American Coatings ASSOCIATION℠

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SUBMITTED VIA ELECTRONIC MAIL

Members of the Scientific Review Panel on Toxic Air Contaminants
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Re: **Review of "p-Chloro-α,α,α-trifluorotoluene (p-Chlorobenzotrifluoride, PCBTF) – Cancer Inhalation Unit Risk Factor – Technical Support Document for Cancer Potency Factors – Appendix B" – Scientific Review Panel Draft – January 2020**

Distinguished Members of the Scientific Review Panel on Toxic Air Contaminants:

The American Coatings Association (ACA) offers the following comments on the Office of Environmental Health Hazard Assessment (OEHHA) draft document, titled "p-Chloro-α,α,α-trifluorotoluene (p-Chlorobenzotrifluoride, (PCBTF) Cancer Inhalation Unit Risk Factor Technical Support Document for Cancer Potency Factors: Appendix B (January 2020).[1] As noted in previous comments, the ACA maintains that it has serious concerns with the draft document and believes that the concerns should be considered carefully by the Scientific Review Panel (SRP). In several key aspects of the draft document, it appears that OEHHA did not use the best available science, failed to evaluate all of the available data, and did not employ generally accepted methods, as discussed in further detail throughout this letter.

Because of the highly technical nature of the OEHHA (2020a) draft document, as well as OEHHA's (2020b) response to ACA's previous comments, it should be noted that the ACA worked closely with consultants from Ramboll US Corporation to review the draft document and prepare these comments.

[1] ACA is a voluntary, nonprofit trade association working to advance the needs of the paint and coatings industry and the professionals who work in it. The organization represents paint and coatings manufacturers, raw materials suppliers, distributors, and technical professionals. ACA's mission includes programs and services that support the coatings industry's commitment to environmental protection, sustainability, product stewardship, health and safety, corporate responsibility, and the advancement of science and technology. Additional information is available on the ACA website, https://www.paint.org.

SUMMARY

In reviewing the OEHHA (2020a) documentation of the recommended IUR for PCBTF as well as OEHHA (2020b) responses to previous ACA comments, ACA requests that the SRP consider the need to revise the draft document because the evaluation contained within it demonstrates that, in key places, OEHHA did not employ the best available science for evaluating the potential carcinogenicity of PCBTF in humans, OEHHA did not fully evaluate and integrate the available data to investigate hypothesized modes of action critical to correctly estimating the potential for carcinogenicity in humans, and OEHHA did not rely on generally accepted methods for the dose-response modeling of selected endpoints from the NTP (2018) study. Specifically, the ACA has the following concerns:

- In the estimation of the Cancer Slope Factor (CSF) or Inhalation Unit Risk (IUR) for PCBTF, OEHHA (2020a) has applied linear low-dose extrapolation, which is grounded in science and policy that are decades old and does not consider the current state of the science or the available science for PCBTF. The extrapolation method is built on the assumption of direct mutagenicity of a chemical with no consideration of repair mechanisms. The best available approaches used today would apply the linear low-dose extrapolation or linear no-threshold approach *only* if a chemical was mutagenic or if data relevant to understanding the mode of action suggested linearity associated with low concentrations of exposure. OEHHA's approach is also inconsistent with conclusions reached by NTP (2018), which found that PCBTF is neither mutagenic nor more generally genotoxic. OEHHA (2020a) itself observed that "All studies of PCBTF mutagenicity have reported negative findings." Further, the PCBTF data relied upon to estimate the IUR fall into two categories that USEPA (2005) indicates will overestimate risk if a linear no-threshold approach is used: use of data from a highly susceptible animal strain (mouse liver tumors) and extrapolation that extends over several orders of magnitude (lowest animal exposure 100 ppm, expected occupational exposure approximately 1 ppm). Instead, OEHHA should have used a nonlinear approach that is consistent with USEPA's (2005) established guidelines.[2] ACA continues to contend that

[2] The existence of a threshold for effects should be welcome news to all stakeholders, including regulators and public health advocates. Even if one accepts OEHHA's assertion that PCBTF poses a risk of cancer to humans, if the risk of those effects only occurs above a certain threshold -- which could possibly be at a level that is above most, if not all, levels of human exposure -- then health protective measures can be clearly identified and communicated to users of the chemical, while also enabling the public to continue receiving the health benefits of reduced ground level ozone that is achieved through industry's use of this chemical as an "exempt" solvent in coatings. Results from available worker studies provide evidence of exposures for which higher than expected rates of the types of cancers observed in animals following exposure to PCBTF were <u>not</u> observed in the workers (Occidental Chemical Corporation 1992). This despite PCBTF exposure having occurred in combination with more than 80 other chemicals and workers potentially having elevated levels of exposure compared to traditional consumers. Currently, there are no viable alternatives available to replace PCBTF where it is used as an exempt solvent. Hence, any regulatory action taken on this chemical must be based on an accurate, carefully calibrated and data-driven assessment of the potential risks to human health, if any. Over-regulating this chemical to avoid

based on the available PCBTF data, as well as the current state of the science, OEHHA's use of linear, low-dose extrapolation is not the best available science and likely overestimated the potential carcinogenic risk of PCBTF to humans. [3] Staying with an outdated, but well-intentioned, assumption is bad science that undermines public trust in agency decision-making while also providing no tangible public health benefit.

- OEHHA (2020a) concluded that the mechanisms by which PCBTF causes tumors are not known. However, for the mouse liver tumors -- the endpoint upon which the recommended IUR is based -- OEHHA gave no consideration to the mode of action discussed by NTP (2018) for these tumors. Moreover, it appears that OEHHA made no attempt to evaluate the available toxicity data relevant to understanding the mode of action. Had OEHHA undertaken such a review, it would have discovered that there is evidence available for PCBTF that are consistent with the mode of action discussed by NTP for rodent liver tumors and that tumors occurring in rodents by this mode of action are <u>not</u> relevant to human health. In addition, the comparison of the dose-response curve and background incidence of these tumors indicates that the mice relied upon for estimating the IUR have a high spontaneous background rate and susceptibility to liver tumors. As such, the mouse liver tumor data should <u>not</u> be used to derive the CSF/IUR. Use of these data likely overestimates the potential for human health risk.

- When estimating the recommended IUR for PCBTF, OEHHA (2020a) did not provide adequate information to fully document the modeling input, output and decisions related to the agency's selection of the model that provided the basis for the IUR. In attempting to replicate the modeling results, Ramboll scientists noted that "best fit to the data" may have not been the sole justification that OEHHA used when selecting a model for IUR estimation. Rather OEHHA appears to have focused on selecting the model that provides the lowest obtainable Point of Departure (POD) (i.e., the point on the dose-response curve selected for extrapolation to lower concentrations), rather than selecting a model that provides the best fit to the data. Selecting best fit to the data is standard practice unless adequate justification is provided, which OEHHA did not provide. However, because OEHHA did not publicly share an adequate amount of information (e.g., modeling input/output), this conclusion is difficult for Ramboll scientists to verify. In addition, OEHHA (2020a) failed to use generally accepted time-to-tumor models to adjust for survival that incorporate all of the animal-specific data provided by the NTP

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an uncertain hazard (i.e., potential health effects in humans) will only bring about the near-certain public health impacts of increased ground level ozone. If the SRP questions this assertion, it should consult with CARB and other air regulators throughout the state.

[3] The ACA continues to assert that the data are insufficient to support listing PCBTF under Proposition 65. As indicated in its letter to Dr. Lauren Zeise, Ph.D., dated September 19, 2019, the association has chosen not to seek judicial review of the listing at this time. OEHHA and the SRP should not interpret the ACA's decision as agreement with the PCBTF listing. As discussed in it comments to the proposed listing, the association believes that the PCBTF listing is inconsistent with the applicable legal and factual requirements for listing. ACA reviewed OEHHA's response to the Association's comments and did not find it persuasive.

(2018) study. These failures may have resulted in the agency over- or under-estimating the potential potency of PCBTF.

DISCUSSION

I. OEHHA Is Not Using the Best Available Science to Derive the CSF/IUR – Specifically, the Agency Relied Upon an Outdated Policy Assumption that Will Overestimate Risk and Is Inconsistent with both the Current State of the Science and the Available Science for PCBTF

In the estimation of the CSF or IUR, OEHHA (2020a) has assumed linear low-dose extrapolation, a default assumption that is not the best available science for assessing the potential carcinogenicity of PCBTF. In short, PCBTF is not mutagenic. Applying linear-low dose extrapolation to a chemical that is not mutagenic is applying outdated scientific principles that have been recognized as such by leading scientists in the field. The agency fails to consider that the available science for PCBTF is inconsistent with the default policy assumption, as well as the scientific issues surrounding the use of this outdated default assumption. Staying with an outdated, but well-intentioned, assumption is bad science that undermines public trust in agency decision-making while also providing no tangible public health benefit. OEHHA must instead use the best available science. Additional details are provided below.

