Executive Office of the President: Council on Environmental Quality, Office of Information and Regulatory Affairs, and Office of Science and Technology Policy
Comments on the Interagency Science Consultation
Draft IRIS Toxicological Review of Perfluorohexanesulfonic Acid (PFHxS)
January 2023
(Date Received February 7, 2023)

Thank you for the opportunity to review the draft IRIS Toxicology Review of PFHxS. Overall, we found the assessment well organized, logical, and easy to follow. We greatly appreciated the sections of the text that synthesized the PFHxS findings in the context of other PFAS (in particular, the thyroid section). We have some constructive comments and suggestions that we hope will bolster an already well-written evaluation.

General Comments

- Was read across considered for the evidence bases for PFHxS? There are limited studies for this PFAS, but an abundance of PFAS literature more broadly. EPA has inconsistently considered read across PFAS evidence in their assessments…
    - Throughout chapter 1, EPA discusses PFAS more broadly as evidence for the physical/chemical properties of PFHxS. Was that same translation considered for health effects?
- Suggest EPA mention the PFOA and PFOS assessments that the EPA Office of Water is doing, even though they are a different group within EPA. It seems a glaring omission in the executive summary.
- Consistency: it seems that the opposite directions observed in some epidemiological studies for thyroid hormones decreased confidence in those studies. However, it is noted that some sub-analyses for immune endpoints resulted in effects in opposite directions. It is unclear if EPA downgraded this body of evidence for that. These seems an important consideration especially considering the inverse effects were found in the same population. Consider whether it is appropriate to include "unexplained inconsistency" in Table 3-14 under antibody response – factors that decrease certainty.
- Can PFHxS be detected at the level proposed for the RfD?
    - Lowest mean serum value in NHANES was 0.715ug/L; compared to the RfD internal POD of 0.00054ug/L
    - Can current analytical methods detect PFHxS at the level?
- Please include DDEF in the acronym/abbreviation list. In Table 5-5, it makes sense to include the DDEF (ratio of clearance rates between animals and humans) for the studies that used NOAEL/LOAEL. However, it is unclear why DDEFs are included for the immune and developmental endpoints that used BMD. Where did those values come from? Were they applied to serum concentrations in some way?
- Did EPA independently validate the modeling results from Budtz-Jørgensen and Grandjean (2018a)?

- EPA presented amended modeling results for PFDA in summer 2022 based on this same study and endpoint. EPA found that the modeling presented in the paper should be modified slightly based on the BMR, correct? Was a similar approach taken here? if not, why?

- Including neurodevelopmental measurements in studies assessing thyroid function is very important. The lack of these measurements makes interpreting the thyroid effects in rodents difficult. However, taking the rodent data on reproduction, growth, and neurodevelopment together, there is very little evidence that the PFHxS-mediated thyroid hormone decrements in rats or mice resulted in typical adverse outcomes associated with thyroid hormone deficiencies. There were no consistent effects on reproduction parameters, fetal size, pup weight, or postnatal growth. There were no observed effects on neurodevelopment. It would be helpful for EPA to consider this set of evidence in the context of the changes in thyroid hormone levels. What critical data are needed to increase confidence in interpreting the thyroid hormone changes, in addition to co-measurement of neurodevelopment and thyroid hormone levels? This information could be brought forth as a data gap and inform future studies. (Page 3-173, lines 26-27 and pages 5-3 onward)

- Could EPA emphasize that the evidence about hepatic, cardiometabolic, and development (including neurodevelopment) effects is driven by human, not animal evidence? Most of the animal evidence was indeterminate to slight. Epidemiological information about the association of health effects with chemical exposure is given greater weight in hazard identification and risk assessment, thus supporting EPA's conclusions in these domains. (Page 4-1)

Specific sections or pages

- 3.1.5 is difficult to parse. It seems that there are details about various models and then conclusions not to use. It would be helpful to have a summary statement at the start or end of the section to indicate that EPA did not use any of the PBPK models identified. This information may also be better suited to a table rather than text, with the model and then limitations clearly identified (and in accessible language).

