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SUBJECT: Response to comments by the Office of Environmental Health Hazard Assessment on DPR's 2020 Allyl Isothiocyanate Draft Risk Characterization Document

Background

At the request of the Department of Pesticide Regulation (DPR), the Office of Environmental Health Hazard Assessment (OEHHA) reviewed the July 2020 Draft Risk Characterization Document (RCD) for Allyl Isothiocyanate. OEHHA was asked to respond to a series of charge questions covering the hazard identification, exposure assessment, risk characterization, and worker and bystander margins of exposure, and provided comments to DPR on October 28, 2020.

This memorandum summarizes DPR's responses to OEHHA's comments on the draft RCD in an itemized fashion, and is divided into the following sections: Detailed Comments; Response to Charge Statements; and Minor Comments. Corresponding revisions were also made to the final RCD and its appendices as appropriate. Responses specific to the exposure assessment are detailed in a separate memorandum.

Note that references cited in this memorandum are specific to OEHHA comments or DPR's response, and not necessarily duplications of those in the draft or final RCD. Likewise, every effort has been made to ensure that any references to tables found in the draft or final RCD are clear. Tables specific to this memorandum are numbered independently of the RCD. All OEHHA comments in this memorandum are direct quotes from the documents, which can be found at https://oehha.ca.gov/media/downloads/pesticides/document/commentsaitc110320.pdf.

OEHHA Detailed Comments – Toxicity Evaluation and Point of Departure Determination

1. Non-cancer Toxicity Evaluation and Point of Departure Determination

a. Pharmacokinetics: The absorption, distribution, metabolism, and excretion of AITC are adequately addressed in the draft RCD. A lack of inhalation absorption data led DPR to assume a default inhalation absorption of 100%. OEHHA notes that the increased levels in urinary bladder tissue in male rats occurs following both oral and intravenous exposure (Ioannou et al., 1984). This study also observed nearly twice the volume of urine in female rats relative to males. Lower urine volume in male rats may have led to more concentrated levels of AITC in the urine and thus in the bladder tissue. However, a previous study by Muztar et al. (1979) observed a two-fold increase in urinary output in male rats administered AITC, compared to controls. Thus, the effect of urinary volumes on AITC disposition is unclear.

> **DPR Response:** The reference Muztar *et al.* (1979) has been added to the final RCD. As mentioned by OEHHA, Muztar *et al.* (1979) showed that AITC has a diuretic effect (increased urine production) in rats. Ioannou *et al.* (1984) showed that urine volume production is not the same in untreated or treated males and females, but rather untreated and treated females produced approximately twice the urine volume than male counterparts. Ioannou et al. (1984) also showed that AITC diuretic effect was more evident in the females and not in males compared to vehicle-treated rats. Although the mechanism of AITC's diuretic effect is not known, the sexually differentiated diuretic effect was demonstrated in rats by Ioannou *et al.* (1984). While Muztar *et al.* (1979) reported an increased urinary output in male rats, the study did not capture sex differences because only male rats were tested. Taken together, the evidence suggests that this sex difference may have contributed to the relatively higher urinary concentrations of AITC and metabolites in males than females, which explains the correspondingly higher bladder toxicity in males than in females.

a. Pharmacokinetics, continued: There is no data regarding the possibility and extent of pulmonary metabolism of AITC in rodents and humans following inhalation exposure. However**,** there is also no available data to indicate that the metabolites of AITC through the inhalation and oral routes, at least qualitatively, are expected to be different. As the main route of excretion following oral or intravenous exposure appears to be via urine, it seems likely that increased levels of AITC metabolites in urinary bladder tissue could result from inhalation exposure as well, though there may be quantitative differences depending on the route. Urinary bladder hyperplasia was the critical effect observed in male and female rats following oral exposure, with bladder tumors also observed in male rats.

DPR Response: No data are available to characterize the toxicokinetics of AITC by the inhalation route. Multiple reports examining AITC toxicokinetics by the oral route indicate that its metabolites are predominantly excreted in urine, and are consistent with the hyperplastic effect seen in bladder epithelium. Additionally, multiple oral toxicity studies showed positive correlations between incidence and severity of urinary bladder hyperplastic lesions and AITC dose. The urinary hyperplastic lesions in oral toxicity studies were observed after only two weeks and persisted through two years of oral AITC administration. In contrast, following 13 weeks of inhalation exposure, urinary bladder lesions were not observed (Randazzo, 2017). Therefore, AITC and/or its toxic metabolites, if present in urine after inhalation exposure, did not achieve concentrations sufficient to induce toxicity. Furthermore, other systemic effects such as retinopathy and cataracts that were observed at higher oral doses also did not occur in the 13-week inhalation study. Therefore, based on Randazzo (2017), DPR determined that inhalation exposure to AITC at 25 ppm and lower does not induce urinary bladder hyperplasia in rats.

b. Acute Toxicity: DPR selected a critical acute POD of 2.5 ppm based on decreased motor activity in rats following a single four-hour nose-only exposure to AITC vapor (Herberth et al, 2017). OEHHA agrees that the Herbeth et al. (2017) study is the most sensitive data set available and concurs with the use of a 10-fold LOAEL-to-NOAEL extrapolation factor. It should be noted that AITC was found to be a dermal sensitizer in studies in humans and mice, and is a respiratory irritant. There is potential for AITC to also be a respiratory sensitizer in humans following repeated exposures.

DPR Response: Experimentally and clinically AITC or mustard oil is known to induce dermal sensitization in humans (Landsteiner and Di Somma, 1938; Lerbaek et al., 2004; Gaul, 1964) and in multiple animal species (Landsteiner and Di Somma, 1938; Durndo 2012c). These reports show that the severity of the sensitization has a concentration threshold (Landsteiner and Di Somma, 1938; Durndo 2012c). While no data on respiratory sensitization were identified, it is plausible that AITC can induce respiratory sensitization following inhalation exposure. A statement to this effect has been added to the final RCD.

c. Subchronic Toxicity: A single subchronic inhalation study was identified by DPR… OEHHA used the benchmark dose model (BMD) to estimate the PODs of the dataset presented in Table 1 [in the OEHHA comments document] and found some models predict BMDLs lower than the NOAEL of 6.6 mg/kg-day. OEHHA suggests DPR model the data and select the most health protective estimate, after taking into account route-specific issues (e.g., toxicokinetics).

