating (since they do not follow EPA recommended protocols), that the Rice et al. (2007) study is suggestively-confirming, but that the Biesemeier et al. (2010) is convincingly negative and does not confirm the potential effect. The additional analysis by Goodman (2009) on the statistics of the Rice et al. (2007) work, and the additional opinions of Williams and DeSesso (2010) that the overall dataset does not indicate developmental neurotoxicity, leads to the panel consensus that a database uncertainty factor of 1 is the best overall judgment, but with all of the previous discussions and caveats of the various studies included.

The panel also consulted with Dr. Ray York, a board-certified toxicologist with extensive experience in experimental developmental and reproductive toxicity on several of the effects described in the Rice et al. (2007). Dr. York was able to respond only after the review meeting, but his response was consistent with the panel's consensus on the developmental endpoints.

## Supplemental Information

Based on the panel's recommendation, Hardy et al. provided additional supplemental materials (the table of RfD values listed below) to assist *ITER* users in evaluating the derived RfD.

**Summary of oral reference doses and minimal risk level derived for decabromodiphenyl ether**

| Year | Oral RfD or MRL | | | Reference |
|---|---|---|---|---|
| | Value, mg/kg-d | Basis | Comment | |
| 1987 | 0.01 | Kociba et al. 1987; rat 2 yr | $NOEL_{rat}$ 1 mg/kg-d; highest dose tested; UF | EPA 2008 |
| 2000 | 4 | NTP 1986; rat, mouse 2 yr | $NOAEL_{rat}$ 1120 mg/kg-d liver thromobosis, | NRC 2000 |

| | | | | |
|---|---|---|---|---|
| | | | degeneration males; UF | |
| 2004 | 10* | Hardy et al. 2002; rat prenatal developmental | $NOEL_{rat}$ 1000 mg/kg-d; MRL for intermediate duration exposure ; UF | ATSDR 2004 |
| 2008 | 0.007 | Viberg et al. 2003; neonatal mouse single dose | $NOEL_{mouse}$ = 2 mg/kg; UF | EPA 2008 |
| 2009 | 4 | NTP 1986; rat, mouse 2 yr | $NOAEL_{rat}$ 1120 mg/kg-d liver thromobosis, degeneration males; BMD modeling | Hardy et al. 2009 |

* Minimal Risk Level

# Appendix C

## Acrylamide

CAS 79-06-1

*ITER* PR-February 2011

**Source Document**

Dourson M., Hertzberg R., Allen B., Haber L., Parker A., Kroner O., Maier A., Kohrman M. Evidence-Based Dose–Response Assessment for Thyroid Tumorigenesis from Acrylamide(2008) *Reg. Toxicol. Pharmacol.* 52: 264-289.

**Data Summary**

| | **Key Information/Data** |
|---|---|
| **Chemical Name** | Acrylamide |
| **CASRN** | 79-06-1 |
| **Risk Value** | 3 E-2 mg/kg-day |
| **Year of publication** | 2008 |
| **Cancer Classification** | See below |
| **Target Organ** | Thyroid |
| **Species** | Rat |
| **Study** | Johnson et al. (1986); Friedman et al. (1995) |

**Available Toxicity Literature and Mode-of-Action (MOA) Analysis:**

HAZARD IDENTIFICATION

Acrylamide has not been shown to cause cancer in humans (Marsh et al., 2007; Swaen et al., 2007; Mucci and Adami, 2005). Four long-term experiments in rats were relevant for the assessment of potential risk in humans, and showed that acrylamide can cause tumors. Johnson et al. (1986) published results for two experiments, one in male and one in female rats. Friedman et al. (1995) also published results for two experiments, one in male and one in female rats, using the same strain of rats. Thyroid tumors in rats exposed to acrylamide were observed to be statistically significant in all four experiments; three of these significances were confirmed by a Fisher exact test. Although, scientists have not identified any chemical that has caused thyroid tumors in humans and the rat thyroid is different from the human thyroid in ways that may be significant, conservative risk assessments use these rat tumors unless data suggest otherwise, therefore, they are considered as relevant to humans. The type of thyroid tumors formed in rats is generally recognized as resulting from growth stimulation and/or mutation, and these modes of action also operate in humans. Up to four different kinds of mammary tumors were observed in two of the four experiments, three of these tumors were statistically significantly observed, one of these significances was confirmed by a Fishers exact test. The statistically significant tumors only occurred in females, and tumors developed were not consistent among experiments. In addition, Friedman et al., 1995 questioned the relevance of their experiment's control animals in comparison to historical controls. Multiple modes of action are likely to be occurring with these inconsistently observed mammary tumors and not all of these modes of action are

likely to be relevant to humans. Therefore, we conclude that these mammary tumors were neither consistent nor fully relevant to humans (Maier et al., 2010). We did not consider tumors of the adrenal gland, central nervous system, clitoral gland, oral tissues, pituitary gland, tunica vaginalis, and uterus to be sufficiently consistent among the rat experiments, and/or relevant to humans for a more comprehensive dose-response assessment. This reasoning for discounting the relevance of tumors of the tunica vaginalis in particular is further explained by Haber et al. (2009).