Scientific Issues Surrounding the Linear No Threshold Assumption

The linear-low dose extrapolation approach was originally incorporated into the first Guidelines for Carcinogen Risk Assessment (USEPA 1986) at a time when mechanisms of carcinogenesis were "largely unknown and data were generally limited" (USEPA 1986). This linear no-threshold approach was proposed based on studies of cancer induced by high doses of ionizing radiation at a time when little was known about processes governing the development of cancer. This assumption is premised on exposure to a chemical causing alterations in the DNA (i.e., mutagenicity) that are transmitted to successive cell generations. OEHHA's (2009) Technical Support Document for Cancer Potency Factors, which sets forth the methods OEHHA uses to derive IURs and CSFs, embraces this assumption stating:

> "The procedures used to extrapolate low-dose human cancer risk from animal carcinogenicity data assumed that a carcinogenic change induced in a cell is transmitted to successive generations of cells descendants, and that the initial change in the cell is an alteration (e.g., mutation, rearrangement, etc.) in the cellular DNA. Non-threshold models are used to extrapolate to low dose human cancer risk from animal carcinogenicity data." (Emphasis added.)

Therefore, this approach applied in the estimation of an IUR for PCBTF assumes a change in the cell that could be transmitted (e.g., mutation) and that there is a risk of cancer with any exposure to PCBTF. Science has evolved since this assumption was proposed and it has been challenged repeatedly highlighting the need to consider biological data (e.g., Tubiana et al. 2006; Doss 2014; Vaiserman et al. 2018). Applying linear low dose extrapolation to PCBTF – a substance

that is not mutagenic – ignores the available data and fails to use the best available science when assessing potential carcinogenicity.

Since the development of the USEPA (1986) Guidelines, multiple processes at the molecular, cellular and organism level have been identified that work to prevent transient DNA damage from causing permanent mutations that would result in cancer (e.g. Clewell et al. 2018; Bryce et al. 2010; Gocke and Muller 2009; Johnson et al. 2009; McMullin et al. 2016). Consideration of these processes for multiple chemicals indicate a threshold below which these protective mechanisms would prevent the development of any health effects. Scientific understanding of carcinogenesis has evolved, and the methods that OEHHA uses need to evolve as well.

In the recent proposed rule by the USEPA in 2018 on Strengthening Transparency in Regulatory Science, it is noted, related to the need to increase transparency of the assumptions in underlying dose-response models, that:

> "…there is growing empirical evidence of non-linearity in the concentration-response function for specific pollutants and health effects. The use of default models, without consideration of alternatives or model uncertainty, can obscure the scientific justification for EPA actions."

Further, in a recent review of USEPA's Guidelines for Carcinogen and Non-Cancer Risk Assessment, members of USEPA's Chemical Assessment Advisory Committee of its Science Advisory Board provided a number of comments outlining the lack of scientific support for the linear no-threshold approach as well as the need to consider available mode of action data (USEPA 2019). Therefore, there are two main things that should be considered in determining whether or not to apply a linear no-threshold approach: whether or not a chemical is mutagenic and whether the scientific evidence supports a linear no-threshold approach. OEHHA (2020a) shows that the agency failed to give these considerations adequate weight or failed to consider them at all.

OEHHA continues to rely upon the multistage model to characterize the potential potency of chemicals, even when the original 1986 USEPA Guidelines noted that no single mathematical procedure is recognized as the most appropriate for low-dose extrapolation in carcinogenesis and that models relied upon for low-dose extrapolation should be consistent with the relevant biological evidence on mechanism of action. The available evidence on PCBTF shows that it is neither genotoxic nor mutagenic.

Using the Available PCBTF Data to Inform the Dose-Response Approach

As ACA has noted in previous comments, when evaluating the potential for mutagenicity of PCBTF or for any compound, it is important to understand the differences between mutagenicity and genotoxicity, two terms which are often used interchangeably. Mutagenicity refers to direct damage to DNA that can be heritable or passed on from cell to cell, while genotoxicity covers a broader range of endpoints that are not transmissible from cell to cell or generation to generation. In other words, if a chemical is mutagenic, it is also genotoxic, but a

chemical could be genotoxic without being mutagenic. Assays that measure mutagenicity are also considered measures of genotoxicity; however, all assays that measure genotoxicity are not indicative of mutagenic potential. Examples of assays that are measures of genotoxicity include unscheduled DNA synthesis (UDS), sister chromatid exchanges (SCEs) and DNA strand breaks. While UDS and SCEs are measures of genotoxicity, they are not measures of mutagenicity because the endpoints measured are not transmissible from cell to cell or generation to generation (Preston and Hoffman 2013). These differences need to be kept in mind when evaluating the data that NTP and others have generated in determining the potential mode of action of PCBTF and the relevant ***dose-response modeling approach*** (i.e., whether to use linear, low-dose extrapolation and assume there is no threshold for effects, *or* determine that there is a threshold below which effects are not anticipated).

The available data clearly demonstrate that PCBTF is not mutagenic and are therefore inconsistent with the default assumption of low-dose linearity that OEHHA has applied. Further, OEHHA's approach is inconsistent with conclusions reached by NTP (2018), which state:

> "p-Chloro-α,α,α-trifluorotoluene is nongenotoxic (Ames assay negative, chromosomal aberration assay negative) and may not directly cause mutations and initiate carcinogenesis."

While NTP noted that additional mechanistic studies are needed, NTP (2018) also stated:

> Overall these results suggest that while p-chloro-α,α,α-trifluorotoluene may be capable of inducing chromosomal damage at high levels of inhalation exposure in male mice, the mode of action for the carcinogenicity of p-chloro-α,α,α-trifluorotoluene observed in rats and mice is unlikely to be driven by genotoxicity".

These NTP (2018) conclusions are critical as the results from this study are the only ones relied upon by OEHHA (2020a) for the estimation of an IUR for PCBTF. NTP (2018) also is the authoritative review that initiated the Proposition 65 listing of PCBTF as a potential carcinogen.

OEHHA (2020a) itself observed that "All studies of PCBTF mutagenicity have reported negative findings." In the absence of data supporting mutagenicity, it is inappropriate for OEHHA to use a linear no-threshold approach to derive a CSF/IUR for PCBTF, when the scientific evidence is clearly inconsistent with this assumption. Instead, OEHHA should have used a nonlinear approach, as explained further in the paragraphs below.

When a chemical is not mutagenic – as is the case with PCBTF – the application of non-threshold or linear approaches are inappropriate. This opinion is shared by other authorities such as the United States Environmental Protection Agency. For example, the current USEPA (2005) guidelines indicate that:

> "Linear extrapolation should be used when there are [Mode of Action] MOA data to indicate that the dose-response curve is expected to have a linear component below the [Point of Departure] POD. Agents that are generally considered to be linear in this region include:

- agents that are DNA-reactive and have direct mutagenic activity, or
- agents for which human exposures or body burdens are high and near doses associated with key precursor events in the carcinogenic process, so that background exposures to this and other agents operating through a common mode of action are in the increasing, approximately linear, portion of the dose-response curve."

However, when a chemical is not mutagenic, USEPA (2005) provides guidelines for use of a nonlinear approach. This approach is the same as dose-response assessments conducted for noncancer endpoints, using the POD, which currently serves as the basis for the IUR; however, instead of calculating a slope factor as OEHHA has done, a reference dose or reference concentration is calculated instead, in accordance with USEPA's established practice for developing such values.

In its Response to Comments, OEHHA (2020b) attempts to distance itself from the USEPA (2005) guidance that OEHHA's own guidance and practice has embraced historically. However, OEHHA's stated reasons for this departure are not relevant to ACA's previous comments. OEHHA has responded that, unlike USEPA, the OEHHA cancer methodology does not make a sharp distinction between genotoxicity and mutagenicity. OEHHA's responses focus on language in OEHHA's guidance related to genetic damage in the context of carcinogenic potential, which is strictly considering these endpoints in the context of a qualitative hazard assessment. However, estimating an IUR is a quantitative assessment. A quantitative assessment must consider how mutagenicity or genotoxicity contribute to the mode or mechanism of action and therefore how it informs the approach to be applied in the extrapolation from the observations at high concentrations in the NTP (2018) study to the low dose region of the dose-response curve. In quantitatively estimating the potential for carcinogenicity in humans, it is critical to understand how mutagenic or genotoxic endpoints relate to any observed carcinogenicity in animal studies. OEHHA has not done this critical piece of the assessment. Instead, the agency has relied upon default policy approaches, rather than determine if the available science for PCBTF is inconsistent with these approaches as explained below. Incomplete and outdated science does not instill public confidence, nor does it provide a public health benefit.

OEHHA (2020a) provides a summary of all available genotoxicity data for PCBTF from published and unpublished studies considered by OEHHA. (*See* OEHHA 2020a, Table 4.) The evidence provided in this table demonstrates that the weight of evidence for the genotoxicity and mutagenicity of PCBTF is negative. OEHHA (2020a) itself concluded that "All studies of PCBTF mutagenicity have reported negative findings."

The limited positive evidence (e.g., two studies) summarized in Table 4 has uncertainties related to the association between PCBTF administration and the endpoints observed. In addition, the *in vivo* and *in vitro* assays reported only provide measures of potential genotoxicity, but not mutagenicity. Each measure has serious limitations, as discussed below.

The only positive evidence of *in vivo* genotoxicity (and not mutagenicity) provided in Table 4 of OEHHA (2020a) is micronucleus formation reported in NTP (2018). The increase in the incidence of micronuclei is only reported in male mice at the highest concentration of PCBTF tested (2000 ppm), with no similar increase noted in female mice or in male or female rats tested at similar concentrations. Further, the concentrations at which micronucleus formation was observed did not correspond with the concentrations at which tumors were observed in the NTP (2018) study, suggesting micronuclei are not part of the mode of action for the observed tumors in rodents. Considering the results from this *in vivo* assay, NTP (2018) concluded that genotoxicity is not part of the mode of action for the tumors observed in rodents following PCBTF exposure.