DPR Response: As suggested by the reviewers, we modeled the urinary bladder hyperplasia in the 13 week oral study by Hasumura et al (2011) for both males and

females using BMDS 3.1.2 (Appendix C to the final RCD).The resultant $BMDL_{10}$ values were 6.0 and 6.3 mg/kg/day horseradish extract in males and females, respectively. The equivalent calculated dose of AITC was 4.9 and 5.2 mg/kg/day after correcting for the AITC content in HRE (82%). These BMDLs were very similar to the study NOEL of 6.6 mg/kg/day AITC (8 mg/kg/day HRE) determined by Hasumura *et al.* (2011). Moreover, the air concentration derived by route-to-route extrapolation for the lowest BMDL of 4.9 mg/kg/day in males was 7.1 ppm[1], which was similar to the 5 ppm AITC NOEL derived from the subchronic inhalation study (Randazzo, 2017). Consequently, we expect 5 ppm to be protective of the urinary bladder epithelial hyperplasia that was observed in the oral study (Hasumura et al., 2011). The modeling results from Hasumura et al (2011) are now included in Appendix C and in Section E.1.3 in the final RCD.

c. Subchronic Toxicity, continued: On page 53 of the draft RCD, DPR also reasoned that because urinary bladder hyperplasia were not observed in the inhalation rat study reported by Randazzo et al. (2017), even in the high-dose rats at 25 ppm, this effect appeared to be specific to the oral route of exposure. OEHHA disagrees that hyperplasia and tumor formation in the urinary bladder are unique to oral exposure:

1) There is no absorption, distribution, metabolism, and excretion (ADME) data to suggest that different metabolites of AITC are formed following inhalation than with oral exposures, though they may be quantitatively different…

DPR Response: As mentioned in the RCD, due to a lack of inhalation ADME data, the toxicokinetic profile of AITC by the inhalation route is not known. However, from the oral studies it is evident that AITC is absorbed and metabolized, leading to systemic effects including urinary bladder hyperplasia, retinopathy, and cataracts. Urine was the major route of excretion for the reactive metabolite N-acetyl-S-(N-allylthiocarbamoyl) cysteine. Its appearance in urine corresponds with the occurrence of urinary bladder hyperplasia within 2 weeks of oral exposure to AITC. However, none of the histopathological effects (bladder hyperplasia, retinopathy, cataracts) were detected following 13 weeks of inhalation exposure in Randazzo (2017). Therefore, it is reasonable to conclude that toxicokinetic disposition differs by the inhalation and oral routes and suggests that air concentrations greater than 25 ppm (the highest tested concentration in Randazzo (2017)) might be required to induce the systemic effects seen on oral exposure. Using the breathing rate default and duration assumptions indicated in

[1] *Route to route extrapolation, internal dose to equivalent air concentration:*
Inhalation POD ppm = Rat Oral POD (mg/kg) / rat BR (m^3/kg) / AITC conversion factor:
Subchronic oral POD = 5 mg/kg/day; Default rat breathing rate (BR) = 0.17 m^3/kg, derived from the 24-hour default breathing rate of 0.96 m^3/kg adjusted by duration of inhalation exposure (6 hours per day; 5 days per week), as follows – 0.96 m^3/kg x 6h/24h x 5 days/7 days; AITC conversion factor, mg/m^3 to ppm = 4.06; Therefore, POD = 4.9 mg/kg/day / 0.17 m^3/kg / 4.06 = 7.1 ppm

footnote 1, 25 ppm by the inhalation route is equivalent to 17 mg/kg/day by the oral route after extrapolation. Therefore, the subchronic inhalation NOEL of 5 ppm should protect against effects observed in both the subchronic oral (bladder hyperplasia) and inhalation (decreased motor activity and olfactory epithelial degeneration) studies. While the effects noted in inhalation and oral studies are qualitatively different, it is our assertion that urinary bladder hyperplasia is the most sensitive non-acute endpoint and that protecting against this endpoint will protect against the occurrence of other subchronic or chronic effects, including urinary bladder tumors.

c. Subchronic Toxicity, continued:

… Furthermore, the assumption of 100% absorption by inhalation also suggests that exhalation of unchanged AITC is not expected to be significant, and most of the AITC inhaled would be absorbed into systemic circulation. As there is no data indicating an alternative route of excretion, it can only be assumed that these metabolites are mainly excreted through the urine. High concentrations of one or more of these metabolites in the urinary bladder could be expected to cause hyperplasia in this target organ via either route.

DPR Response: Due to the lack of data on AITC absorption by the inhalation route, DPR assumed a default of 100 % for AITC. However, this should not be taken to mean that inhaled AITC is excreted primarily in urine as it is following oral exposure or that the metabolite profile is similar for the two routes. The absence of bladder hyperplasia by the inhalation route in the subchronic study suggests that something is indeed different between the two routes. Ultimately, based on the actual histopathological data, the NOEL of 5 ppm is protective of all effects that were examined for inhalation route, including urinary bladder hyperplasia.

c. Subchronic Toxicity, continued:

2) The fact that no urinary bladder hyperplasia was reported in the 13-week inhalation study (Randazzo et al., 2017) could be explained by either the relatively low exposure levels or the short exposure duration or a combination of both. The study results of Randazzo et al (2017) cannot conclusively prove the effects observed in the subchronic oral studies are not relevant for inhalation exposure. OEHHA recommends DPR to consider the factors discussed in their evaluation of the subchronic oral studies.

DPR Response: The most sensitive effects by the inhalation route were motor activity alterations and histopathological changes to olfactory and respiratory epithelium (Randazzo, 2017). Therefore, regardless of the presence or absence of ADME differences between the oral and inhalation routes, urinary bladder hyperplasia did not appear to be relevant to inhalation exposure, as it was not observed even at the highest concentration of 25 ppm. Moreover, the subchronic inhalation POD of 5 ppm based on motor activity, rearing counts and changes to nasal epithelium should be protective of other effects,

including any urinary bladder hyperplasia that might occur at the doses higher than 25 ppm. Additionally, oral-to-inhalation route extrapolation of the 5 mg/kg/day $BMDL_{10}$ for urinary bladder hyperplasia observed in the oral study by Hasumura et al. (2011) resulted in an extrapolated value of 7.1 ppm, which is greater than the NOEL of 5 ppm in Randazzo (2017) study.

d. Chronic Toxicity: No inhalation studies for chronic toxicity were available for evaluation in the draft RCD. Two high quality chronic oral toxicity studies are available and were evaluated by DPR… It should be noted that though transitional cell papillomas and epithelial hyperplasia of the urinary bladder were observed in male rats (NTP, 1982), NTP noted that these effects did not occur in the same animals. This would suggest that hyperplasia may not be a required precursor for the urinary bladder tumors, which is contradictory to the statement on pages 55 and 56 in the draft RCD...