MODE OF ACTION ANALYSIS

Acrylamide is genotoxic, but not directly mutagenic. A principal metabolite of acrylamide, glycidamide, is mutagenic. Acrylamide also causes growth stimulation and oxidative stress, the latter of which can lead to mutations and other genotoxicity. Mutagenicity and genotoxicity from acrylamide exposure have only been seen at doses higher than those that caused tumors in the four experiments mentioned above. Furthermore, tumors precede genotoxicity in the dose scale, and the shapes of dose-response curves for tumors and mutagenicity and genotoxicity are generally much different. Accordingly, it is unlikely that the tumors evoked are solely caused by either mutagenicity or genotoxicity. Unmeasured mutagenicity might be occurring at low doses and might be responsible for some of the low dose tumors. In fact, mutagenicity appears to lead low-dose *only* tumors. The weight of scientific evidence supports growth stimulation as contributing to thyroid tumors. Specifically:

- Khan et al. (1999) showed statistically significant morphological changes in the thyroid consistent with stimulation after acrylamide for 2 or 7 days.
- Lafferty et al. (2004) observed three measures of growth stimulation in the thyroid after acrylamide for 7, 14 & 28 days; DNA labeling was statistically significant by pairwise comparison. Chico-Galdo et al. (2007) results suggest that DNA labeling in Lafferty et al. (2004) was growth stimulation.
- Friedman et al. (1999) showed thyroid hormones to be statistically significantly decreased in males after 28 days of exposure (trend test & pairwise comparison). Females appeared to be affected, but less so.
- Johnson et al. (1986) showed thyroid hyperplasia in both male and female rats after 2 years of exposure (statistically significantly-trend test).

Comparison with EPA (1998) examples also supports this mode of action for acrylamide. Tumors evoked by acrylamide exposure were generally benign, occurred late in life, and were more often in hormonally-active organs, in all four experiments. Such tumor appearance is more consistent with manners of tumor formation that are different from direct mutation. These observations also mean it is unlikely that direct mutations are causing all of the tumors in these experiments. Thus, both a mutagenic and non-mutagenic manners of tumor formation are likely to contribute to thyroid tumors.

**Quantitative Estimate:**

EPA (2005) suggests "decoupling" data when several modes of action occur in different parts of the dose response curve. Thus, a

- Mutagenic, non-threshold, linear mode of action may be occurring at doses of less than 1 mg/kg-day, and
- Growth stimulation, threshold, non-linear mode of action likely dominates at doses in excess of 1 mg/kg-day.

EPA (2005) suggests that selection of a point of departure be close to the lower range of data of interest. Pooling thyroid tumors over 19 dose groups show that 2% extra risk is:

- ~Double the background rate of the pooled data, and
- 1/3 of the highest low dose response rate of ~6%.

Thus, a 2% BMR is comfortably within the interpolation range allowing for a stable estimate for the point of departure. Using EPA (2005) guidelines, we compared different mathematical models in an attempt to fit these "decoupled" data.

- Multistage model did not fit “"decoupled" data well;
- Weibull model fit these "decoupled" data well with fixed power of 2, but EPA software did not allow a positive value for control doses with power unfixed; controls animals had a positive dose.
- Probit model fit these "decoupled" data well, showing a linear response for tumors in the low dose range and a curvilinear upward trend for tumors from growth stimulation in the high dose range;
- When low dose "decoupled" data only were considered, a weighted linear regression and a multistage model both confirmed the use of the probit model to generate a point of departure for the low dose extrapolation.

The probit model Benchmark dose (BMD) of 2% extra risk is 0.81 mg/kg-day, which is associated with a slope of 0.025 $(mg/kg\text{-}day)^{-1}$. Slope value adjusted by 1.2 for known kinetic rat and human differences; not further adjusted for dynamic variability because:

- Williams (1995) states that thyroid tumors in humans do not form in the presence of mutagens if TSH-stimulated growth is prevented;
- EPA (1998) considers an adjustment factor of 1 for chemicals having a growth stimulation mode of action, unless specific data suggest otherwise;
- Allen et al. (1988) showed in published studies of human and animal tumor slope factors that the most likely value for an overall factor is roughly 1-fold;
- Goodman and Wilson (1991) consider the best estimate of the interspecies factor to be log normally distributed around a value of 1.

The adjusted slope factor is 0.030 (mg/kg-day)<sup>-1<sup>.

## Results of Review

**Overall assessment:**

Based on the reading and analysis of the information provided, the panel identified their overall recommendation for the proposed *ITER* materials they reviewed as:

☑ Acceptable with comments (as indicated)

**Panel Conclusions and Recommendations**

The panel determined on February 15, 2011 that the risk value derived in *Evidence-Based Dose–Response Assessment for Thyroid Tumorigenesis from Acrylamide* by Dourson et al. (2008) should be included on the *ITER* database.

The panel agreed that there are multiple modes of action for the thyroid tumors in rats caused by acrylamide, but the relevance of these thyroid tumors to humans was questioned because there are known problems extrapolating from rat to human thyroid (physiologically). Rats may not be an adequate predictor of humans, with estimations of human risk from rats erring on the side of conservatism.

The panel agreed that two different modes of action cause the dose-response curve to have different slopes in the low and high dose regions. Mutagenicity appears to dominate in the low dose region and stimulation of growth appears to dominate in the high dose region. Although the panel agreed with the "decoupling" of the data as suggested by EPA (2005), it saw the effects more working together to evoke the overall tumor response.

As part of the dose-response assessment, the authors modeled control groups at a 0.002 mg/kg-day dose level because acrylamide was found in rat chow at this level. The panel suspected that this did not result in much change to the estimated risk and requested the authors re-run their model using control groups at a zero dose, rather than the 0.002 mg/kg-day. The authors reported that setting control values to zero was only influential in the estimation of a slope value at the 4th digit of precision, indicating very little influence on the outcome from a risk assessment point of view. The authors were not able to run the log-dose-probit model with the controls at zero dose because of the inability of the model to show zero doses on the log-dose scale.

The authors explored several models for the dose-response assessment, including the multistage, probit, and Weibull model, the latter of which EPA (1998) recommends for thyroid tumors. The Weibull and probit models yielded similar results; both of these model results were confirmed by a linear regression on low dose tumors only. The multistage model did not fit the overall data as well. The panel requested that a table or figure comparing the models be provided as supplemental information on *ITER*.

