EPA's Response to Selected Interagency Comments on the Final Interagency Science Discussion Draft of the IRIS Toxicological Review of 1,4-Dioxane (with Inhalation Update)

September 2013

Purpose: The Integrated Risk Information System (IRIS) assessment development process of May 2009 includes two steps (Step 3 and 6b) where the Executive Office of the President and other federal agencies can comment on draft assessments. Comments on the Final Interagency Science Discussion draft of the IRIS Toxicological Review of 1,4-Dioxane (with Inhalation Update) and IRIS Summary for 1,4-Dioxane (Step 6b) were provided by the Department of Defense (DoD), National Institute for Occupational Safety and Health (NIOSH), Council on Environmental Quality (CEQ), Agency for Toxic Substances and Disease Registry (ATSDR), and jointly by the Office of Management and Budget (OMB) and the Office of Science and Technology Policy (OSTP). A few comments were general questions about IRIS and were not related to how EPA responded to the recommendations on 1,4-dioxane from the public and external peer reviewers. Additionally, a number of comments were editorial in nature and were incorporated where appropriate. The following are EPA's responses to selected interagency comments. All interagency comments were taken into consideration in revising the draft assessment prior to posting on the IRIS database.

For a complete description of the IRIS process, including Interagency Science Discussion, visit the IRIS website at www.epa.gov/iris.

Selected Interagency Science Discussion Comments and Responses:

Topic #1: Toxicological Significance of Nuclear Enlargement – *NIOSH noted the interagency science discussion draft assessment for 1,4-dioxane included statements such as the "toxicological significance of nuclear enlargement is uncertain" and requested that EPA further explain why the toxicological significance is uncertain and clarify that it has been found to be associated with exposure to certain carcinogens and that it is possibly related to early carcinogenic effects. Clawson et al.* (1992) *was provided by NIOSH as a suggested reference.*

> **EPA Response:** After review of the Clawson et al. (1992) reference suggested by NIOSH and additional references, EPA concluded that nuclear enlargement may be found in any cell type responding to microenvironmental stress or undergoing proliferation. It may also be an indicator of exposure to a xenobiotic in that the cells are responding by transcribing mRNA. Several studies indicate that it may also be identified as an early change in response to exposure to a carcinogenic agent (Wiemann et al., 1999; Enzmann et al., 1995; Clawson et al., 1992; Ingram and Grasso, 1987, 1985); however, its relationship to the typical pathological progression from initiated cell to tumor is unclear. Therefore, nuclear enlargement as a specific morphologic diagnosis was not considered an adverse effect of exposure to 1,4-dioxane.

The discussion around nuclear enlargement was updated to reflect this conclusion in the final version of the Toxicological Review.

Topic #2: Metabolism of 1,4-Dioxane – *DoD requested additional consideration regarding metabolic saturation and near complete metabolism of 1,4-dioxane to β-hydroxyethoxy acetic acid (HEAA) at low-level exposures. DoD suggested including the findings from a new peer-reviewed and published paper* (Koissi et al., 2012) *that addresses the stability of 2-dioxanone, a metabolite of 1,4-dioxane. In addition, DoD also suggested that the U.S. Army report titled "Studies on Metabolism of 1,4-Dioxane"* (2010) *be reconsidered and included in the Toxicological Review.*

EPA Response: The information on metabolism of 1,4-dioxane was reconsidered by EPA. It was concluded that metabolic saturation is seen in the single dose studies; however, metabolic saturation is not seen in the chronic exposure studies where metabolic induction was observed instead (Section 3.3). There is insufficient evidence to determine that metabolic saturation occurs at the low-level environmental exposures. Koissi et al. (2012) found that 2-dioxanone is rapidly degraded ($t_{1/2}$ is approximately 2 hr) at physiological conditions (pH = 7.0 and 25ºC). Information on the stability of 2-dioxanone was added to the assessment in Section 3.3. The U.S. Army report (2010) was reconsidered and included in the Toxicological Review (Section 3.3) supporting that HEAA appears to be the major metabolite of 1,4-dioxane and the response to charge question 2 in Section A.3.1 was updated.