DPR Response: Sustained AITC metabolite(s) in urine leading to bladder hyperplasia and eventual tumors is supported as the mode of action (MOA) for bladder tumors by weight of evidence analysis. The relevance of this MOA to human health risk assessment is supported by US EPA and other agencies (US EPA, 2006; EFSA, 2010). Urinary bladder hyperplasia is a necessary intermediate step in the pathogenesis of neoplasia by this MOA in rats (USEPA, 2006), where hyperplasia appears earlier and at lower doses than papillomas. Time and dose concordance for AITC-induced hyperplasia and tumors is also evident in data reported by Cho et al. (2017), Hasumura et al. (2011), and NTP (1982). Cho et al. (2017) demonstrated that AITC induced bladder hyperplasia at both 4.1 and 15.7 mg/kg/day, but papillomas were observed only at 15.7 mg/kg/day, supporting the interpretation that hyperplasia is a key precursor event for AITC-induced papillomas. Where there is a transition from a primary to a secondary lesion, the identification of papillomas might preclude identification of hyperplasia in the same animal tissue (Bryan and Cohen, 1983). It is possible that hyperplasia developed into papilloma and was recorded as such in those animals, while the rest of the 7/50 animals with hyperplastic lesions did not develop papilloma. It is also possible that only the most severe lesions were observed in any animal, resulting in papillomas but not hyperplasia when both occurred in the same animal.

e. Reproductive and Developmental Exposure: Teratology studies were available in mice, rats, hamsters, and rabbits…For the hamster, the draft RCD (page 56) reported an increase in incidence of incomplete sternebral ossification in fetuses of hamsters at the highest tested dose (23.3 mg/kg-day), but stated the effect was not found to be statistically significant or toxicologically relevant. However, in the Summary of Toxicological Data on AITC (2018), DPR reported the increased litter and fetal incidence of incomplete ossification of sternebrae and determined a developmental NOAEL of 5.1 mg/kg-day. OEHHA suggests that DPR address their inconsistencies in the interpretation of the data.

DPR Response: The Summary of Toxicology Data document prepared by HHA's Active Ingredient Section was designed to summarize reviews of toxicology data specifically in support of registration and to determine if there were data gaps in the submitted studies. Those findings are specific to the suitability of Registrant or other data submitted to support registration pursuant to FIFRA guidelines. However, when developing an RCD, DPR considers not only these reviews, but all relevant data and interpretations and conducts further evaluations of the data to establish and refine critical endpoints for risk assessment and to propose regulatory targets for risk management consideration. Because the purpose of the reviews in the Summary of Toxicology Data are to support registration evaluation, they may arrive at different conclusions than the corresponding reviews in the RCD to support risk assessment.

With regard to the developmental study in hamsters, the incidences of incomplete sternebral ossification in fetuses were as follows: 17/21 for control, 21/22 (0.2 mg/kg/day group), 18/20 (1.1 mg/kg/day group), 21/23 (5.1 mg/kg/day) and 22/23 (23.8 mg/kg/day). DPR determined that the marginal effects in hamsters at 23.8 mg/kg-day noted in the Summary of Toxicology and by OEHHA was not of toxicological significance because the incidences of ossification delays were not statistically elevated compared to controls. Therefore, the 5.1 mg/kg/day in hamsters was not selected as the developmental NOEL. Instead, the lowest relevant maternal and developmental NOELs were set at 6 mg/kg/day based on increased litters with resorption sites, and numbers of dead fetuses in mice at 28 mg/kg/day. Route-to-route conversion indicates the equivalent oral dose for the acute inhalation POD (2.5 ppm, the ENEL from Herberth, 2017) would be 1.4 mg/kg/day. This is lower than the 6 mg/kg/day oral maternal and developmental NOEL. Therefore, the selected acute POD (and subchronic and chronic PODs) is protective of maternal and developmental effects.

e. Reproductive and Developmental Exposure, continued: OEHHA disagrees with DPR's determination that fetal and pup effects were plausibly secondary to maternal toxicity and were thus not considered toxicologically significant. Co-occurrence of fetal and maternal toxicity does not necessarily indicate causation. Even if there are sufficient mechanistic data to determine that a fetal effect is due to a specific maternal deficit, the fetal effect still represents developmental toxicity. The US Environmental Protection Agency (US EPA) notes that whether developmental effects are secondary to maternal toxicity or not, the maternal effects may be reversible while effects on the offspring may be permanent (US EPA, 1991) … The PODs selected in the draft RCD were 2.5 ppm for acute, 5 ppm for subchronic, and 0.5 ppm for chronic exposure. These PODs appear to be protective of reproductive and developmental effects observed in the animal toxicity studies, and OEHHA suggests DPR include dose equivalent calculations for the most sensitive developmental endpoint in their discussion.

DPR Response: DPR conducted a critical analysis of Food and Drug Research Labs (FDRL) (1973), particularly the effects in hamster. Only summary data were available. Individual pup or litter level data were not available for evaluation. Based on our statistical analysis of the available summary data, we concurred with the study authors and concluded that slightly increased incidence in the incomplete sternabral ossification in fetuses of hamsters at the highest tested dose was not statistically significant by Fisher's test comparing the control against dose groups for incidence to total number of pups born. However, mice exhibited increased number of litters with resorption sites and dead fetuses at a 28 mg/kg/day, resulting in a study maternal and developmental NOEL of 6 mg/kg/day. Our calculations show that the critical acute (2.5 ppm), subchronic (5 ppm), or chronic (0.5 ppm) inhalation PODs result in 1.75, 3.5, and 0.35 mg/kg/day inhalation-to-oral calculated doses, respectively, which are all lower than the NOEL (6 mg/kg/day) derived for maternal and developmental effects in the developmental toxicity study by FDRL (1973). Therefore, the effects observed in pups and dams appear at higher doses than the inhalation-to-oral converted doses for acute, subchronic, and chronic critical PODs in the RCD.

2. Carcinogenicity:

The available carcinogenicity studies are two-year gavage studies of food-grade AITC (purity > 93%) in male and female F344/N rats and B6C3F1 mice (NTP, 1982) and two-year drinking water studies of horseradish extract (HRE) containing 82 – 86% AITC in male and female F344/DuCrj rats (Cho et al., 2017). The draft RCD summarized the three tumor sites (urinary bladder tumors, leukemia, and fibrosarcomas) observed in oral studies. It is OEHHA's position that the three tumor sites are treatment related, and the cancer potency should be based on the multisite analysis for the bladder papilloma and leukemia from the NTP (1982) male rat study.

DPR Response: DPR agrees with OEHHA that urinary bladder tumors and possibly subcutaneous fibrosarcomas are caused by AITC. DPR does not agree with OEHHA's position that the undifferentiated leukemia is treatment related, nor that a multisite tumor analysis is appropriate for the observed tumor sites (or at least for bladder papillomas and leukemia). With respect to the undifferentiated leukemia, DPR's position is corroborated by the study authors and the EFSA panel on AITC (EFSA, 2010).

Subcutaneous fibrosarcomas were observed in one study (NTP, 1982) in one sex and only at the high dose. Linear extrapolation to derive a cancer potency slope is recommended by US EPA when a chemical is mutagenic or when it lacks data to show an alternative non-mutagenic MOA. DPR concluded that mutagenicity is unlikely to be an MOA for AITC-induced tumors based on evaluation of the genotoxicity database, nor was there evidence for an alternative MOA. Furthermore, absence of a dose response relationship within the chosen dose range precluded cancer potency calculation.