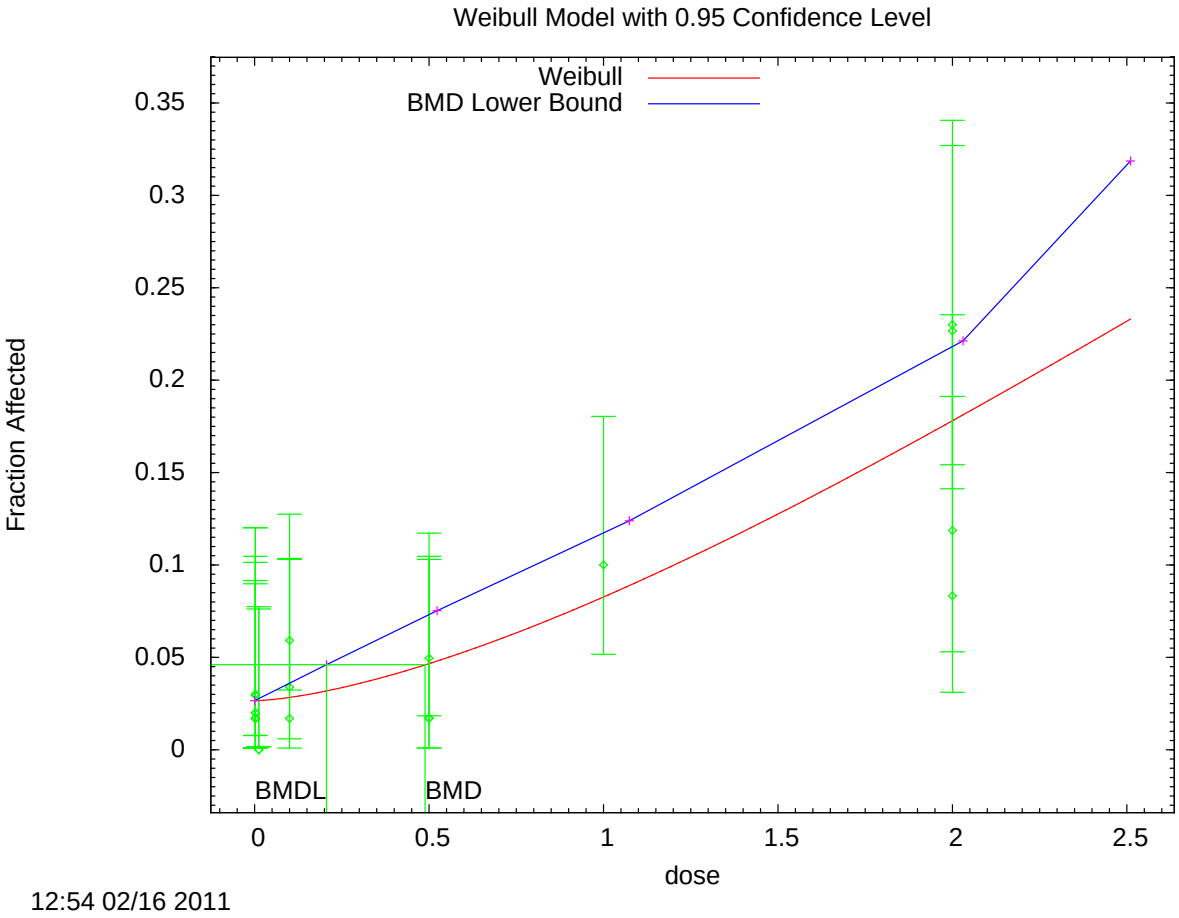
The panel considered that the risk assessment methodology used was appropriate, especially since it was based on EPA (2005) and used EPA BMD software, but while the guidelines recommend decoupling the data given competing MOAs, this has not been applied routinely in previous risk assessments. The panel was concerned that some *ITER* users would find it difficult to fully evaluate the complexities of this assessment and suggested the authors provide additional explanation to enhance transparency and understanding. Specifically, the panel asked the authors to explain how the *ITER* user can interpret this cancer value and compare with other available values, describe the range of margins of exposure for the risk specific dose, and clarify the purpose of the reference dose found in Dourson et al. (2008) and how it might be used. The authors requested additional information so that users do not misuse this value, and it is fully explained when users should choose the cancer slope value over the RfD. For example, the authors could mention that the slope factor is applicable for low dose extrapolation, but that due

to multiple modes of action and other effects taking over at higher dose range. The authors need to specify this range.

## Supplemental Information

Based on the panel's recommendation, Dourson et al. provided additional supplemental materials (Weibull Model run with the control dose at 0.002 to account for background exposure in rat chow vs. model run with control dose at 0.000) to assist *ITER* users in evaluating the derived RfD.

**Weibull Model run with the control dose at 0.002**

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Weibull Model using Weibull Model (Version: 2.15; Date: 10/28/2009)
Input Data File: C:/USEPA/BMDS212/Data/wei_ACR Thyroid Control 0_Opt.(d)
Gnuplot Plotting File: C:/USEPA/BMDS212/Data/wei_ACR Thyroid Control 0_Opt.plt

Wed Feb 16 12:54:33 2011

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BMDS_Model_Run

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The form of the probability function is:

P[response] = background + (1-background)*[1-EXP(-slope*dose^power)]

Dependent variable = response
Independent variable = dose
Power parameter is restricted as power >= 1.000000

Total number of observations = 19
Total number of records with missing values = 0
Maximum number of iterations = 250
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

Default Initial (and Specified) Parameter Values
Background = 0.0192308
Slope = 0.000598375
Power = 8.69999

Asymptotic Correlation Matrix of Parameter Estimates

| | Background | Slope | Power |
|---|---|---|---|
| Background | 1 | -0.61 | 0.56 |
| Slope | -0.61 | 1 | -0.93 |
| Power | 0.56 | -0.93 | 1 |