Topic #3: Reference Concentration (RfC) Database Uncertainty Factor – *DoD commented that although the peer reviewers generally agreed with the selection of uncertainty factors, EPA should reconsider whether the database uncertainty factor (UF_D) of 3 is warranted since there were several requests for clarification to support this value.*

EPA Response: Four of the six reviewers agreed with the selection and justification of the UFs applied to the point of departure (POD) for the derivation of the RfC. None of the reviewers disagreed with the UF_D of 3; however, one suggested that it be noted that the reproductive toxicity and teratogenicity indices monitored in rats by Giavini et al. (1985) were unremarkable. Two other reviewers agreed with the selection of the UFs but requested clarification of the justification for the database uncertainty factor. EPA revised the text (Sections 5.1.3, 5.2.4, and in response to charge question 5 in Section A.3.2) to clearly state that the reason for a UF_D of 3 is because of the lack of a multigenerational reproductive study (U.S. EPA, 2002).

Topic #4: Mode of Action – *DoD suggested adding a figure for potential modes of action (MOAs) for nasal carcinogenicity, similar to the figure included for liver carcinogenicity. DoD stated that if the MOA is unknown, then EPA should "downgrade" the language describing the evidence. Additionally, DoD commented that EPA inadequately responded to peer review comments regarding the carcinogenic MOA for 1,4-dioxane and the low dose extrapolation approach and that in addition to the linear extrapolation approach presented, EPA should present a nonlinear extrapolation approach and illustrate the data limitations to this approach..*

> **EPA Response:** EPA agreed with DoD that a figure for nasal tumors similar to that for liver tumors would be helpful and added Figure 4-2 to the Toxicological Review that depicts possible MOAs for nasal tumors as a result of exposure to 1,4-dioxane. In agreement with interagency reviewers, EPA modified the MOA language for nasal and liver tumors from "unknown" to state that the evidence is "not conclusive". These changes were made throughout the document, especially in Sections 4.7.3.7, 5.4.3, and 6.2.3.
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> With regards to the cancer extrapolation approach, when EPA evaluates whether the available data provide significant biological support for a mode of action for cancer the goal is to identify key events and to have reasonable confidence in the sequence of events and how they relate to the development of tumors including information on the shape of the dose-response curve at low doses. It is EPA's judgment that there are insufficient data to establish the shape of the exposure-response curve at low doses based on the mode of action data for cancer effects following exposure to 1,4-dioxane, for both oral and inhalation routes of exposure; thus a default linear extrapolation was used. The external peer review panel weighed in on the issue and 5 of the 6 reviewers agreed with the approach that was presented in the Toxicological Review. A nonlinear approach was not added to the assessment.

Topic #5: Benchmark Dose Modeling (BMD) for Cancer Endpoints – *DoD suggested evaluating the inhalation cancer dose-response data using all available BMD models in EPA's BMD Software (BMDS) then selecting the best fitting model, rather than examining the fits of the multistage model first. DoD also commented that the BMDS models used to fit the oral and inhalation data were different and the same model should have been used for consistency. Additionally, DoD commented that a response to a peer review comment implied that the WinBUGS approach had not been peer reviewed and they suggested that it be peer reviewed if necessary.*

> **EPA Response:** In IRIS assessments and as stated in the EPA's BMD Technical Guidance document (2012), of the suite of models available in the BMDS, multistage models are preferred for cancer endpoints and if multistage models do not fit the data, then other BMDS models are explored. For the 1,4-dioxane inhalation assessment, multistage models provided an adequate fit to the cancer data. For the oral assessment,

the multistage models did not adequately fit the cancer data; therefore, the remaining BMDS models were evaluated and the best fitting model was selected. Thus, different models were used for the oral and inhalation datasets. The WinBUGS approach has been peer reviewed and published in the scientific literature (Kopylev et al., 2009; Spiegelhalter et al., 2003). These references are cited in the Toxicological Review and provided in the list of references.

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