Urinary bladder tumors were observed in both chronic oral studies (NTP, 1982; Cho et al., 2017). DPR's weight of evidence analysis supports hyperplasia as a requisite precursor event. Consequently, DPR concluded that AITC exposures at doses that do not result in urinary bladder hyperplasia would not cause urinary bladder tumors. Unlike the oral subchronic studies, the subchronic inhalation study did not show evidence of urinary bladder hyperplasia even at 25 ppm. Therefore, a cancer potency analysis was not deemed appropriate for any of the observed tumors, either separately or through multisite tumor analysis.

Again emphasizing the results of the available studies, while there was a rapid induction of urinary bladder hyperplasia in the oral study, there was no evidence of bladder hyperplasia at the end of the 13-week inhalation study. Therefore, repeated inhalation exposure to AITC does not present a risk for bladder tumors at the air concentrations tested. The route-to-route extrapolation (see RCD Section E.1.4) showed that the critical chronic inhalation POD for decreased motor activity and olfactory epithelial degeneration will be protective of any systemic toxicity, including bladder hyperplasia and tumors.

2. Carcinogenicity, continued: …With regards to undifferentiated leukemia in male rats, OEHHA does not agree with the conclusion in the draft RCD that relies on comparison of leukemia incidence with the historical controls, and suggests including this treatment-related tumor site in the cancer potency estimate. The incidences were 2/50, 6/50, 8/50, or 4%, 12%, and 16%, in control, low-dose, and high-dose, respectively. The incidence in the high-dose group was significantly increased by pairwise comparison with control, and there was a dose-related trend (draft RCD Table 11). NTP (1982) reported that the increase was not statistically significant from the historical controls (96/999 or 10%). However, the NTP (2015) Handbook for Preparing Report on Carcinogen Monographs states that while historical control data from the testing laboratory can be helpful, "the concurrent controls are considered to be the most relevant comparison group for evaluating potential exposure-related tumor effects." As a generally accepted scientific principle, this approach is also used by the US EPA (2005) in its Guidelines for Carcinogen Risk Assessment, which states that the preferred standard for determining statistical significance of tumor incidence comes from a comparison of tumors in dosed animals with those in concurrent control animals.

DPR Response: DPR's position is that the increased incidence of undifferentiated leukemia was not treatment related. The relationship between F344/N strain of rats and undifferentiated leukemia is unusual due to the high variability and high background rate of this tumor in this rat strain. Therefore, in addition to dose response data, DPR considered historical background data (NTP 1982; Thomas et al), meta-analysis information by other authors (Thomas et al), and lack of reproducibility in a related sub-strain (F344/DuCrj used by Cho et al. 2017) while assessing undifferentiated tumors

response. Taken together, DPR agrees with the study authors and an independent EFSA panel (EFSA, 2010) that the leukemias seen in the NTP 1982 study were unlikely to be related to AITC treatment.

2. Carcinogenicity, continued: OEHHA also does not agree with the statement in draft RCD that "there was compelling evidence that the observations were artifacts of the study design and the selected rat strain (F344/N) rather than AITC treatment." The undifferentiated leukemia in rats is also known as mononuclear cell leukemia (MNCL) (NTP, 1982). Although OEHHA doesn't assume or require tissue concordance between tumors found in animals studies and those that occur in humans, rat MNCL does have a human counterpart and there is human relevance. US EPA (2012a) noted that several authors have concluded that rat MNCL is similar to human natural killer cell (NK)-large granular lymphocyte leukemia (Stromberg et al., 1985; Ishmael and Dugard, 2006; Thomas et al., 2007). MNCL was also one of the tumor types in the same strain of rat (F344) used by OEHHA to derive a cancer potency estimate for Diisononyl Phthalate in the development of a No Significant Risk Level (NSRL) under California's Proposition 65 (OEHHA, 2015) …

DPR Response: DPR does not dispute the human relevance of these tumors. Rather, DPR concludes that the undifferentiated leukemia seen in the NTP (1982) study was due to a propensity of the rat strain (F344/N) for these tumors. In other words, the incidence data were not a reliable indicator of a treatment effect. In addition, the apparent effect was not reproducible in a different rat strain (F344/DuCrj in Cho et al., 2017; see response above).

2. Carcinogenicity, continued: … In Table 11 of the draft RCD, there is a mistake indicating significance by pairwise comparison in the high dose males, when the p value is in fact not statistically significant … There was also a typo in the table legend indicating statistical significance at $p<0.5$, rather than $p<0.05$ …

DPR Response: These corrections have been made in the final RCD.

2. Carcinogenicity, continued: … [W]hen calculating animals at risk, OEHHA suggests using animals alive at the appearance of the first tumor. There was an approximate 25% mortality in the high dose group at the appearance of the first bladder tumor. When analyzing tumor incidences with animals at risk as the sample size, the incidence of transitional-cell papilloma in the high dose males was statistically significant by pair-wise comparison. OEHHA recommends DPR reevaluate incidences of all tumors using this method.

DPR Response: DPR agrees that performing an animals-at-risk analysis would be appropriate if the data required a dose response analysis of urinary bladder tumors. However, low dose linear extrapolation was determined to be inappropriate in this case

because the data indicated a threshold effect. The hyperplasia-based POD is expected to protect from the development of urinary bladder papilloma.

2. Carcinogenicity, continued: In the two-year drinking water study by Cho et al (2017), there were also increases in urinary bladder papilloma in high-dose male rats (1/32, 0/32, 3/32), but the incidences were not statistically significant by pairwise or by Exact trend test. Regardless, urinary bladder transitional-cell papilloma is a rare tumor type (Haseman et al. 1998) and OEHHA considers the urinary bladder transitional-cell papilloma to be treatment-related with the data from male rats in the NTP study (1982) adequate for cancer potency estimation.

DPR Response: DPR agrees that the data from male rats in NTP (1982) would be adequate for cancer potency estimation if not for the weight of evidence indicating that urinary bladder tumors result from sustained urinary bladder hyperplasia due to urinary metabolite(s) leading to eventual bladder tumors. Because the weight of evidence supports urinary bladder hyperplasia as a precursor lesion, cancer potency estimation was not supported by the data. Again, the route-to-route extrapolation (See RCD section E.1.4) showed that the critical chronic inhalation POD for decreased motor activity and olfactory epithelial degeneration would be protective of any systemic toxicity, including bladder hyperplasia and tumors.