Parameter Estimates

| | | | 95.0% Wald Confidence Interval | |
|---|---|---|---|---|
| Variable | Estimate | Std. Err. | Lower Conf. Limit | Upper Conf. Limit |
| Background | 0.0265286 | 0.00634709 | 0.0140885 | 0.0389687 |
| Slope | 0.0594013 | 0.0251168 | 0.0101734 | 0.108629 |
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| | | | | |
|---|---|---|---|---|
| Power | 1.5052 | 0.626322 | 0.277627 | 2.73277 |

Analysis of Deviance Table

| Model | Log(likelihood) | # Param's | Deviance | Test d.f. | P-value |
|---|---|---|---|---|---|
| Full model | -301.631 | 19 | | | |
| Fitted model | -314.143 | 3 | 25.0247 | 16 | 0.06939 |
| Reduced model | -349.402 | 1 | 95.5427 | 18 | <.0001 |

AIC: 634.287

Goodness of Fit

| Dose | Est._Prob. | Expected | Observed | Scaled Size | Residual |
|---|---|---|---|---|---|
| 0.0020 | 0.0265 | 1.592 | 1.000 | 60 | -0.476 |
| 0.0020 | 0.0265 | 1.539 | 1.000 | 58 | -0.440 |
| 0.0020 | 0.0265 | 2.653 | 3.000 | 100 | 0.216 |
| 0.0020 | 0.0265 | 1.327 | 1.000 | 50 | -0.287 |
| 0.0020 | 0.0265 | 2.706 | 3.000 | 102 | 0.181 |
| 0.0020 | 0.0265 | 1.327 | 1.000 | 50 | -0.287 |
| 0.0120 | 0.0266 | 1.543 | 0.000 | 58 | -1.259 |
| 0.0120 | 0.0266 | 1.570 | 0.000 | 59 | -1.270 |
| 0.1000 | 0.0283 | 1.672 | 2.000 | 59 | 0.258 |
| 0.1000 | 0.0283 | 1.672 | 1.000 | 59 | -0.527 |
| 0.1000 | 0.0283 | 5.752 | 12.000 | 203 | 2.643 |
| 0.5000 | 0.0467 | 2.755 | 1.000 | 59 | -1.083 |
| 0.5000 | 0.0467 | 2.708 | 1.000 | 58 | -1.063 |
| 0.5000 | 0.0467 | 4.715 | 5.000 | 101 | 0.134 |
| 1.0000 | 0.0827 | 8.267 | 10.000 | 100 | 0.629 |
| 2.0000 | 0.1776 | 10.477 | 7.000 | 59 | -1.185 |
| 2.0000 | 0.1776 | 10.655 | 5.000 | 60 | -1.910 |
| 2.0000 | 0.1776 | 13.319 | 17.000 | 75 | 1.112 |
| 2.0000 | 0.1776 | 17.758 | 23.000 | 100 | 1.372 |