Regarding *in vitro* measures of potential genotoxicity, only two out of twenty entries in Table 4 of the IUR documentation provided evidence of genotoxicity *in vitro* (Benigni et al. 1982; Litton Bionetics 1979). The *in vitro* assays reported in these studies are the UDS assay in human embryonic epithelial cells (Benigni et al. 1982) and the SCE assay conducted in mouse lymphoma cells (Litton Bionetics 1979b). In addition to being nearly forty (40) years old, these assays have other serious limitations.

The Benigni et al. (1982) study was conducted at high concentrations that are not relevant to the question of low-dose linearity or the question of mode of action in the low-dose region. Benigni et al. (1982) reports a significant increase in the incidence of UDS following administration of PCBTF (1, 2 and 10 µl/ml) administered to cells from human skin and muscle explant; however, in OEHHA's (2020b) response to the ACA's comments, the agency concludes that that the concentrations tested in the Benigni et al. (1982) study are much higher concentrations than what would be obtained in the blood of animals exposed to PCBTF in the NTP (2018) studies. An example is provided in OEHHA (2020b) for female rats that demonstrates that following exposure to 50 ppm PCBTF for six hours, blood levels of 6 µg/ml (0.0045 µl/ml) would be expected (Newton et al. 1998). In addition, modeled blood concentrations reported by Knaak et al. (1998) for a 250 ppm exposure to PCBTF for 6 hours were approximately 36 µg/ml (0.027 µl/ml) (Knaak et al. 1998). Therefore, while the Benigni et al. (1982) may provide limited evidence of genotoxicity, it is associated with high concentrations of exposure well above those concentrations where tumors were observed in the NTP (2018) study. Therefore, these results are not relevant to understanding the potential mode or mechanism of action of PCBTF needed to inform low-dose extrapolation.

Further, in responding to ACA's previous comments, OEHHA (2020b) does not discuss the negative results reported by Benigni et al. (1982) for mutagenicity in the Ames assays. Benigni et al. (1982) reported that the lack of mutagenicity observed in the Ames assay they conducted was consistent with a lack of mutagenicity of PCBTF in a separate study (unpublished; University of Trieste) in which Wistar rats were administered 100 mg PCBTF/kg bw/day for three days. This demonstrates no attempt by OEHHA to integrate the results within or across studies in drawing conclusions about both the potential mutagenicity or genotoxicity of PCBTF and therefore the potential mode of action.

The Litton Bionetics (1979) study is an unpublished report that provides the results of a SCE assay conducted in mouse lymphoma cells. While the frequency of SCEs reported is statistically significantly increased compared to the solvent control (DMSO), the frequency following administration of PCBTF is much closer to the solvent control incidences of SCE and much lower than those reported with the positive control (EMS) (Table 1). This would suggest only weak genotoxic potential for PCBTF, at best. In addition, as noted in Preston and Hoffman (2013), the results from both the UDS and SCE *in vitro* assays provide evidence of potential genotoxicity, but not mutagenicity. Furthermore, while OEHHA (2020b) demonstrates that a regression line can be draw through the data, the changes in SCE do not increase in a monotonic fashion with the increases in PCBTF (Table 1). In addition, Litton Bionetics (1979) provides results from the SCE assay in the presence of metabolic activation. The authors characterized the results of the assay with activation as erratic. While three of the five dose levels yielded frequencies that were significantly greater than the solvent control frequency, there were concentrations, including the highest concentration tested, that failed to show any significant effect. The authors considered the results of the assay as positive but noted the lack of a clearly defined dose-response.

Table 1. SCE Frequencies in cells exposed to PCBTF without activation

Treatment	Dose	No. of Chromosomes	No. of SCE's	SCE/Chromosome ±SE	SCE/Cell
Negative Control (Medium)	---	740	151	0.204 ± 0.017	8.16
Solvent Control (DMSO)	0.1 ml	758	200	0.264 ± 0.017	10.55
Positive Control (EMS)	0.5 µl/ml	767	1186	1.546 ± 0.045*	61.85*
Test Compound					
PCBTF	0.0025 µl/ml	756	261	0.345 ± 0.021*	13.81*
PCBTF	0.0050 µl/ml	770	259	0.336 ± 0.021*	13.45*
PCBTF	0.0100 µl/ml	745	338	0.454 ± 0.025*	18.15*
PCBTF	0.0200 µl/ml	779	320	0.411 ± 0.023*	16.43
PCBTF	0.0400 µl/ml	742	357	0.481 ± 0.025*	19.25*

*Significantly greater than solvent control value, $P < 0.01$ (t-test)
Values tested against solvent control

If OEHHA had conducted a review of the PCBTF science in an integrated approach to determine whether the evidence supports a linear no-threshold approach, they would have determined that the NTP (2018) study, that was the focus of the dose-response modeling by OEHHA (2020a), provides evidence of doses where the tumor incidences are not statistically different from controls (see Figure 1). The solid lines in the graph represent doses where a difference from background is achieved. The dots not connected to a line represent doses that are not significantly different from background. It is clear that a range of concentrations can be

observed that are not statistically significantly different from background and for the majority of tumors, it is around 200 mg/kg/day, which may suggest a POD for a nonlinear approach.

In its Response to Comments, OEHHA (2020b) has provided a dose-response curve for the male mouse liver tumor data (top curve in Figure 1) to try and provide evidence of a linear mode of action. However, this curve does not provide information in the low dose region of the curve (below 100 ppm), which is the area of the curve relevant to human exposure (approximately 1 ppm). Therefore, strict reliance on this curve in male mice alone to justify linearity, in the absence of consideration of the available data that informs the potential mode of action for PCBTF, will result in potential overestimation of the risk of cancer in humans. Further, in looking at the dose-response curves (Figure 1), the shape of all the dose-response curves appear consistent, with the exception of the mouse liver tumors (top two curves in the graph) – meaning that the curves for the mouse liver tumors appear to be outliers.

This difference between the curves for the mouse liver tumors and all the other curves requires careful consideration. Mouse liver tumors have a very high background rate, indicating they are spontaneously formed and represent a susceptible species. All of the other dose-response curves considered for modeling by OEHHA (2020a) have a similar very low slope, including the only other endpoint considered in the mouse, Harderian tumors. The observation of very low slopes for the many of the dose-response curve combined with the high concentrations administered to the animals, suggest low potency or potential for carcinogenicity.

Integrating the information on doses where tumor incidences are not significantly different from control values suggest a range of doses which could become the basis for a nonlinear approach using USEPA standard approaches. A POD would be identified as a starting point for the nonlinear approach within this range of concentrations or doses. The ranges of PODs for tumors other than the mouse liver tumors represent estimates of PODs (38 to 459 mg/kg/day) that are consistent with the range of concentrations for which there is no statistically significant increase in tumors. These values could be relied on to apply a nonlinear approach and derive a reference dose or reference concentration in accordance with USEPA's established practice for developing such values.

While a mode of action for mouse liver tumors has been hypothesized and may not be clearly defined, the available science for PCBTF is clearly inconsistent with a default linear no-threshold approach based on no evidence of mutagenicity. USEPA (2005) provides examples of traits that tend to overestimate risk if a linear extrapolation approach is used. These include:

- The slope factor is derived from data on a highly susceptible animal strain.
- Linear extrapolation is used as a default and extends over several orders of magnitude.
- The largest of several slope factors is chosen.