2. Carcinogenicity, continued: Furthermore, OEHHA does not see evidence that these tumors were caused by route-specific mechanisms. AITC has not been adequately tested by inhalation in two-year cancer bioassays, and it is inappropriate to make conclusions for the inhalation route based on results from sub-chronic studies. There is no evidence for route-specific differences in ADME that supports the hypothesis that the carcinogenic effect of AITC is limited to the oral route. The draft RCD noted in the ADME section that "The oral absorption in rats and mice was estimated to be > 90%. DPR considers oral absorption > 90% as complete (100%). In the absence of data for inhalation uptake, DPR assumes a default inhalation absorption of 100%." In addition, positive findings related to some cancer key characteristics (electrophilicity, genotoxicity and induction of oxidative stress) indicate that AITC acts systemically...

DPR Response: As discussed above, there are no inhalation toxicokinetic data for AITC. However, there are subchronic studies with which it is possible to compare the effects of AITC by the oral and inhalation routes. Urinary bladder hyperplasia, a prerequisite for bladder tumors, was observed by the oral but not by the inhalation route after 13 weeks of exposure. Urinary bladder hyperplasia was the most sensitive subchronic and chronic effect by oral route. This effect correlates with oral toxicokinetic studies that demonstrate the excretion of high levels of the major metabolite of AITC in urine. These observations indicate that effects of AITC by the oral route are different from the inhalation route and likely relate to differences in ADME. More specifically, quantitative differences in the amounts of AITC and its reactive metabolites at internal sites of toxicity likely contribute

to the differences in effects observed between oral and inhalation routes. DPR considers the absence of urinary bladder hyperplasia after inhalation to be comparable to the hyperplastic oral doses (compared after oral-to-inhalation dose conversion, see response to OEHHA Comment 5a) where the lesion was observed to be the critical factor in determining the route dependency of the effect.

Although DPR assumes 100% absorption by both oral and inhalation route, similarity in metabolism and excretion by these two routes is not assumed. It is plausible that differences in metabolism and excretion by the inhalation route account for the absence of urinary bladder effects observed in studies using the oral route.

2. Carcinogenicity, continued: … OEHHA disagrees with the conclusion in the draft RCD that "any positive results for AITC may not have been mediated by direct DNA-reactivity." AITC is a highly reactive compound, which has been shown in vitro to form adducts with proteins (Kawakishi & Kaneko, 1987) and glutathione (Kawakishi & Kaneko, 1985). A study by Kassie and Knasmuller (2000) found that AITC induced formation of thiobarbituric acid reactive substances (a marker of lipid peroxidation) in HepG2 cells in vitro, and that reactive oxygen species may be involved in the AITC induced DNA damage in E coli. These findings are related to electrophilicity and induction of oxidative stress, two key characteristics of carcinogens (Guyton et al., 2018). Positive results of several genotoxic endpoints as summarized in the draft RCD and by IARC (1999) support that AITC is genotoxic to various cellular targets in vitro, and/or in vivo. Notably, AITC induced DNA strand breaks and oxidative damage to DNA in humans in vivo (Charron et al. 2013). While there are negative and some weakly positive or equivocal findings in the genotoxicity database, it is OEHHA's opinion that they are not sufficient to discount the positive genotoxicity findings.

DPR Response: DPR agrees with OEHHA that AITC is positive in many genotoxicity assays. DPR analyzed the genotoxicity database according to US EPA's Guidelines for Carcinogen Risk Assessment (US EPA, 2005) to determine if AITC was a direct DNA reactive and a mutagenic agent. DPR also agrees that AITC is a highly reactive compound and forms protein adducts *in vitro*. However, the available evidence did not support a determination that AITC is mutagenic *in vivo*. Critically, AITC was negative in multiple *in vivo* mutagenic assays and was negative-to-weakly positive (~2-fold compared to control) in *in vitro* bacterial mutagenicity assays at cytotoxic concentrations. DPR concluded that AITC is unlikely to be mutagenic under physiological conditions, in agreement with USEPA and EFSA assessments.

With respect to other forms of genotoxicity, DPR agrees with OEHHA that AITC was demonstrably genotoxic in several assays. However, in an *in vivo* human trial, AITC consumption for 10 days did not result in DNA strand breaks, nor did it significantly induce 8-Oxo-7,8-dihydro-2'-deoxyguanosine (8-oxodG), a marker of DNA oxidation.

DNA strand breaks were only transiently present when bolus doses of AITC were consumed. No other evidence of DNA adduct formation by AITC *in vivo* or *in vitro* was identified. In an *in vitro* DNA alkylating assay, AITC showed poor or borderline reactivity compared with other positive chemicals according to the study authors. It is plausible that reactive AITC is suppressed by cellular proteins, thus making it unavailable for reaction with DNA. At high doses, AITC was positive in other genotoxicity assays indicating DNA damage, and clastogenicity. These were considered as likely due to indirect reactions.

These findings are presented in both the draft and final RCD. According to USEPA guidance (USEPA, 2005), evidence of indirect genotoxicity in the absence of direct genotoxicity is not sufficient to support a mutagenic mode of action. Thus linear extrapolation of tumor incidence data is not appropriate in this case, when there is strong evidence of a non-DNA reactive mode of action.

2. Carcinogenicity, continued: Based on consideration of all the information available, the default approach is to apply a linearized multistage model to derive a cancer potency estimate for each tumor site. For carcinogens that induce tumors at multiple sites in a particular species and sex, US EPA's Benchmark Dose Software (BMDS) can be used to derive maximum likelihood estimates (MLEs) for the parameters of the multisite carcinogenicity model by summing the MLEs for the individual multistage models from the different sites and/or cell types. This multisite model provides a basis for estimating the cumulative risk of carcinogen treatment-related tumors.

DPR Response: As discussed above, AITC is unlikely to be mutagenic under physiological conditions. Weight of evidence analysis for urinary bladder tumors shows strong support for a threshold MOA with urinary bladder hyperplasia as a prerequisite event. A POD that protects against urinary bladder hyperplasia should be sufficient to prevent downstream tumors. Additionally, DPR concluded that undifferentiated leukemia is unlikely to be related to AITC treatment. Subcutaneous fibrosarcoma occurred only at the high dose in one sex. As such, the incidence data for this tumor are not amenable to linear, low-dose extrapolation. Therefore, protecting against urinary bladder hyperplasia, which is the most sensitive effect by oral route and was not observed in repeated inhalation exposure, is likely to protect against the development both of urinary bladder tumors and subcutaneous fibrosarcomas.