Chi^2 = 22.08 d.f. = 16 P-value = 0.1406

Benchmark Dose Computation

Specified effect = 0.02

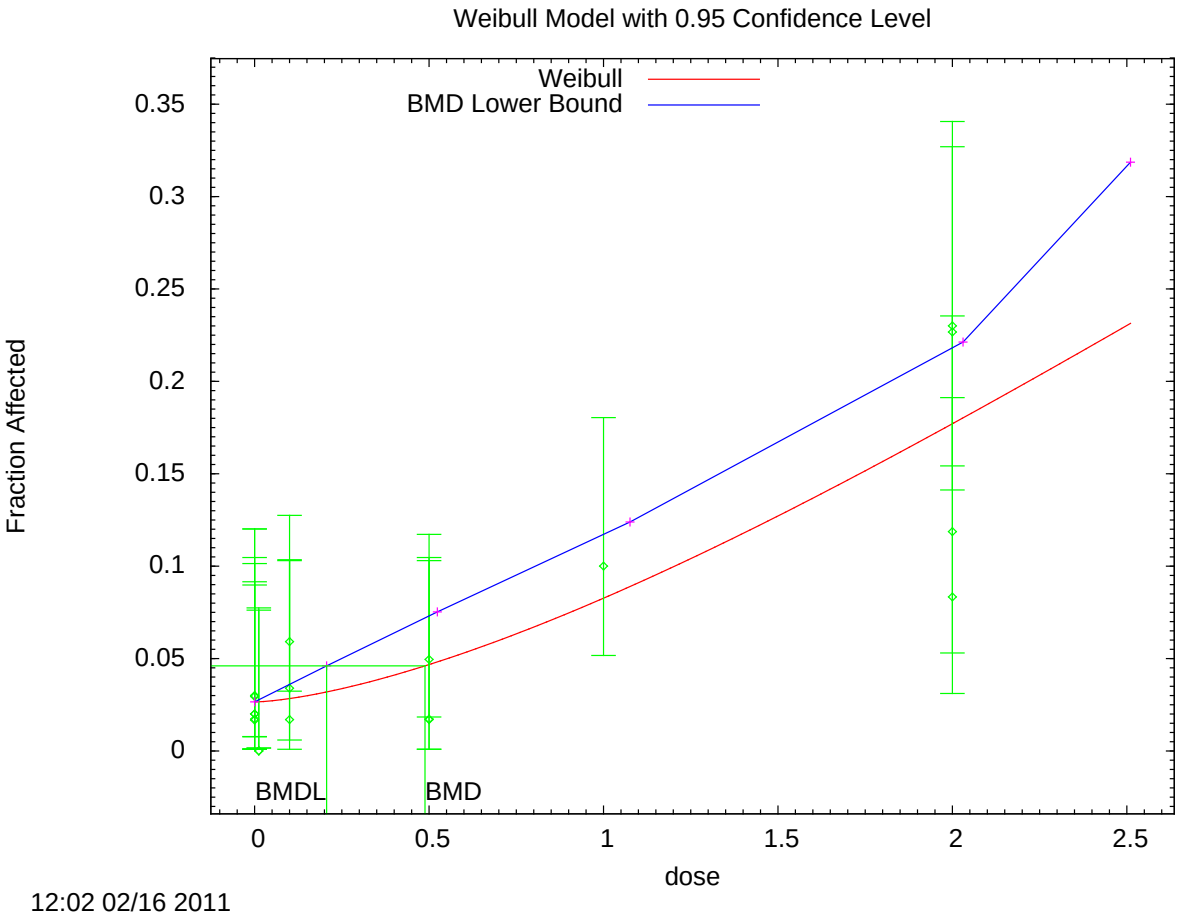
Risk Type = Extra risk

Confidence level = 0.95

BMD = 0.48845

BMDL = 0.205964

**Weibull Model run with the control dose at 0.000**

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Weibull Model using Weibull Model (Version: 2.15; Date: 10/28/2009)
Input Data File: C:/USEPA/BMDS212/Data/wei_ACR Thyroid Control 0_Opt.(d)
Gnuplot Plotting File: C:/USEPA/BMDS212/Data/wei_ACR Thyroid Control 0_Opt.plt

Wed Feb 16 12:02:12 2011

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BMDS_Model_Run

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The form of the probability function is:

P[response] = background + (1-background)*[1-EXP(-slope*dose^power)]

Dependent variable = response
Independent variable = dose
Power parameter is restricted as power >= 1.000000

Total number of observations = 19
Total number of records with missing values = 0
Maximum number of iterations = 250
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

Default Initial (and Specified) Parameter Values
Background = 0.0384615
Slope = 0.114522
Power = 1

Asymptotic Correlation Matrix of Parameter Estimates

| | Background | Slope | Power |
|---|---|---|---|
| Background | 1 | -0.61 | 0.56 |
| Slope | -0.61 | 1 | -0.93 |
| Power | 0.56 | -0.93 | 1 |

Parameter Estimates

| | | | 95.0% Wald Confidence Interval | |
|---|---|---|---|---|
| Variable | Estimate | Std. Err. | Lower Conf. Limit | Upper Conf. Limit |
| Background | 0.0265282 | 0.00634418 | 0.0140939 | 0.0389626 |
| Slope | 0.0594171 | 0.025131 | 0.0101612 | 0.108673 |
| Power | 1.50475 | 0.626701 | 0.276443 | 2.73307 |

Analysis of Deviance Table

| Model | Log(likelihood) | # Param's | Deviance | Test d.f. | P-value |
|---|---|---|---|---|---|
| Full model | -301.631 | 19 | | | |
| Fitted model | -314.143 | 3 | 25.0243 | 16 | 0.0694 |
| Reduced model | -349.402 | 1 | 95.5427 | 18 | <.0001 |

AIC: 634.286

Goodness of Fit

| Dose | Est._Prob. | Expected | Observed | Scaled Size | Residual |
|---|---|---|---|---|---|
| 0.0000 | 0.0265 | 1.592 | 1.000 | 60 | -0.475 |
| 0.0000 | 0.0265 | 1.539 | 1.000 | 58 | -0.440 |
| 0.0000 | 0.0265 | 2.653 | 3.000 | 100 | 0.216 |
| 0.0000 | 0.0265 | 1.326 | 1.000 | 50 | -0.287 |
| 0.0000 | 0.0265 | 2.706 | 3.000 | 102 | 0.181 |
| 0.0000 | 0.0265 | 1.326 | 1.000 | 50 | -0.287 |
| 0.0120 | 0.0266 | 1.543 | 0.000 | 58 | -1.259 |
| 0.0120 | 0.0266 | 1.570 | 0.000 | 59 | -1.270 |
| 0.1000 | 0.0283 | 1.672 | 2.000 | 59 | 0.258 |
| 0.1000 | 0.0283 | 1.672 | 1.000 | 59 | -0.527 |
| 0.1000 | 0.0283 | 5.752 | 12.000 | 203 | 2.643 |
| 0.5000 | 0.0467 | 2.755 | 1.000 | 59 | -1.083 |
| 0.5000 | 0.0467 | 2.709 | 1.000 | 58 | -1.063 |
| 0.5000 | 0.0467 | 4.717 | 5.000 | 101 | 0.134 |
| 1.0000 | 0.0827 | 8.268 | 10.000 | 100 | 0.629 |
| 2.0000 | 0.1776 | 10.477 | 7.000 | 59 | -1.184 |
| 2.0000 | 0.1776 | 10.655 | 5.000 | 60 | -1.910 |
| 2.0000 | 0.1776 | 13.318 | 17.000 | 75 | 1.112 |
| 2.0000 | 0.1776 | 17.758 | 23.000 | 100 | 1.372 |

Chi^2 = 22.08 d.f. = 16 P-value = 0.1407

Benchmark Dose Computation

Specified effect = 0.02

Risk Type = Extra risk

Confidence level = 0.95

BMD = 0.488261 BMDL = 0.206288

Figure 2a. Multistage model fitted to pooled-all thyroid tumor data, showing little change in slope between the low and high dose regions.