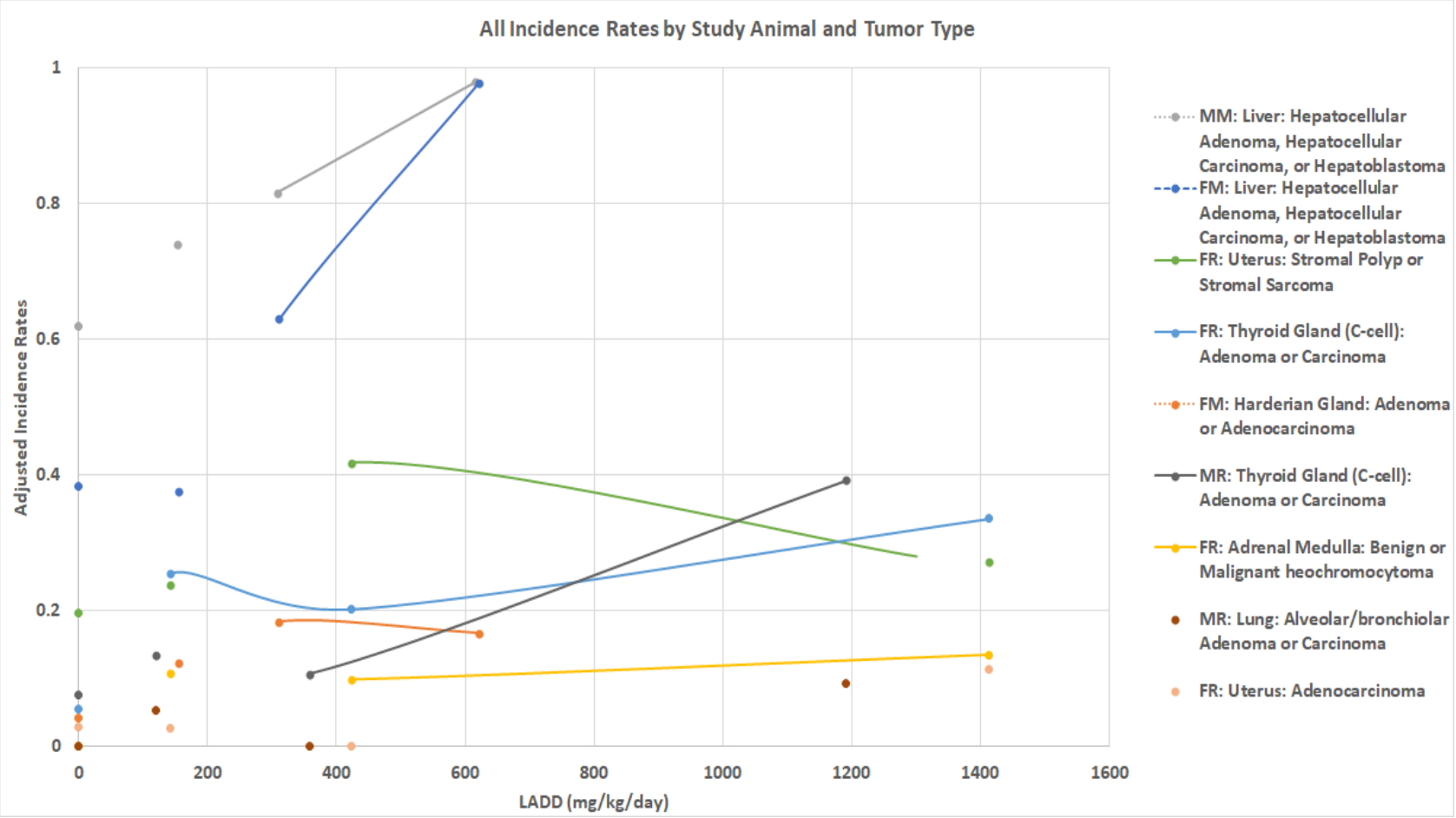
The available data for PCBTF fall into two of these categories. The recommended IUR is derived from data on a highly susceptible animal strain (see discussion below of mouse liver

tumors) and linear extrapolation is used as a default and extends over several orders of magnitude. The lowest concentration tested in male mice (100 ppm), which is the species upon which the recommended IUR is based, is 100 ppm, which is orders of magnitude higher than expected occupational exposures (approximately 1 ppm; Lee et al. 2015), with exposures to the general public expected to be even lower.

While OEHHA (2020a, 2020b) has responded to ACA's previous comments to indicate that rather than "some" evidence there is "limited" evidence of the genotoxicity of PCBTF; it is clear, based on the evidence provided in Table 4 of OEHHA (2020a), there is no evidence that PCBTF is mutagenic. There is, at best, as noted by OEHHA (2020a) evidence from two studies, one of

Figure 1

Dose-Response Relationships from NTP (2018) Considered by OEHHA (2020a) in Estimating the IUR



Solid lines indicate concentrations where incidences are significantly increased compares to concurrent controls.

which (Benigni et al. 1982) OEHHA (2020b) has concluded is not relevant to the expected blood concentrations and therefore the tumors observed in the NTP (2018) study. This leaves one study to provide any evidence of genotoxicity (Litton Bionetics 1979), while all of the remaining evidence is inconsistent with these conclusions. Again, this elevates the importance of NTP's conclusions that PCBTF is not genotoxic or mutagenic and that the carcinogenicity of PCBTF is unlikely to be driven by genotoxicity in determining a dose-response approach for estimating the potential for cancer in humans from exposure to PCBTF. As such, OEHHA should abandon use of its linear, no-threshold approach and instead determine an acceptable exposure level using a nonlinear approach consistent with USEPA's established practice for developing such values. Clearly, the available science for PCBTF is inconsistent with a mode of action that would indicate low-dose linearity. Further, the current state of the science does not support this policy-based assumption and is therefore incorrect and will likely overestimate potential risk to humans.

II. OEHHA Did Not Conduct a Proper Assessment of the Mode of Action Discussed by NTP, which is Supported by Available Data, and OEHHA did not consider the Mouse as a Excessively Sensitive and Susceptible Species and Strain for Liver Tumors.

OEHHA (2020a) concluded that the mechanisms by which PCBTF causes tumors are not known. However, for the mouse liver tumors -- the endpoint upon which the recommended IUR is based – there is nothing to suggest in OEHHA's (2020a) documentation that OEHHA critically reviewed the available data in relationship to the evidence put forth by NTP (2018). The evidence put forth by NTP (2018) is consistent with a constitutive androstane receptor (CAR) mode of action (MOA) for mouse liver tumors (e.g., CYP2B induction, increases in liver weights and nonneoplastic responses). NTP (2018) further noted data in their study that was consistent with a CAR mode of action. While OEHHA (2020b) argued that NTP (2018) did not "propose" this mode of action, the discussion by NTP (2018) of data available to support this mode of action certainly suggests a hypothesized mode of action that warrants investigation. There are methods outlined in the USEPA (2005) guidelines for evaluating hypothesized modes of action for cancer. However, it appears that OEHHA made no attempt to evaluate the available data for PCBTF to follow up on NTP's discussion to determine if any additional data for PCBTF are consistent with NTP's hypothesized mode of action. The absence of a publication in the peer-reviewed literature on this topic does not dismiss the need to review the available data in evaluating the potential relevance of this mode of action to the mouse liver tumors observed in the NTP (2018) study. It would be a component of applying the best available science in evaluating the potential carcinogenicity of PCBTF.

In considering the methods outlined by USEPA (2005) for evaluating a hypothesized mode of action, a weight of evidence evaluation is relied upon in understanding the data to support a hypothesized mode of action. USEPA (2005) provides a framework for evaluating a hypothesized mode of action which includes:

- Description of the hypothesized mode of action
    - Identification of key events

- Discussion of experimental support
    - Strength, consistency specificity of association
    - Dose-response concordance
    - Temporal relationship
    - Biological plausibility and coherence

Further, USEPA (2005) notes that:

- The topics listed for analysis should not be regarded as a checklist of necessary "proofs." The judgment of whether an hypothesized mode of action is supported by available data takes account of the analysis as a whole.
- The framework provides a structure for organizing the facts upon which conclusions as to mode of action rest. The purpose of using the framework is to make analysis transparent and to allow the reader to understand the facts and reasoning behind a conclusion.

If OEHHA had undertaken a review of the PCBTF toxicity and mechanistic data as it related to a CAR mode of action, it would have discovered that there is available evidence from multiple studies relevant to evaluating the mode of action discussed by NTP (2018) for liver tumors in rodents. In addition, it would have found that this mode of action is not relevant to human health. As such, the mouse liver tumor data should not be used to derive the CSF/IUR. A discussion of the available data is set forth below.

In its Response to Comments, OEHHA (2020b) discussed two studies conducted in CAR/RPXR humanized mice to suggest that rodent tumors occurring by the CAR adverse outcome pathway (AOP) may be relevant to human health (Lusier et al. 2014; Braeuning et al. 2014). However, neither or these studies are specific to PCBTF and OEHHA failed to note some of the authors' conclusions regarding the results of these studies: Specifically, the authors challenge the suitability of these animal models for predicting potential risk in humans.

- Luisier et al. (2014) concluded that while humanized CAR mouse models should reflect human transcriptional responses, the data reported in this study suggest that humanized nuclear receptor mice may not be a simple model for extrapolating the risk of rodent tumor findings to humans. Luisier et al. (2014) further acknowledged that based on the weight of evidence of human relevance framework phenobarbital-induced rodent nongenotoxic hepatocarcinogenesis is not considered to be a relevant mechanism for humans and there is no evidence of phenobarbital liver cancer risk in epidemiological data in epileptics.

Further, results from a study by Haines et al. (2018) support that the response of the humanized CAR/PXR mouse differs markedly from that of human hepatocytes and is, therefore, not a suitable animal model for studies on the hepatic effects of nongenotoxic rodent CAR activators. In addition, a study by Ross et al. (2010) in several types of knockout mice, including humanized CAR/PXR mice, provided evidence that human receptors are able to support the chemically

induced hypertrophic responses but not the hyperplastic (cell proliferation) responses, which are necessary for cancer development.