3. Extrapolation, Variability and Uncertainty

a. Interspecies Extrapolation and RGDR Approach: … OEHHA agrees with DPR's assumptions and calculations of HECs, yet has some comments on the UFs used to calculate MOEs. For the RGDR approach for non-cancer effects, DPR decreased the conventional interspecies UF of 10 to √10. This is based on the assumption that the RGDR already accounted

for the pharmacokinetic portion of the interspecies factor. OEHHA agrees that if a chemical is causing a portal of entry effect and local metabolism is generally not a concern, the reduction in the pharmacokinetic portion of the UF to a value of 1 is appropriate. However, when the critical effect is systemic in nature, and may involve metabolism, a UF for interspecies pharmacokinetics should be retained with a value of 2 to account for potential uncertainty (OEHHA, 2008). This is especially warranted for AITC due to the absence of pharmacokinetic data following inhalation exposure, and the concern for effects seen in the urinary bladder following oral exposure which are attributed to excretion of AITC-metabolites**.** Thus, this interspecies UF_K of 2 should be considered for all durations of exposure, as the critical effects are considered systemic effects. The total interspecies UF would then be 6, not 3 (rounded).

> **DPR Response:** Dosimetric adjustment for both local and systemic effects with an RGDR of 1 was applied according to guidelines established by US EPA (US EPA, 1994, 2012). As recommended by US EPA and consistent with DPR's current practice, the interspecies UF was reduced from 10 to 3 to acknowledge reduction of the pharmacokinetic UF from 3 to 1. It is important to note that the interspecies UF is not expected to account for the difference between exposure routes. Instead, it accounts for the variability between the animals and humans.

b. Intraspecies Extrapolation: In the draft RCD, a default UF of 10-fold was applied to account for intraspecies variability within the human population (UF_H) … It is OEHHA's opinion that an intraspecies UF of 10 is insufficient as there are many factors affecting human variability in response to a chemical exposure (OEHHA, 2008; Zeise et al. 2013). The scientific basis for this recommendation is detailed in OEHHA's peer reviewed Air Toxics Hot Spots Risk Assessment Guidelines, Technical Support Document for the Derivation of Reference Exposure Levels (OEHHA, 2008). Based on analyses of human pharmacokinetic variability, OEHHA's practice is to increase the traditional intraspecies pharmacokinetic UF of $\sqrt{10}$ to 10. This increase would account for the wide variability in pharmacokinetics in the population, especially among subpopulations such as infants and children, pregnant women, and the elderly. Thus, OEHHA recommends DPR expand their concerns for these subpopulations and increase the intraspecies pharmacokinetic UF to 10, resulting in a total UF_H of 30.

> **DPR Response:** Consistent with current practice, DPR used a default intraspecies UF of 10 that is comprised of a pharmacokinetic UF of 3 and a pharmacodynamic UF of 3 (DPR, 2011). The toxicology data indicated that the most sensitive effect by the oral route was equally protective of young and adult animals in a 2-generation reproductive toxicity study. The data do not support raising the intraspecies pharmacokinetic UF from 3 to 10 at this time.

c. Sensitive Population and Limited Inhalation Toxicity Database: OEHHA recommends an additional UF of √10 be applied to address the limited inhalation toxicity database as there are major data gaps in chronic exposure, oncogenicity, and reproductive and developmental toxicity by the inhalation route, which is the primary route of human exposure. In addition, there is no DNT data by any route on the potential effects of AITC on the developing brains of fetuses, infants and children. Evidence of neurotoxicity was observed as the primary critical effect in the three inhalation toxicity studies and there is evidence that AITC can impact fetuses as indicated by an oral developmental toxicity study of AITC at doses that, when converted to external air concentrations, were similar to the subchronic inhalation POD (Morgareidge, 1973).

> **DPR Response:** To account for the lack of a chronic inhalation study, DPR has included a duration extrapolation factor of 10 to convert the subchronic POD to a chronic POD. With respect to reproductive and developmental toxicity, DPR concluded that fetal or pup effects occurred at similar or higher doses to those eliciting maternal effects. The most sensitive fetal and maternal effects (increased litters with resorption sites, and increased numbers of dead fetuses) were observed in the oral developmental study in mice (LOEL of 28 mg/kg/day and NOEL of 6 mg/kg/day). The equivalent external air concentrations for developmental effects is 9 ppm. Based on the above, DPR's critical subchronic inhalation POD of 5 ppm based on motor activity decrements in rats will be protective of any developmental and maternal effects by the oral route. Based on the data and interpretation of the relevant guidance, DPR does not consider adding an additional UF of 3 to account for lack of inhalation reproductive and developmental toxicity studies to be necessary at this time.

d. Risk Characterization: The Margin of Exposure (MOE) approach was used to evaluate non-cancer hazards … OEHHA recommends a target MOE of 600 for all age groups, occupational and non-occupational, to take into account the recommended higher pharmacokinetic portions of the interspecies (2) and intraspecies (√10) UF's, and an additional UF (√10) to protect potentially sensitive individuals from potential health effects, given the very limited inhalation toxicity database, and to protect fetuses, infants, and children from concern for developmental neurotoxicity.

> **DPR Response:** DPR's rationale for using an interspecies UF of 3, and an intraspecies UF of 10, resulting in a target MOE of 30 is detailed in the responses to OEHHA Comments on Extrapolation, Variability and Uncertainty above. The target MOE was applied for all exposure scenarios involving residential bystanders (children and adults), and workers (adults). In combination with the subchronic-to-chronic duration extrapolation factor of 10 to derive the chronic POD and the dose extrapolation factor of 10 to derive the acute POD, these were considered adequate to account for the uncertainties inherent to this risk assessment.

e. Pesticide illness data: OEHHA suggests that DPR consult the SENSOR program at NIOSH and ask if any AITC-related illnesses associated with soil fumigation have been reported in the US… California does have many reports of MITC-related illnesses and injury that were associated with bystander or re-entry worker exposure. MITC is regulated as a toxic air contaminant. Because AITC and MITC share similar chemical structures and many chemical properties as well as some application methods, there is a concern that AITC may pose a similar health hazard. OEHHA recommends that DPR evaluate this possibility in the draft EAD.

> **DPR Response**: Pesticide illness data related to AITC-containing products was collected from SENSOR (details of SENSOR database are described in the next response). There were six SENSOR reports related to exposure to AITC-containing products, mainly with respect to its use in repellants. There were no reports related to the use of AITC as a fumigant. One adverse effect report to US EPA cited a potential AITC exposure in Florida. These records contained self-reported clinical signs such as eye irritation and pain, cough, respiratory irritation, shortness of breath, and asthma attack/exacerbation.
>
> Physiochemical properties, and exposure related comparisons between AITC and MITC are described in the final EAD (DPR, 2022). AITC and MITC have substantive similarities as well as differences in their toxic effects in animal studies. Both compounds cause irritation at the site of contact (e.g., nasal passages after inhalation exposure, and stomach lining after oral exposure). However, MITC appears to be more toxic to the nasal epithelium than AITC after inhalation exposure in rats (DPR, 2004). Specifically, in a 4-week subchronic rat inhalation study MITC induced nasal epithelial atrophy at 0.3 ppm (LOEL). In contrast, in a 13-week rat inhalation study AITC induced nasal epithelial degeneration at 10 ppm (LOEL) (Randazzo, 2017), indicating that AITC is less irritating than MITC. Therefore, it is appropriate to use AITC toxicity testing data to identify hazard and to characterize risk from exposure to AITC.