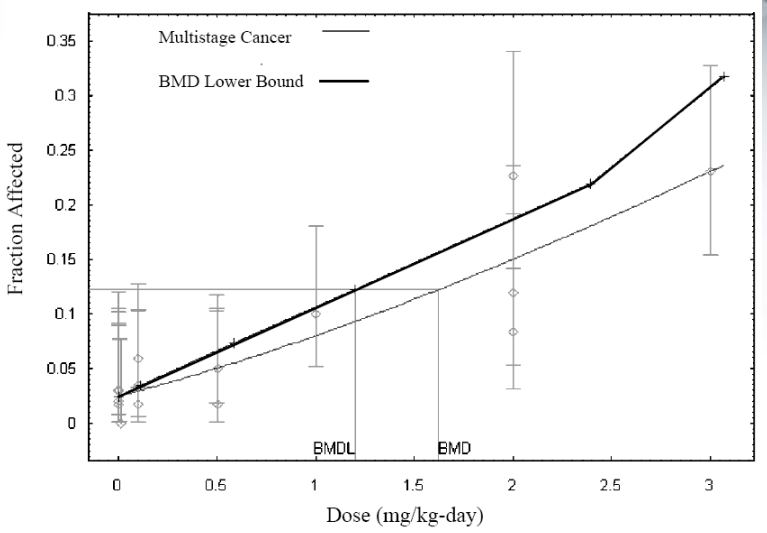
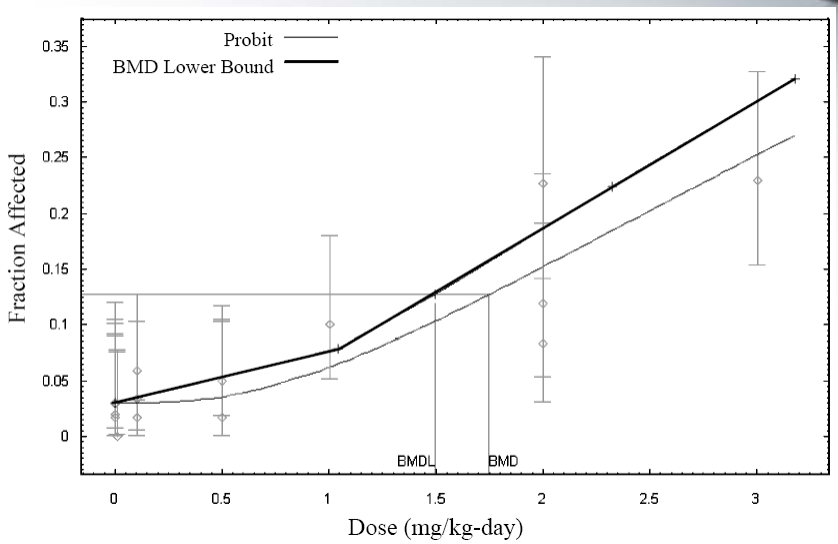



Figure 2b. Probit model fitted to pooled-all thyroid tumor data, showing differing slopes between the low and high dose regions.



## Table 5. Comparison of 2% BMD and BMDL from various models for acrylamide induced thyroid tumors in rats & projected slope factors (SF).

| Data Set | Model | $BMD_{02}$ | SF $BMD_{02}$ | $BMDL_{02}$ | SF $BMDL_{02}$ | BMD/BMDL |
|---|---|---|---|---|---|---|
| All pooled tumors | EPA Multistage | 0.39 | 0.052 | 0.23 | 0.087 | 1.7 |
| Low-dose pooled tumors | EPA Multistage | 0.80 | 0.025 | 0.23 | 0.088 | 3.5 |
| **All pooled tumors** | **EPA Probit** | **0.81** | **0.025** | **0.69** | **0.029** | **1.2** |
| All pooled tumors | EPA Weibull | 0.82 | 0.024 | 0.72 | 0.028 | 1.1 |
| Low-dose pooled tumors | Weighted linear regression | 0.92 | 0.022 | 0.33 | 0.061 | 2.8 |

P-values for all EPA model regressions exceed 0.1 and thus are acceptable; AICs and residuals for the multistage and probit models with all pooled tumors were comparable, however, BMD/BMDL ratios (above) and visual fit differ (see Figure 2a,b). Bold printed row indicates model chosen for extrapolation to humans.

# Appendix D

## 2,3,7,8-Tetrachlorodibenzo(p)dioxin

CAS 1746-01-6
Review Date: February 15-16, 2011

**Source Document**

Simon T., Aylward L.L., Kirman C.R., Rowlands J.C. and Budinsky R.A. Estimates of Cancer Potency of 2,3,7,8-Tetrachlorodibenzo(p)dioxin Using Linear and Nonlinear Dose-Response Modeling and Toxicokinetics (2009) *Toxicol. Sci.* 112(2): 490-506.

**Data Summary**

| | Key Information/Data |
|---|---|
| Chemical Name | 2,3,7,8-tetrachlorodibenzo-(p)-dioxin (TCDD) |
| CASRN | 1746-01-6 |
| Risk Value | 1E-7 mg/kg-day |
| Year of Publication | 2009 |
| Point of Departure (POD) (Experimental) | $BMC_{01}$ 2.61E-3 mg/kg |
| POD (Adjusted) | $BMD_{01[HEC]}$ 1.3E-6 mg/kg-d |
| Uncertainty Factors | Composite: 1 |
| Target Organ | Liver (Combined hepatocellular adenomas and cholangiocarcinomas) |
| Species | Rat |
| Study | NTP, 2006 |

**Potential for Human Carcinogencity:**

Simon *et al*. (2009) developed a human-equivalent reference dose of 1E-7 mg/kg-day for carcinogenic effects of 2,3,7,8-tetrachlorodibenzo-(p)-dioxin (TCDD) based on the occurrence of hepatocellular and biliary tumors in female Sprague Dawley rats observed in a two year bioassay (NTP, 2006). The scientific consensus is that dioxin's effects, including its carcinogenic effects in animals, involve activation of the aryl hydrocarbon receptor (AHR).

In September of 2010, a workshop on *Dose-Response Approaches for Nuclear Receptor-Mediated Modes of Action*. In this workshop, for the first time in an expert panel format, the aryl hydrocarbon receptor (AHR) expert panel rigorously applied the MOA/Human Relevance framework and agreed on an MOA.