- Page 3-15: "The EPA assumed that the ratio of body water in the newborn versus adults (2.4) also applies to the fetus…"

    - The body composition of a newborn is different than a fetus – a newborn at term has much higher fat content than a fetus at pretty much any gestational age, for example. Is there a citation to support assuming that the same ratio can be applied to the fetus as to the newborn?

- 3-57, Line 1 – please clarify that the effects on T4 and T3 were observed in the animal studies.

- 3-92 – In the epidemiological studies, are the endpoints for gestational age or gestation duration calculated from the last menstrual period or based on ultrasound measurement?

- Figures 3-22, 3-24, 3-26 – even zoomed in at 200%, the figures are unreadable. Please consider splitting the figures over two pages so that the text is legible.

- 3-106: In the studies of birth length and head circumference, was birth weight also measured? Birth weight, length, and head circumference can be inter-related. Was birth weight used as a covariate in the analysis?

- 3-113 – line 1 – please change to "some evidence for postnatal growth deficits," "postnatal developmental deficits" could include more than just growth and thus far in the assessment, only postnatal growth has been considered.

- Page 3-114 – decreased AGD and ACD are known to be related to estrogenic or anti-androgenic signaling. However, PFHxS did not elicit significant responses in the endocrine disruptor screening battery for these domains. Could EPA provide a bit of context about the implications of the lack of activity in these reporter assays in light of changes in AGD/ACD? Do the tests lack sensitivity? Is there another reason that could explain the discrepancy (alternative mode of action, etc)?

- Page 3-117 – We are having trouble following this section. What definition did the authors use to define "preterm"? For example, a decrease in GA at birth of 1 week (39 weeks versus 40 weeks gestation) would not qualify as preterm or premature under current clinical guidelines.

- Page 3-120 – This section is a little confusing. If the women were enrolled at 8-16 wk gestation, they would not have had early losses for this pregnancy, correct? It's our understanding that under the study parameters, if women had experienced spontaneous abortion prior to 8 weeks, they would not have been enrolled in the study.

- 3-170:
    - Footnote 13 states that in Ramhoj et al, 2020, the activity boxes recorded *horizontal* activity. This suggests that rearing behaviors (vertical activity) were not included in the measurements. However, on page 3-171, line 7, the assessment states that there were no differences in "motor activity and rearing behavior" in Butenhoff 2009 and Ramhoj 2020. Were the rearing behavior

measurements captured in Butenhoff? It's unclear from the text where rearing behavior was evaluated.
    - What were the results of the radial arm maze from the Ramhoj study? It is the only test of learning and memory that was performed in the animal studies on neurodevelopment. Since there was some evidence of behavioral issues in humans, mentioning the results of the radial arm maze (even if negative) would be helpful.

- Is figure D-1 repeated? Or are there differences in these figures?

- Suggested edits to the following charge question are to solicit more actionable feedback:
  1. The Toxicological Review for PFHxS describes and applies a systematic review protocol for identifying and screening pertinent studies. The protocol is described in brief detail in Section 1.2.1 (*Literature Searching and Screening*) and in full detail in Appendix A (*Systematic Review Protocol for the PFAS IRIS Assessments*). Please comment on whether the literature search strategy and screening criteria for PFHxS reflects a comprehensive review of the available scientific literature on the noncancer and cancer health effects in humans and experimental animals exposed to PFHxS or related saltsand are clearly described. Please identify additional peer-reviewed studies of PFHxS that the assessment should incorporate.

3.2.2. Immune effects

EPA could consider adding some text to the Evidence Integration section to acknowledge that immunosuppression, increased susceptibility to infections, and autoimmunity are mechanistically interconnected via cross-talk between protective and pathological inflammation. Therefore, studies exploring links between PFHxS exposures and autoimmune diseases are needed to further our knowledge of the immune toxicity of this chemical. Additionally, persistent inflammation is one of the driving factors of cellular and tissue damage leading to organ dysfunction. Therefore, a better understanding of immune and inflammatory responses to PFHxS itself, and any subsequent effects on organ-specific inflammation will be important to understand the effects of PFHxS on cardiac, thyroid, hepatic and nervous system function.