In the discussion of the NTP (2018) study, NTP offers the following conclusions related to the mode of action for mouse liver tumors:

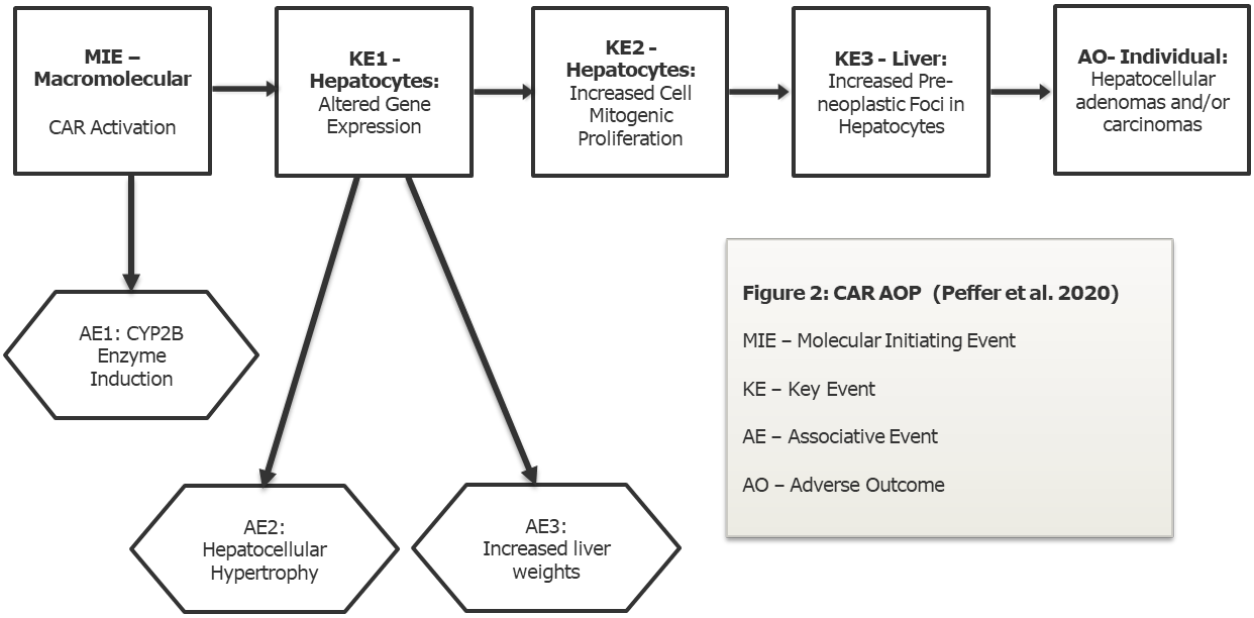
- There is evidence that PCBTF exposure can lead to cytochrome P4502B (CYP2B) induction in the liver of rodents (Pelosi et al. 1998).
- Other cytochrome isoforms evaluated (e.g., cytochrome P4502E) showed higher activity in animals exposed to PCBTF; however, the strongest induction was CYP2B.
- CYP2B activation via the constitutive androstane receptor (CAR) is a known mechanism for tumor promotion activity in the liver of rodents (Sakamoto et al. 2013).
- Liver weights and nonneoplastic lesions observed in the NTP 3-month and 2-year studies are also consistent with a potential CAR-mechanism (Bucher et al. 1994; Parkinson et al. 2006).

Based on NTP's conclusion that the increased incidence of hepatocellular carcinomas reported in male and female mice following inhalation exposure to PCBTF could occur through a potential CAR-mechanism of action (MOA), Ramboll scientists conducted a review of the available results from toxicity studies for PCBTF. NTP (2018) suggested a CAR mode of action for the observed mouse liver tumors based on: (1) the observation of key events for the CAR-MOA including reported increases in CYP2B activity in rats following oral exposure to PCBTF (Pelosi et al. 1998), (2) concentration-related increased liver weights in mice exposed to PCBTF via inhalation for 3 months (NTP 2018), and (3) the consistent evidence from standard *in vitro* assays that PCBTF is not genotoxic (NTP 2018). The key events focused on by NTP (2018) are also consistent with an adverse outcome pathway (AOP) for CAR activation available on the AOP Wiki (Figure 1), which is hosted by the Society for the Advancement of Adverse Outcome Pathways (SAAOP) and endorsed and supported by the US Army Engineer Research & Development Center (ERDC), US Environmental Protection Agency (USEPA), Organisation for Economic Co-operation and Development (OECD), National Toxicology Program (NTP) and European Commission (EC).

The data for PCBTF follow a familiar pattern for other well-known CAR-mediated chemicals, such as phenobarbital. Phenobarbital induced hepatocellular carcinomas in rodents are reported to occur through a CAR-MOA (Holsapple et al. 2006). Phenobarbital has been well-studied and the mode of action for rodent hepatic tumors well established; therefore, potential modes of action of other chemicals are often compared to the evidence for phenobarbital to establish the potential of a CAR-MOA. Holsapple et al. (2006) reports that phenobarbital is the prototype rodent hepatocarcinogen that induces liver tumors through the activation of CAR (a non-genotoxic mechanism) with associated key events that include increased cell proliferation, inhibition of apoptosis, hypertrophy, and the development of altered hepatic foci (Holsapple et al. 2006). The authors conclude that for compounds for which the data are consistent with a phenobarbital-like or CAR-MOA, the carcinogenic response is not relevant to humans. Evaluations for other compounds have concluded that rodent hepatocellular tumors

occurring by the CAR-MOA are considered not relevant to human health (Elcombe et al. 2014; Yamamoto et al. 2004; Holsapple et al. 2006; Yamada et al. 2009).

The results from Ramboll's review of the toxicity data for PCBTF provide evidence of dose-response relationships (both oral and inhalation) between PCBTF and multiple key events and associative events in an established adverse outcome pathway for CAR-MOA (Figure 2) or the induction of hepatocellular adenomas and carcinomas in rodents (Peffer et al. 2020). These key events and associative events are also consistent with the proposed AOP for CAR (Peffer et a. 2020) and those associated with phenobarbital-induced liver tumors in rodents (Holsapple et al. 2006; Elcombe et al. 2014; Yamamoto et al. 2004; Numazawa et al. 2005; Yoshiniari et al. 2001; Waxman and Azaroff 1992), all of which are not relevant to human health.



In addition to the Key Events (KE) associated with the CAR MOA, Associative events (AE), defined as biological processes that are not necessary for the AOP but can be used as surrogate markers for a particular KE, especially when a particulate KE is difficult to measure, have also been identified for the CAR MOA (Peffer et al. 2020). The AEs that follow CAR activation include:

1) Increased CYP2B or CYP3A enzyme activity and/or protein in hepatocytes

2) Increased hepatocellular hypertrophy

3) Increased liver weight.

In drawing their conclusions regarding the potential MOA associated with PCBTF liver tumors, NTP (2018) noted the observation of key events similar to those for the CAR MOA

including reported increases in CYP2B activity in rats following oral exposure to PCBTF (Pelosi et al. 1998), concentration-related increases in liver weights in mice exposed to PCBTF via inhalation for 3 months (NTP 2018) and the consistent evidence from standard in vitro assays that PCBTF is not mutagenic (NTP 2018). Table 2 provides a summary the available evidence for PCBTF that supports a CAR MOA. The table demonstrates evidence over a wide range of doses, as well as over multiple time points. While evidence is lacking for one of the midstream key events, evidence is available to support initiating events, as well as the adverse outcome; therefore, evidence is not necessary for every key event to establish consistency with a known mode of action, such as CAR activation.

For the initiating event, increased CYP2B induction associated with CAR activation has been reported in rats following PCBTF exposure (Pelosi et al. 1998) and is an associative event that provides evidence of CAR activation. Pelosi et al. (1998) exposed Sprague-Dawley rats to PCBTF concentrations of 0, 10, 50 or 250 ppm for 6 hours per day for 90 days and reported a significant increase in CYP2B and liver weight in female rats exposed to 250 ppm. No data was identified in the literature regarding CYP2B induction in mice following PCBTF exposure; however, other rodent hepatocarcinogens with known CAR MOAs (e.g., phenobarbitol and metofluthrin) are inducers of CYP2B enzymes in both rats and mice (Lake 2018; Elcombe et al. 2014) and that endpoint alone has been used to establish evidence of this initial key event. Therefore, the molecular initiating event (MIE), CAR activation, is supported by evidence of the associative event, induction of CYP2B.

Similar evidence has been used to establish CAR activation of other compounds (e.g. metofluthrin; Yamada et al. 2009; Deguchi et al. 2009) in rodents. For KE1, altered gene expression, increased incidences of hepatocellular hypertrophy and increased liver weights are considered associative events as outlined by the AOP. Table 2 presents the data from multiple studies that report increases in both the incidence of hepatocellular hypertrophy and liver weights in both rats and mice following exposure to various concentrations at various exposure durations of PCBTF (NTP 1992, 2018; Newton et al. 1998, Yuan et al. 1992, Macri et al. 1987). The table presents evidence of dose-response, as well as temporal, relationships with increasing exposure to PCBTF in rodents. Significant increases in preneoplastic hepatic foci (KE3) were reported in both mice and rats exposed to concentrations of 100 ppm and greater for 2 years (NTP 2018). Finally, significant increases in the adverse outcome (AO), hepatocellular tumors, were noted in mice, but not rats, exposed to 100 ppm and greater for 2 years (NTP 2018). This may be due to the susceptibility of this mouse strain to the development of liver tumors, which has a high incidence (males – 62%; females – 36%) in concurrent control animals.