B. Exposure Assessment.

1. Off-site Workers and Residential Bystanders.

f. Pesticide Related Illness: The Isagro AITC products have been used outside of California for several years since US EPA approval in 2014. The Sentinel Event Notification System for Occupational Risk (SENSOR) program at NIOSH may have reports of pesticide illness related to AITC use in the 13 other participating states. OEHHA suggests that DPR consult the SENSOR program at NIOSH and ask if any AITC-related illnesses associated with soil fumigation have been reported in the US.

Secondly, California does have many reports of MITC-related illnesses and injury that were associated with bystander or re-entry worker exposure. MITC is regulated as a toxic air contaminant. Because AITC and MITC share similar chemical structures and many chemical

properties as well as some application methods, there is a concern that AITC may pose a similar health hazard. OEHHA recommends that DPR evaluate this possibility in the draft EAD.

DPR Response: The SENSOR data as well as any case reports in the open literature and all relevant adverse effects reports mandatorily submitted to US EPA and DPR pursuant to the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), section (6)(a)(2)) were analyzed when developing the RCD. As of December 2020, there were six SENSOR records related to AITC. Those records did not list the pesticide product(s) being used, but none of them is related to AITC use as a fumigant. It is worth mentioning that there are only 13 US states that participate in the SENSOR pesticide program. Considering the lack of SENSOR records available for AITC fumigant use, this information is not included in the exposure assessment document. There is one Adverse Effect Report available from US EPA (under Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), section (6)(a)(2)) that lists potential AITC exposure in Florida. However, there is not enough detail in the report to evaluate any human health consequences.

As discussed in Appendix 1 of the EAD, MITC compounds (e.g., metam sodium and metam potassium) are often applied using methods different from those used for AITC. In addition, the available MITC data often showed maximum MITC emissions at night, which is different from available AITC emission data which show maximum emissions during the day. As application methods and soil emission are major causes of bystander exposures, DPR does not agree with analyzing MITC-related illness cases as an estimate of possible AITC illness. The remainder of the response to comments regarding pesticide related illnesses is addressed in a separate memorandum on responses to the Exposure Assessment Document.

Comments on Charge Statements – Hazard Identification

Charge Question 1. Acute POD: A default 10x LOAEL-to-NOAEL extrapolation factor was used to establish the critical acute POD of 2.5 ppm.

OEHHA Comment: OEHHA agrees with the use of a dose extrapolation factor of 10, as the critical study included neurobehavioral effects at the lowest dose tested. This default factor is typically applied when extrapolating from a LOAEL to a NOAEL.

DPR Response: No response required.

Charge Question 2. The critical chronic inhalation POD was estimated from the subchronic critical POD by applying a default duration extrapolation factor of 10. This was necessitated by the lack of chronic inhalation studies.

OEHHA Comment: OEHHA concurs with the use of an extrapolation factor of 10 to extrapolate from subchronic to chronic exposures, as a study with a longer exposure duration is not available. However, OEHHA recommends the most health protective studies for each exposure duration are used to derive PODs, after taking into account ADME differences. OEHHA suggests DPR use BMD or other appropriate dose-response evaluation methods to confirm that the most health protective POD is selected from the available inhalation and oral toxicity studies.

> **DPR Response**: In response to OEHHA's suggestion, DPR conducted a BMD analysis of the subchronic oral critical endpoint, then estimated the oral-to-inhalation extrapolated POD, comparing that value to the POD derived from the subchronic inhalation POD. The subchronic inhalation POD of 5 ppm derived from the inhalation study was lower than oral-to-inhalation extrapolated POD of 7.1 ppm (See RCD Section E.1.3) and is therefore protective of the urinary bladder hyperplastic effects seen in oral studies. Ultimately, PODs from oral studies were not used to establish critical PODs

Charge Question 3. PODs from oral studies were not used to establish critical PODs.

OEHHA Comment: While oral toxicity studies were evaluated in the draft RCD and were used to satisfy data requirements to support registration, only inhalation studies were considered when evaluating critical effects and PODs. OEHHA finds this approach problematic. Critical effects identified in the oral toxicity studies were dismissed over "concerns about route specificity of observed effects," yet the inhalation studies available are too limited to adequately characterize chronic, reproductive, or developmental effects resulting from inhalation exposure.

Urinary bladder hyperplasia was a common adverse effect in rats following oral exposure to AITC for various exposure durations, including a two-generation reproductive and developmental study. The draft RCD attributed the development of urinary bladder hyperplasia to sustained high levels of AITC-metabolites in urine (section E.1.7, draft RCD). Because urinary bladder hyperplasia was not found in a single subchronic inhalation study, DPR determined that bladder effects were specific to the oral route. While detailed ADME data following inhalation exposure are not available, it is clear that both oral and IV administration lead to increased AITC-metabolite levels in bladder tissues, particularly in male rats. There is no data on first pass metabolism by the lung, nor data to suggest that the expected route of excretion of AITC metabolites following inhalation is different than following oral exposure. The fact that no urinary bladder hyperplasia was reported in the 13-week inhalation study (Randazzo et al., 2017) could be explained by either the relatively low exposure levels or the short exposure

duration or a combination of both. Thus, OEHHA disagrees that urinary bladder effects are specific to the oral route and are irrelevant to the inhalation route.

DPR Response: DPR carefully considered all available studies with relevant toxicity data in the draft and final RCDs as part of a weight-of-evidence approach. Studies were evaluated at comparable dose levels to ensure the selected inhalation PODs were protective of the observed effects irrespective of the route of exposure. DPR determined that urinary bladder hyperplasia was the critical effect by the oral route in subchronic and chronic durations, then compared the oral PODs for urinary bladder hyperplasia observed in subchronic, chronic, and reproductive toxicity studies against the critical inhalation PODs. This comparative approach using estimated oral-to-inhalation air concentrations demonstrated that the critical subchronic and chronic inhalation PODs were lower than the corresponding oral PODs. Therefore, the selected inhalation PODs are anticipated to protect against all effects observed by oral route.

Although there were no toxicokinetic studies for inhalation route, the well-designed subchronic inhalation study (Randazzo, 2017) revealed inhalation route-specific effects (for example, olfactory epithelial degeneration), and the effects that were observed in oral studies were not observed even up to 25 ppm AITC by the inhalation route. This difference between the routes suggests that exposure to AITC by inhalation does not induce the same effects as occurred by the oral route at the equivalent concentrations. Therefore, the lack of inhalation toxicokinetic data did not influence the determination of critical POD selection, which were selected to be protective of all effects observed by either inhalation or oral exposure.