The AHR expert workshop panel concluded that sustained AHR activation was a key event in the MOA and that it would be possible to identify NOELs for induction of xenobiotic metabolizing enzymes (XME) and likely other downstream effects. Via AHR activation,

TCDD stimulates the early clonal growth of spontaneously occurring altered hepatic foci and the growth stimulation related to biliary cell proliferation and fibrosis. A late stage event is a collection of histopathological changes referred to as hepatopathy. Hepatopathy is comprised of histological changes that were identified as associative events, including high-dose cytoxicity that leads to regenerative repair, another growth stimulus for spontaneously initiated cells. The workshop panel indicated that hepatocellular adenomas were associated with the occurrence of multinucleated hepatocytes and that cholangiocarcinomas were associated with oval cell hyperplasia.

Although the relevance of rodent liver tumors for human risk assessment may be questionable, these tumors have historically been used by U.S. Environmental Protection Agency (US EPA) as the basis of cancer risk assessment of dioxin-like compounds in humans. Hence, Simon *et al*. (2009) also chose this endpoint.

NTP (2006) was one of a series of two-year cancer bioassays for dioxin-like chemicals in female Harlan Sprague-Dawley rats. These bioassays were conducted using a sophisticated design that incorporated six dose groups, including a stop-exposure group, interim sacrifices, and measures of tissue concentrations and enzyme activity at multiple time points. As indicated, the most prominent responses were remarkable histological changes in the liver grouped under the term hepatopathy, cholangiocarcinoma and hepatocellular adenoma.

**Quantitative Estimate:**

Hepatocellular adenomas and cholangiocarcinomas were combined and the dose-response relationship was assessed based on the lifetime average liver concentration (LALC) estimated with a toxicokinetic model. The same model was used to back-extrapolate the human equivalent-external dose. The best-fitting dose-response model was the dichotomous Hill model. The use of a Hill model is consistent with the underlying biology of receptor-based toxicity, inappropriate gene expression, and dysregulation of homeostasis. The $BMD_{01}$ was expressed as the LALC was 2.61E-3 mg/kg.

<p>Based on the nonlinear mode of action, an interspecies adjustment factor of 0.1 was applied to the $BMD_{01}$. This choice reflects the fact that humans are less sensitive to the activation of the AHR by TCDD (Connor and Aylward, 2006). Following back-extrapolation, the human equivalent dose (HED) was 1.3E-6 mg/kg-d. An intraspecies adjustment factor of 10 was applied to obtain the reference dose of 1E-7 mg/kg-d.

## Results of Review

**Overall assessment:**

Based on the reading and analysis of the information provided, the panel identified their overall recommendation for the proposed *ITER* materials they reviewed as:

☑ Acceptable with comments (as indicated)

**Panel Conclusions and Recommendations**

The panel determined on February 16, 2011 that the risk value derived in *Estimates of Cancer Potency of 2,3,7,8-Tetrachlorodibenzo(p)dioxin Using Linear and Nonlinear Dose-Response Modeling and Toxicokinetics* by Simon et al. (2009) should be included on the *ITER* database.

The panel agreed that the proposed mode of action for the liver tumor endpoint, activation of the AHR receptor and subsequent events, is appropriate basis for the assessment, similar to what others have done, and that the authors, and the supporting study of Conner and Aylward (2006), followed EPA (2005) in the discussion of alternative modes of action, viewpoints, and review of existing assessments.

The authors chose a $BMD_1$ as the point of departure for the tumor endpoint, and not the $BMDL_1$. The choice of a benchmark response of 1% is consistent with EPA (2005) guidelines of between 1 and 10%. One panelist thought that using this best estimate over the lower limit was a good choice, especially since a value of 1% is used as the POD, and lower limits are known to diverge among mathematical models at this level and below.

Some concern was raised by the panel on the choice of critical effect, specifically the developmental NOAEL/LOAELs, which for monkeys are close to the point of departure for rat liver tumors. However, developmental NOAELs for rats are up to 1000-fold higher than liver tumors in rats. The panel agreed with the choice of critical effect (liver tumors in rats), and thought it was more appropriate than the developmental toxicity in monkeys as a basis of the dose-response assessment.

The use of a 0.1 uncertainty factor for rat to human toxicodynamics was discussed extensively. The panel reviewed the IPCS (2005) guidelines on this factor and unanimously agreed that the authors should provide additional supplemental information for *ITER* users. Specifically, they asked the authors to further describe the coefficient of variation (COV) that forms the basis of the rat to human toxicodynamic ratio. The IPCS guidelines state that the COV needs to be 20% or less, in order for the data-derived value of 0.1 (in this case) to be appropriate. If the COV is found to be greater than 20% for the best choice of comparison between the rat and human toxicodynamic metric, then the use of an uncertainty factor less than 1 is appropriate, but that a specific value such as 0.1 cannot be determined with accuracy.

The panel agreed that the choice of other uncertainty factors and dose metric between rats and humans were appropriate.

The panel also commented that this is a highly valuable assessment since there were no other cancer values for this chemical on *ITER* at the time of the review. Overall the assessment demonstrates a good application of EPA (2005) guidance.

## Supplemental Information

Based on the panel's recommendation, Simon provided additional supplemental materials to assist *ITER* users in evaluating the derived risk value.

### Additional Support for the Chemical-Specific Adjustment Factor (CSAF) for Interspecies Toxicodynamic Differences for TCDD
### *ITER* Database Submission

**Introduction**

Simon *et al*. (2009) used a value of 0.1 as the CSAF for interspecies toxicodynamic adjustment, $AD_{AF}$. To provide additional support for this factor, data from Schrenk *et al*., (1995), Xu *et al*. (2000), Silkworth *et al*. (2005) and Budinsky *et al*. (2010) were examined for consistency with WHO-IPCS (2005). Supplemental information from Simon *et al*. (2009) provided a qualitative discussion of this consistency whereas this supplement to *ITER* provides a quantitative estimate.