Table 2. PCBTF Evidence Relevant to Hypothesized Mouse Liver Tumor Mode of Action			
Key Events	**10 -100 ppm**	**125 -500 ppm**	**1000 -2000 ppm**
MIE: Constitutive androstane receptor (CAR) Activation (Associative Event: CYP2B induction)	✓ 4 weeks	✓ 4 weeks	
KE1: Altered gene expression specific to CAT activation (Associative Events: Increased incidence of liver hypertrophy/increased liver weights)	✓ 14 days 4 weeks 13 weeks 2 years	✓ 14 days 4 weeks 3 months 13 weeks 2 years	✓ 28 days 3 months 2 years
KE2: Increased cell mitogenic proliferation			
KE3: Increased pre-neoplastic foci in hepatocytes	✓ 2 years	✓ 2 years	✓ 2 years
AO: Hepatocellular adenomas, carcinomas	✓ 2 years	✓ 2 years	✓ 2 years

References: Macri et al. 1987; NTP 1992, NTP 2018; Newton et al. 1998; Pelosi et al. 1998; Yuan et al. 1992

Even if OEHHA determines that there is not enough evidence to support a CAR mode of action for PCBTF, liver tumors in rodents in general have been questioned as to whether they are relevant to human health. While typical standardized carcinogenicity testing methods using both rats and mice to predict carcinogenicity in humans have been utilized for decades, it is imperative to consider the interspecies differences among humans and rodents when animal data are used to estimate carcinogenic risk in humans. B6C3F1 mice, which are the strain used in the NTP (2018) study, have been noted as a highly susceptible strain of mice, with males more susceptible than female mice (Lake 2018). Figure 1, above, demonstrates the clear difference in dose-response curves for the mouse liver tumors, compared to all other endpoints, including female mouse harderian gland tumors, considered by OEHHA (2020a) in the estimation of the IUR. This significant difference should give OEHHA and the Scientific Review Panel pause – the mouse data is an outlier of questionable utility.

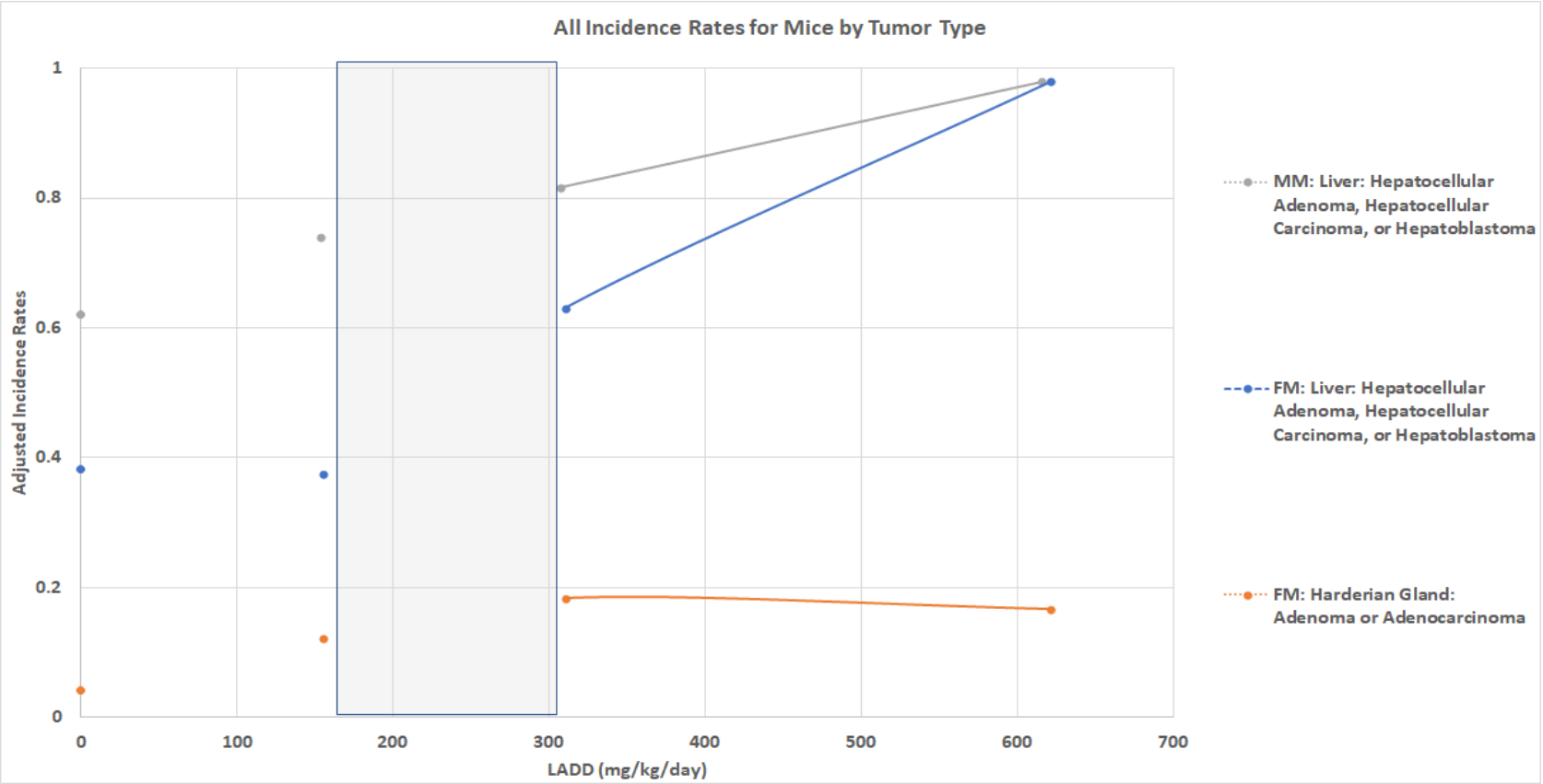
Use of animal carcinogenic data to estimate risk in humans presumes that the process of carcinogenesis in mice and rats is similar enough to precisely and reliably predict outcomes in humans. When this presumption might not hold, the risk assessor must proceed with caution. The liver is the most frequent site for cancer following exposure to mutagenic or nonmutagenic chemicals in mice and rats (Grisham 1996). Further, OEHHA (2009) discusses mouse liver tumors as an example of an endpoint that is a common site for spontaneous tumors and an endpoint that is relatively sensitive to chemical carcinogenesis, while the human liver is more resistant. Hepatocellular carcinoma in humans is related to multiple factors and is predominately linked to chronic hepatitis infections, aflatoxin B exposures, ethanol abuse and tobacco smoking.

The contrasting origins of hepatocarcinogenesis in rodents and humans suggests that the results of carcinogenicity testing in rats and mice may not accurately predict human hepatocarcinogenesis and may overpredict the potential for carcinogenicity.

When Ramboll scientists looked closer at the dose-response curves for the mouse liver tumors, they discovered that there are concentrations for the incidences of liver tumors that are not statistically significantly different from incidences in the concurrent control animals (Figure 3). In Figure 3, the points of the dose-response curve lacking a connecting line represent concentrations for which the tumor incidence was not statistically significantly different from concurrent controls. The range of doses noted in gray in Figure 3 represent a range of doses between doses that have a lack of and observance of statistical significance of mouse liver tumors. The recommended PODs (BMDLs) for mouse liver tumors reported by OEHHA (2020a) are all below this range. When applying standard approaches outlined in USEPA (2005), the lowest value could be considered as the POD for extrapolation. In considering the lack of mutagenicity of PCBTF combined with the susceptibility of the mice in the NTP (2018) study to liver tumors, this provides support for the use of the currently recommended PODs as a starting point for nonlinear extrapolation to an acceptable level of exposure, rather than assuming linearity.

Figure 3

Dose-Response Relationships for Mouse Liver Tumors Considered by OEHHA (2020a) for the Estimation of the IUR



The Gray area indicates a range of doses between dose groups where there is a lack of statistical significance to the doses where statistical significance is noted.

OEHHA's decision to rely on the male mouse liver tumors reported in the NTP (2018) study to establish the potential for carcinogenicity in humans is not based on a critical review of the available science for PCBTF. As demonstrated in Table 2, there is evidence to support an evaluation of the PCBTF data as it relates to a CAR MOA, and as discussed by NTP (2018), the available science for PCBTF is consistent with a mode of action (CAR activation) for male mice liver tumors (the endpoint relied upon for the OEHHA recommended IUR). Further, tumors occurring by this mode of action in rodents are not relevant to human health. In addition, the strain of mice in the NTP (2018) study have a high background rate of liver tumors, demonstrating a high spontaneous incidence and susceptibility, making reliance on this endpoint of questionable validity for extrapolation to humans. As such, OEHHA should either abandon use of the mouse liver tumor data when developing the CSF/IUR or conduct a thorough analysis of the available data to evaluate the CAR mode of action, as well as the relevance of the mouse liver tumor data in general to human health. OEHHA should not proceed any further with the draft CSF/IUR without making these changes.

III. The Dose-Response Modeling Conducted by OEHHA Lacks Transparency and Did Not Rely on Generally Accepted Methods Using All of the Available Data and Results.

When estimating the recommended IUR for PCBTF, OEHHA (2020a) does not appear to have relied upon generally accepted methods for selecting a dose-response model. However, it is difficult to evaluate because OEHHA (2020a) failed to report all of the information needed to enable the public to adequately assess the selection of the models that OEHHA applied to the data. In addition, in attempting to replicate the modeling results in OEHHA (2020a), the agency appears to have made decisions based on selecting the lower BMDL value, rather than the best fit of a model to the data. The agency also failed to use generally accepted time-to-tumor models to adjust for survival. These failures may have resulted in the agency over- or under-estimating the potential potency of PCBTF.