DPR recognizes that the limited inhalation database constituted the main toxicological uncertainty in this risk assessment. Although there were indications that effects seen in the oral studies were specific to this route, all oral studies (acute, subchronic, and chronic) were evaluated in parallel with inhalation studies in order to identify the most sensitive effect. To ensure that the effects in the oral studies are accounted for, DPR employed a route-to-route extrapolation to generate equivalent external air concentrations from the oral NOELs and compared them to the subchronic and chronic inhalation PODs. As the actual inhalation PODs were lower than those generated by extrapolation, the former were considered protective of effects observed in both the oral and inhalation studies.

Charge Question 4. This RCD did not include a cancer risk estimate for AITC.

OEHHA Comment: The available chronic toxicity/carcinogenicity studies indicate that AITC is an animal carcinogen, and this determination is supported by the induction of urinary bladder tumors, leukemia, and fibrosarcoma in rat oral studies. AITC is a highly reactive compound and can react with protein and DNA in vitro through adduct formation or generation of reactive oxygen species. OEHHA also determined AITC to be genotoxic; this is supported by several positive genotoxic endpoints as summarized in the draft RCD and by IARC (1999). Additional information is provided in the detailed comments. OEHHA suggests DPR quantitatively estimate cancer risk of AITC in its risk assessment.

> **DPR Response:** With respect to the urinary bladder tumors, DPR followed US EPA's cancer guidelines and concluded that AITC is unlikely to act by a mutagenic MOA. Furthermore, DPR identified bladder hyperplasia as a requisite upstream event in a non-mutagenic mode of action for urinary bladder tumors and used it to develop a threshold for urinary bladder tumors, with the assertion that non-hyperplastic doses that prevent hyperplasia will protect from the development of urinary bladder tumor response. Therefore, the chronic RfC derived in the RCD is protective of urinary bladder tumors.
>
> With respect to the undifferentiated leukemia, DPR does not consider these tumors to be AITC treatment related. Detailed discussions were provided earlier in this document (see responses to OEHHA comments on carcinogenicity in this document) and in the final RCD. Finally, due to a lack of quantitative data, subcutaneous fibrosarcoma was not amenable to low-dose, linear extrapolation assessment. For these reasons, DPR did not develop a cancer slope potency.

Comments on Charge Statements – Risk Characterization

Charge Question 7. Dosimetric adjustments of air concentrations to account for pharmacokinetic differences between laboratory animals and humans were used to calculate reference concentrations (RfCs) and risk targets (i.e., target Margins of Exposure).

OEHHA Comment: OEHHA supports the use of the RGDR approach to convert doses in animal inhalation experiments to human equivalent concentrations (HEC) for non-cancer effects. However, for effects that are systemic in nature, it is OEHHA's position that the RGDR approach does not account for interspecies differences in metabolism or excretion (see additional discussion in detailed comments). Therefore, OEHHA recommends retaining an interspecies pharmacokinetic UF of 2, resulting in a total interspecies UF of 6, rather than 3 as presented in the draft RCD.

DPR Response: Dosimetric adjustment was applied according to guidelines developed by US EPA (US EPA, 1994 and 2012) for both local and systemic effects. The resultant RGDR was 1. As recommended by US EPA and according to DPR's current practice, the interspecies UF was reduced from 10 to 3 since the default dosimetric adjustment accounts for variability in pharmacokinetic handling.

Comments on Charge Statements – Worker and Bystander Margins of Exposure

Charge Question 8. Risks to on-site workers were estimated for acute (short term), subchronic (seasonal) and chronic (annual, lifetime) exposures.

OEHHA Comment: OEHHA agrees with the chosen durations to estimate occupational risks of on-site workers in the draft RCD, and noted that many occupational exposure scenarios are far below DPR's target MOE of 30. As noted in the Risk Characterization section, OEHHA suggests a target MOE of 600.

DPR Response: See DPR's responses to OEHHA comments in the Extrapolation, Variability and Uncertainty of this document.

Charge Question 9. Risk to off-site workers and residential bystanders, were estimated for acute exposures.

OEHHA Comment: Based on the proposed uses of AITC and its toxicological properties, OEHHA recommends estimates for seasonal, annual, and lifetime exposures of off-site workers and residential bystanders be included in the assessment. It is of concern to OEHHA that all the acute exposure scenarios for off-site workers and residential bystanders, including children, were below the draft RCD's target MOE of 30, and would be well below OEHHA's suggested target MOE of 600.

DPR Response: See DPR's responses to OEHHA comments in the Extrapolation, Variability and Uncertainty of this document. See further discussion in the separate response memorandum addressing comment on the exposure assessment.

Minor Comments – Risk Assessment

A. Draft RCD

Table 5 on page 28: body weight percent change for females at 25 ppm is incorrect; it should be 12% rather than 125%.

DPR Response: The correction is noted in the final RCD.

Notation for Table 6 does not match the footnotes of the table. There are mixed letters and numbers contained in the table, but only letters are listed in the footnotes.

DPR Response: The correction is noted in the final RCD.

Reference at the top of page 31 for Lewerenz et al, 1988a is incorrect. Decreased total cholesterol was observed in Hasamura et al, 2011.

DPR Response: The correction is noted in the final RCD.

The Estimated AITC Dose in mg/kg-day differs between Tables 12 and 13. Table 12 shows the calculated AITC intake whereas Table 13 lists the estimated HRE intake. Values for Table 13 Estimated AITC Dose (mg/kg-day) should be 0, 2.2, 4.4, and 16.8, assuming 82% AITC content in the HRE used (as assumed when calculating estimated AITC dose in Table 12).

DPR Response: The correction is noted and additional details regarding the average HRE intake to study methods are now included in the final RCD for traceability.

Regarding the genotoxicity evidence, the draft RCD did not include some positive studies summarized by IARC (1999). OEHHA suggests including the following in the genotoxicity section.

- Reverse mutation in Escherichia coli WP67 (Rihová, 1982, as cited in IARC, 1999);
- Chromosomal aberrations in Allium cepa (Sharma and Sharma, 1962, as cited in IARC, 1999);
- Drosophila melanogaster sex-linked recessive lethal mutations (Auerbach and Robson, 1944 and 1947, as cited in IARC, 1999);
- The summary for Tripathi et al. (2015, as cited in the draft RCD) is missing the induction of gamma-H2AX, a marker for DNA damage and/or double-strand breaks.

DPR Response: Additional citations are now included in the summary table of genotoxicity in the final RCD as appropriate. Allium cepa (commonly referred to as the bulb onion), which was the primary organism of interest in Sharma and Sharma (1962), is not relevant for the AITC human health risk assessment. Auerbach and Robson (1944) and Auerbach and Robson (1946) report identical data. A summary of the most complete report, Auerbach and Robson (1946), has been added to the summary table in the final RCD. The induction of γH2AX is now included in the Tripathi et al (2015) study summary.

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