WHO-IPCS (2005) indicates that to ensure adequacy of the concentration-response data, when dose response curves are not parallel, the preferred point for comparison would be the lowest point on the concentration-response curve that would provide reliable information without extrapolation outside the range of data and suggested the $EC_{10}$ for this purpose. In addition, to ensure the adequacy of the number of subjects/samples, this guidance that the standard error of the mean should be less than 20% of the mean value.

**Data Sources and Methods**

Table 4 in Connor and Aylward (2005) provided the support for the $AD_{AF}$ value of 0.1. This support document examines papers that measure TCDD sensitivity in humans and female Sprague-Dawley (SD) rats. These rats are the same strain, gender, and species used in NTP (2006) upon which the *ITER* toxicity estimate in Simon *et al*. (2009) is based. Hence, the data from Schrenk *et al*. (1991) on Wistar rats were not used.

Schrenk *et al*. (1995) measured ethoxyresorufin-O-deethylase (EROD) activity in primary hepatocytes from 6 humans at concentrations from 0.001 nM to 10 nM. These individual data were digitally extracted from Figure 1 in Schrenk et al. (1995). Xu *et al*. (2000) measured ethoxyresorufin-O-deethylase (EROD) activity in primary hepatocytes from humans and rats at concentrations from 0.001 nM to 1000 nM. Some individual responses were not measured at some of the concentrations. Individual data were digitally extracted from Figure 1

of Xu *et al*. (2000) for 5 humans and 4 rats. Raw data for EROD induction in human primary hepatocytes from four donors at a concentration range of 1E-05 nM to 100 nM were kindly provided by Dr. Jay Silkworth (Silkworth *et al.*, 2005). Donor #3 was a poor responder and these data were not used. Pooled rat data was digitally extracted from Figure 1 in Silkworth *et al.* (2005). Raw data were obtained from Budinsky *et al.* (2010) on EROD induction in 5 female humans and 5 female SD rats at a concentration range of 1E-05 nM to 100 nM.

For each individual rat or human, a least-squares method implemented in MS-Excel was used to obtain Hill function parameters for each individual and the $EC_{10}$ was calculated from those parameters.

**Results**

The individual, mean and standard error (SEM) values for $EC_{10}$ are shown in Table 1 below. The values of $AD_{AF}$ as the ratio of the rat $EC_{10}$ to the human $EC_{10}$ is shown in the last column of Table 1. The $AD_{AF}$ values from individual studies range from about 7 to 75 with an average of 30. The overall AFTD from combining available data on humans and Sprague-Dawley rats is 24.5.

| **Study** | **Number** | **Individual EC10 values** | **EC10 (Mean ± SEM)** | **Standard Error CV 20% criterion** | **Reciprocal of $AF_{TD}$ Value** |
|---|---|---|---|---|---|
| Schrenk et al. (1995) | 6 humans | 0.0052, 0.0101, 0.0027, 0.0095, 0.0106, 0.0207 | 0.0098 ± 0.00253 | 25.8% | 10[1] |
| Xu et al (2000) | 5 humans | 0.0285, 0.0155, 0.0102, 0.0196, 0.0120 | 0.0172 ± 0.00325 | 18.9% | 75 |
| | 4 rats | 3.03E-05, 2.06E-05, 8.18E-04, 4.51E-05 | 0.000229 ± 0.000197 | 86.0% | |
| Silkworth et al (2005) | 3 humans | 0.0412, 0.0926, 0.0215 | 0.0518 ± 0.0212 | 40.9% | 6.7[2] |
| Budinsky et al (2010) | 5 humans | 0.0915, 0.0225, 0.0264, 0.0399, 0.0813 | 0.0523 ± 0.0128 | 24.4% | 26.5 |
| | 5 rats | 0.000820, 0.00557, 0.000456, 0.00167, 0.00135 | 0.00197 ± 0.000825 | 41.8% | |
| **Combined data** | | | | | |

[1] $AD_{AF}$ value presented by Schrenk *et al*. (1995) and is based on $EC_{50}$ values.
[2] $AD_{AF}$ value obtained from digital extraction of combined data in Figure 1 in Silkworth *et a*l. (2005)

| | | | | | |
|---|---|---|---|---|---|
| Humans | N=19 | | $0.0259 \pm 0.00648$ | 21.9% | 24.5 |
| Rats | N=9 | | $0.00120 \pm 0.00058$ | 48.4% | |

Table 1. Compilation of $EC_{10}$ values for EROD induction by TCDD from human and rat primary hepatocytes from four studies.

**Discussion**

Humans appear to be less variable than rats with regard to the $EC_{10}$ for EROD induction by TCDD, as indicated by the standard error criterion in Table 1. With the exception of the data from Silkworth *et al*. (2005), the human data from the other three studies meet or approach the 20% criterion. The combined dataset of 19 individual humans comes very close to the criterion.

It should be noted that this is a "soft" criterion and would be used to determine where in the range of potential values, the value of $AD_{AF}$ would be chosen. The value of 0.1 used in Simon *et al*. (2009) is near the lower end of the range and thus would be considered health-protective. It should also be noted that if the mean value of the $AD_{AF}$ values from the combined individual measurements were used the reference dose for TCDD would closer to 300 pg/kg-d rather than 100 pg/kg-d.

While EROD is commonly used as a marker of aryl hydrocarbon receptor (AHR) activation, the relationship of EROD induction to toxicity is less clear (Carlson *et al*., 2009). In humans, TCDD produces expression changes in a much smaller number of genes than in rats. In fact, the expression changes in only five genes are shared between humans and rats (Rowlands *et al*., 2007). Therefore, the use of EROD induction is very likely itself a health-protective choice.

In summary, the value of the interspecies toxicodynamic CSAF of 0.1 used in Simon *et al*. (2009), indicating humans are ten-fold less sensitive than rats, represents a conservative value within the potential range of values and is also conservative because of the lack of a relationship of EROD induction to toxicity secondary to sustained AHR activation.

**References**

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