When selecting a dose-response model, OEHHA (2020a) appears to have used methods taken from a 2014 draft operating procedure for USEPA subcontractors that has not been finalized. While the website providing the guidance indicates that the guidance has been reviewed in accordance with USEPA policy and approved for publication, it is still marked as "Draft" suggesting it has not been finalized. Moreover, these methods provided in the guidance are inconsistent with those found in USEPA's well-established final BMDS Guidance (2012), as well as the OEHHA (2009) Technical Support Document. As noted previously, for detailed methods on dose-response, OEHHA (2009) defers to USEPA (2005) Guidelines for Carcinogen Risk Assessment.

In selecting the model for estimation of the IUR, this draft operating procedure (USEPA 2014) was cited by and relied on by OEHHA (2020a) to choose the number of stages for cancer modeling. The approaches in that draft document are inconsistent with the well-established finalized USEPA (2012) BMDS Guidance which has been through inter- and intra-agency review, an external peer review and a public workshop. This 2012 USEPA BMDS Guidance is

recommended on the USEPA website accompanying the BMDS model and "provides guidance on the application of the benchmark dose approach for determining the point of departure for health effects data." Therefore, USEPA's (2012) BMDS Guidance represents accepted scientific methods across the scientific community whereas the draft operation procedure that OEHHA relied upon does not.

Assessing the goodness-of-fit of a model to the data is critical in selecting a benchmark dose and the first item listed in both the draft Standard Operating Procedure for USEPA subcontractors (USEPA 2014) and USEPA BMDS Guidance (USEPA 2012) is reliance upon the Akaike's Information Criterion (AIC) for comparison across models. The AIC is not reported by OEHHA (2020a). Rather, what is reported in OEHHA (2020a) is the difference between the AIC for the selected and non-selected models. The AIC values themselves, rather than the difference between values, are needed to truly understand fit of the models to the data.

Ramboll attempted to replicate the modeling conducted by OEHHA (2020a). In evaluating the modeling results, it appears that this difference in AIC may have been the sole reason for selection of a model; specifically, in some cases where the alternate model that was not selected has a better fit, based on Chi-square p-value, residual assessment and graphic fit. Further, these results suggest that OEHHA might have focused on selecting the model that provides the lowest obtainable POD, rather than selecting a model that provides the best fit to the data. However, it is important to note that these results cannot be confirmed completely from the OEHHA (2020a) report because OEHHA did not provide sufficient information to the public, but instead these results are determined from Ramboll's independent modeling of the data.

In addition, the method OEHHA (2020a) used to adjust for differential early mortality or significant differences in survival is a crude approach and is not recommended in either the USEPA (2005) Guidelines for Carcinogen Risk Assessment or the OEHHA (2009) Technical Support Document. Rather, the application of time-to-tumor models are noted in both Guidance documents to account for significant decreases in survival. These models are often preferred because they take into account all of the available animal-specific information regarding survival and the time at which the tumor of interest was observed. Moreover, because OEHHA (2020a) is relying upon an NTP study, the pathologists reviewing the results often draw conclusions about whether the tumors observed were incidental of the cause or death of the animal being examined. Relying upon time-to tumor models incorporates all of the available individual animal data from a study where survival problems were noted, rather than relying upon a crude approach which eliminates animals from consideration (determination of the "effective number") if they died before the first tumor was reported. This removal of animals from consideration can be important in accounting for the risk of completing death in the higher concentration groups.

The application of modeling approaches that are inconsistent with both finalized USEPA Guidelines and OEHHA Guidelines have resulted in the use of dose-response models that may not adequately characterize the available data. This may result in significant over- or underestimates of the potential potency of PCBTF. As such, OEHHA should re-evaluate the potential potency using generally accepted methods.

CONCLUSION

ACA and its members take their environmental stewardship responsibilities very seriously. PCBTF was developed as a substitute for use in ACA member products precisely because it assists in reducing the public health effects of ground level ozone. Currently, there are no viable alternatives available to replace PCBTF where it is used for this purpose. Accordingly, it is imperative that OEHHA's CSF/IUR accurately characterize the potential carcinogenicity of PCBTF, assuming there is such potential in humans. ACA urges the Scientific Review Panel to require OEHHA to revise its draft CSF/IUR. We believe the current draft document includes significant errors by not using the best available science, by failing to evaluate all available data, and by not using generally accepted methods.

Respectfully submitted,

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REFERENCES

Benigni R, Bignami M, Conti L, Crebelli R, Dogliotti E, Falcone E, Carere A. 1982. In vitro mutational studies with trifluralin and trifluorotoluene derivatives. Annali dell'lstituto superiore di sanita, 18(1): 123-126.

Braeuning A, Gavrilov A, Brown S, Wolf CR, Henderson CJ, Schwarz M. 2014. Phenobarbital-mediated tumor promotion in transgenic mice with humanized CAR and PXR. Toxicological Sciences, 140(2): 259-270.

Bryce SM, Shi J, Nicolette J, Diehl M, Sonders P, Avlasevich S, Raja S, Bemis JC, Dertinger SD. 2010. High content flow cytometric micronucleus scoring method is applicable to attachment cell lines, Environmental and Molecular Mutagenesis, 51: 260–266 (As cited in Clewell et al. 2018).

Bucher JR, Shackelford CC, Haseman JK, Johnson JD, Kurtz PJ, Persing RL. 1994. Carcinogenicity studies of oxazepam in mice. Fundamental and Applied Toxicology, 23: 280-297 (As cited in NTP 2018).

Clewell RA, Thompson CM, Clewell HJ III. 2018. Dose-dependence of chemical carcinogenicity: biological mechanisms for thresholds and implications for risk assessment. Chemico-Biological Interactions, 301: 112-127.

Deguchi Y, Yamada T, Hirose Y, Nagahori H, Kushida M, Sumida K, Sukata T, Tomigahara Y, Nishioka K, Uwagawa S, Kawamura S, Okuno Y. 2009. Mode of action analysis for the synthetic pyrethroid metofluthrin-induced rat liver tumors: Evidence for hepatic CYP2B induction and hepatocyte proliferation. Toxicological Sciences, 108(1): 69-80.

Doss M. 2014. Adoption of linear no-threshold model violated basic scientific principles and was harmful: Letter from Mohan Doss regarding Edward Calabrese's paper "How the US National Academy of Sciences misled the world community on cancer risk assessment: new findings challenge historical foundations of the linear dose response" (Arch Toxicol (2013) 87:2063-2081) and the letter from Ralph J Cicerone (Arch Toxicol (2014) 88:171-172). Archives of Toxicology, 88(3): 849-852.

Elcombe CR, Peffer RC, Wolf DC, Bailey J, Bars R, Bell D, Cattley RC, Ferguson SS, Geter D, Goetz A, Goodman JI, Hester S, Jacobs A, Omiecinski CJ, Schoeny R, Xie W, Lake BG. 2014. Mode of action and human relevance analysis for nuclear receptor-mediated liver toxicity: A case study with phenobarbital as a model constitutive androstane receptor (CAR) activator. Critical Reviews in Toxicology, 44(1): 64-82.

Gocke E, Muller L. 2009. In vivo studies in the mouse to define a threshold for the genotoxicity of EMS and ENU. Mutation Research, 78(2): 101–107 (As cited in Clewell et al. 2018).

Grisham JW. 1996. Interspecies comparison of liver carcinogenesis: implications for cancer risk assessment. Carcinogenesis, 18(1): 59-81.

Haines C, Elcombe BM, Chatham LR, Vardy A, Higgins LG, Elcombe CR, Lake BG. 2018. Comparison of the effects of sodium phenobarbital in wild type and humanized constitutive androstane receptor (CAR)/pregnane X receptor (PXR) mice and in cultured mouse, rat and human hepatocytes. Toxicology, 396-397: 23-32.

Holsapple MP, Pitot HC, Cohen SM, Boobis AR, Klaunig JE, Pastoor T, Dellarco VL, Dragan YP. 2006. Mode of action in relevance of rodent liver tumors to human cancer risk. Toxicological Sciences, 89(1): 51-56.

Johnson GE, Doak SH, Griffiths SM, Quick EL, Skibinski DO, Zaïr ZM, Jenkins GJ. 2009. Non-linear dose–response of DNA-reactive genotoxins: Recommendations for data analysis. Mutation Research, 678(2): 95-100 (As cited in Clewell et al. 2018).

Knaak JB, Smith LW, Fitzpatrick RD, Olson JR, Newton PE. 1998. In vitro hepatic metabolism of PCBTF: Development of Vmax and Km values and partition coefficients and their use in an inhalation PBPK model. Inhalation Toxicology, 10: 65-85.

Lake BG. 2018. Human relevance of rodent liver tumour formation by constitutive androstane receptor (CAR) activators. Toxicology Research, 7(4): 697-717.

Lee EG, Lewis B, Burns DA, Kashon ML, Kim SW, Harper M. 2015. Assessing exposures to 1-chloro-4-(trifluoromethyl) benzene (PCBTF) in U.S. workplaces. Journal of Occupational and Environmental Hygiene, 12(7): D123-D130.

Litton Bionetics. 1979. Mutagenicity Evaluation of Parachlorobenzotrifluoride (PCBTF) in the Sister Chromatid Exchange Assay in L5178Y Mouse Lymphoma Cells Final Report (1979), US Environmental Protection Agency. Document No. 40-7952010, Fiche No. OTS0508136.

Luisier R, Lempiainen H, Scherbichler N, Braeuning A, Geissler M, Dubost V, Muller A, Scheer N, Chibout SD, Hara H, Picard F, Theil D, Couttet P, Vitobello A, Grenet O, Grsal-Kraupp B, Ellinger-Ziegelbauer H, Thomson JP, Meehan RR, Elcombe CR, Henderson CJ, Wolf CR, Schwarz M, Moulin P, Terranova R, Moggs JG. (2014). Phenobarbital induces cell cycle transcriptional responses in mouse liver humanized for constitutive androstane and pregnane-x receptors. Toxicological Sciences, 139(2): 501-511.

Macri A, Ricciardi C, Stazi AV, Mantovani A, Marci CV, Piccioni A, Badellino E, Bianchi MP, Pepe M, Ceccanti M. 1987. Subchronic oral toxicity of 4-chloro-α,α,α-trifluorotoluene in Sprague-dawley rats. Food and Chemical Toxicology, 25(10): 781-786.

McMullin PD, Pendse S, Adeleye Y, Carmichael PL, Andersen ME, Clewell RA. 2016. Chapter 6: Using transcriptomics to evaluate thresholds in genotoxicity dose-response In: Toxicogenomics in Predictive Carcinogenicity (Editor: Waters MD, Thomas RS). Pp. 185-208 (As cited in Clewell et al. 2018).

Newton PE, Bolte HF, Richter WR, Akinsanya MB, Knaak JB, Smith LW. 1998. Inhalation toxicity, neurotoxicity, and toxicokinetic studies of p-chlorobenzotrifluoride. Inhalation Toxicology, 10(1): 33-48.

NTP. 1992. NTP Technical Report on Toxicity Studies of p-Chloro-α,α,α-Trifluorotoluene (CAS NO. 98-56-6) Administered in Corn Oil and α-Cyclodextrin to F344/N Rates and B6C3F$_1$ Mice in 14-Day Comparative Gavage Studies. NIH Publication 92-3133. United States Department of Health and Human Services. Public. National Institutes of Health. National Toxicology Program, Toxicity Report Series, Number 14.

NTP. 2018. NTP Technical Report: Toxicology and Carcinogenesis Studies of *p*-Chloro-a,a,a-trifluorotoluene (CASRN 98-56-6) in Sprague Dawley Rats (Hsd:Sprague Dawley SD) and B6C3F1/N Mice (Inhalation Studies), TR 594. Research Triangle Park, NC, June 2018.

Numazawa S, Shindo S, Maruyama K, Chibana F, Kawahara Y, Ashino T, Tanaka S, Yoshida T. 2005. Impaired nuclear translocation of CAR in hepatic preneoplastic lesions: Association with an attenuated CYP2B induction by phenobarbital. FEBS Letters, 579(17): 3560-3564.

Occidental Chemical Corporation. 1992. Initial submission: Mortality patterns of workers in the Niagara plant (final report on mixtures of chemical substances) with attachments and cover letter dated 022192. (As cited in NTP 2018).

OEHHA. 2009. Air Toxics Hot Spots Program Risk Assessment Guidelines. Technical Support Document for Cancer Potency Factors: Methodologies for derivation, listing of available values, and adjustments to allow for early life stage exposures. California Environmental Protection Agency Office of Environmental Health Hazard Assessment. Online at: http://oehha.ca.gov/air/crnr/technical-support-document-cancer-potency-factors-2009.

OEHHA. 2020a. Air Toxics Hot Spots Program. p-Chloro-α,α,α-trifluorotoluene (*p*-Chlorobenzotrifluoride, PCBTF). Cancer Inhalation Unit Risk Factor, Technical Support Document for Cancer Potency Factors, Appendix B. Scientific Review Panel Draft. Air and Site Assessment and Climate Indicator Branch, Office of Environmental Health Hazard Assessment, California Environmental Protection Agency. January 2020.

OEHHA. 2020b. Responses to Public Comment on the draft cancer inhalation unit risk factor for p-chloro-α,α,α-trifluorotoluene (Parachlorobenzotrifluoride, PCBTF). Office of Environmental Health Hazard Assessment, California Environmental Protection Agency. PCBTF IUR Document.

Parkinson A, Leonard N, Draper A, Ogilvie BW. 2006. On the mechanism of hepatocarcinogenesis of benzodiazepines: Evidence that diazepam and oxazepam are CYP2B inducers in rats, and both CYP2B and CYP4A inducers in mice. Drug Metabolism Review, 38: 235-259 (As cited in NTP 2018).

Peffer RC, Bailey K, Lichti-Kaiser K, Lake B. 2020. Constitutive androstane receptor activation leading to hepatocellular adenomas and carcinomas in the mouse and the rat. Adverse Outcome Pathway WIKI. Available at: https://aopwiki.org/aops/107.

Pelosi GD, Oberdoerster J, Olson JR, Knaak JB, Smith LW, Newton PE. 1998. Characterization of rat hepatic cytochrome p-450 activities following inhalation exposure to p-chlorobenzotrifluoride. Inhalation Toxicology, 10: 49-63.

Preston RJ, Hoffman GR. 2013. Chapter 9: Genetic Toxicology. In: Casarett & Doull's Toxicology, The Basic Science of Poisons. 8th Edition. (editor – Curtis D. Klaassen). McGraw Hill Education.

Ross J, Plummer SM, Rode A, Scheer N, Bower CC, Vogel O, Henderson CJ, Wolf CR, Elcombe CR. 2010. Human constitutive androstane receptor (CAR) and pregnane X receptor (PXR) support the hypertrophic but not the hyperplastic response to the murine nongenotoxic hepatocarcinogens phenobarbital and chlordane in vivo. Toxicological Sciences, 116(2): 452-466.

Sakamoto Y, Inoue K, Takahashi M, Taketa Y, Kodama Y, Nemoto K, Degawa M, Gamou T, Ozawa S, Nishikawa A, Yoshida M. 2013. Different pathways of constitutive androstane receptor-mediated liver hypertrophy and hepatocarcinogenesis in mice treated with piperonyl butoxide or decabromo-diphenyl ether. Toxicologic Pathology, 41: 1078-1092 (As cited in NTP 2018).

Tubiana M, Aurengo A, Averbeck D, Masse R. 2006. The debate on the use of linear no threshold for assessing the effects of low doses. Journal of Radiological Protection, 26(3): 317-324.

USEPA. 1986. Guidelines for carcinogen risk assessment. United States Environmental Protection Agency. EPA/630/R-00/004, Federal Register 51(185): 33992–43003.

USEPA. 2005. Guidelines for carcinogen risk assessment. United States Environmental Protection Agency, Risk Assessment Forum. Report number EPA/630/P-03/001F.

USEPA. 2012. Benchmark Dose Technical Guidance. United States Environmental Protection Agency, Risk Assessment Forum. Washington, DC.

USEPA. 2014. Choosing Appropriate Stage of a Multistage Model for Cancer Modeling (BMDS Technical Guidance). United States Environmental Protection Agency. https://cfpub.epa.gov/ncea/bmds/recordisplay.cfm?deid=308382.

USEPA. 2018. Strengthening Transparency in Regulatory Science. United States Environmental Protection Agency. 40 CFR Part 30 [EPA-HQ-2018-0259; FRL-9977-40-ORD]. Federal Register, 83(83): 18768-18774 (Monday, April 30, 2018).

USEPA. 2019. Letter to The Honorable Andrew R. Wheeler: Consultation on Updating EPA Guidelines for Carcinogen and Non-Cancer Risk Assessment. United States Environmental Protection Agency, Office of the Administrator Science Advisory Board. July 15, 2019. Washington, DC. EPA-SAB-19-003.

Vaiserman A, Koliada A, Zabuga O, Socol Y. 2018. Health impacts of low-dose ionizing radiation: current scientific debates and regulatory issues. Dose Response, 16(3): 1-27.

Waxman DJ, Azaroff L. 1992. Phenobarbital induction of cytochrome P-450 gene expression. Biochemical Journal, 281: 577-592.

Yamada T, Uwagawa S, Okuno Y, Cohen SM, Kaneko H. 2009. Case study: an evaluation of the human relevance of the synthetic pyrethroid metofluthrin-induced liver tumors in rats based on mode of action. Toxicological Sciences, 108(1): 59-68.

Yamamoto Y, Moore R, Goldsworthy TL, Negishi M, Maronpot RR. 2004. The orphan nuclear receptor constitutive active/androstane receptor is essential for liver tumor promotion by phenobarbital in mice. Cancer Research, 64(20): 7197-7200.

Yoshinari K, Sueyoshi T, Moore R, Negishi M. 2001. Nuclear receptor CAR as a regulatory factor for the sexually dimorphic induction of CYB2B1 gene by phenobarbital in rat livers. Molecular Pharmacology, 59(2): 278-284.

Yuan J, Jameson CW, Coehl TJ, Elwell MR, Leininger JR, Thompson MB, Corniffe, Carlton T. 1992. Application of molecular encapsulation for toxicology studies: Comparative toxicity of p-Chloro-α,α,α-trifluorotoluene in α-Cyclodextrin vehicle versus corn oil vehicle in mal and female Fischer 344 rats and B6C3F1 mice. Fundamental and Applied Toxicology, 18(3): 460-470.