OFFICE OF ENVIRONMENTAL HEALTH HAZARD ASSESSMENT

Notification Level Recommendation

Perfluorobutane Sulfonic Acid in Drinking Water

January 2021

Pesticide and Environmental Toxicology Branch
Office of Environmental Health Hazard Assessment
California Environmental Protection Agency

Notification Level Recommendation for Perfluorobutane Sulfonic Acid (PFBS) in Drinking Water

Prepared by

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SUMMARY

This document presents the notification level (NL) recommendation by the Office of Environmental Health Hazard Assessment (OEHHA) for perfluorobutane sulfonic acid (PFBS) in drinking water. NLs are health-based advisory levels established by the Division of Drinking Water of the State Water Resources Control Board (SWRCB) for chemicals in drinking water that lack regulatory or maximum contaminant levels (MCLs). When a risk assessment for a chemical of concern in drinking water is lacking, SWRCB may request that OEHHA develop a risk assessment for that chemical and derive a health-protective concentration that can be used to establish an NL. Health-protective concentrations are based on the most sensitive, well-conducted and scientifically valid toxicology studies. In developing health-protective concentrations, OEHHA considers the publicly available studies in humans and laboratory animals, as well as in vitro studies of toxicokinetics and mechanisms of toxicity.

OEHHA developed a drinking water health-protective concentration of 0.5 micrograms per liter (µg/L), equivalent to 0.5 parts per billion (ppb) for noncancer effects of PFBS, based on reduction of the thyroid hormone, thyroxine (T4), in pregnant female mice on gestation day 20 (GD20) (Feng et al., 2017). There were insufficient data to evaluate the potential carcinogenicity of PFBS.

OEHHA recommends that SWRCB set the NL for PFBS in drinking water at 0.5 ppb.

INTRODUCTION

At the request of SWRCB, OEHHA has developed a recommendation for a drinking water NL for PFBS. Health and Safety Code Section 116455 defines an NL as the level of a drinking water contaminant that SWRCB has determined, based on available scientific information, does not pose a significant health risk but, when exceeded, warrants notification to a water system's governing body and other specified entities. NLs are nonregulatory, health-based advisory levels that SWRCB establishes as a precautionary measure for contaminants for which regulatory standards have not been set but that may be considered candidates for the establishment of an MCL.

This document reflects OEHHA's focused review of the human and animal toxicity database identified from the open literature and from references cited in the US Environmental Protection Agency's draft human health toxicity evaluation for PFBS (US EPA, 2018a).[1]

BACKGROUND

Perfluorobutane sulfonic acid (PFBS; CASRN 375-73-5) and its related salt (potassium perfluorobutane sulfonate or K^+PFBS; CASRN 29420-49-3) are four-carbon

[1] Supporting documentation and data are publicly available on the Health Assessment Workspace Collaborative (HAWC), https://hawcprd.epa.gov/assessment/100000037/

fluorocarbons with a sulfonic acid functional group that act as anionic surfactants, and are used in numerous commercial products for their water- and stain-repellent properties. They are members of a large class of chemicals known as per- and polyfluoroalkyl substances (PFAS). PFBS was originally manufactured as a replacement for the environmentally and biologically persistent and toxic eight-carbon chain PFAS, perfluorooctane sulfonate (PFOS) (Lieder et al., 2009a). The motivation was that because of their shorter carbon chains and more rapid elimination in animals, certain perfluoroalkylsulfonates like PFBS would present less of a human health risk with less bioaccumulation than the eight-carbon PFAS (Bogdanska et al., 2014). High PFBS levels have been measured in water (including surface, ground, and drinking waters) in Germany, China and Antarctica (Feng et al., 2017). PFBS is expected to eventually replace PFOS as a major perfluorinated environmental contaminant, and has already been detected in human populations (ECHA, 2019). Furthermore, since PFBS does not appear to biodegrade, it is expected that the environmental concentrations will continue to rise, resulting in greater potential for human exposure.

In April 2019, SWRCB ordered potential PFAS source locations (airports, landfills and adjacent water systems) to sample public water supply (PWS) wells quarterly for four consecutive quarters. SWRCB received PFAS monitoring data from over 600 water system sites adjacent to nearly 250 airports with fire training areas and municipal solid waste landfills within California. The results of the PWS wells sampled during the first four quarters are now available to the public.[2] These results indicate that PFBS is the 4th most frequently detected of the PFAS tested, and in general PFBS detections and concentrations were increasing over the time period analyzed in the PWS wells. With the detections and concentrations of PFBS potentially increasing in CA drinking water sources, the need for health-protective levels of PFBS and other short-chain PFAS in drinking water has become apparent.

Various agencies both within and outside of the US have developed reference levels for PFBS, some specifically for human exposure in drinking water. US EPA (2018a) does not have a reference concentration for groundwater, but has developed human health toxicity values (i.e., subchronic and chronic reference doses or RfDs) in a public review draft. These RfDs are meant to eventually be used to develop "health protective levels for chemicals in water, soil, and other media" or regulatory standards for drinking water under the Safe Drinking Water Act (SDWA) (US EPA, 2018b). This public review draft provides evidence integration and hazard characterization summaries for PFBS and K^+PFBS. The RfDs were developed from studies by Feng et al. (2017) and Lieder et al. (2009b), with additional reports supporting these as critical studies. The draft subchronic and chronic RfDs were developed as follows: subchronic RfDs of 0.04 mg/kg-day (based on thyroid effects) or 0.1 mg/kg-day (based on kidney effects); chronic RfDs of 0.01 mg/kg-day (based on thyroid effects) or 0.01 mg/kg-day (based on kidney effects). Prior to the draft toxicity assessment of 2018, US EPA developed provisional peer-reviewed toxicity values (PPRTVs) for PFBS and K^+PFBS (US EPA, 2014). The PPRTVs were based on kidney hyperplasia in female rats (Lieder et al.,

[2] https://www.waterboards.ca.gov/pfas/drinking_water.html, accessed August 2020

2009a). Subchronic and chronic provisional reference doses (or p-RfDs) were 0.2 and 0.02 mg/kg-day, respectively.

Based on the PFBS detected in groundwater and other available information, the Minnesota Department of Health (MDH) developed a chronic noncancer health-based value ($nHBV_{chronic}$) of 2 ppb (MDH, 2017). This value was based on epithelial hyperplasia in the kidneys of dams in the two-generation study by Lieder et al. (2009b) and York (2003b, unpublished study as cited in MDH, 2017). Also, an $nHBV_{subchronic}$ of 3 ppb was developed based on the same endpoint, and an $nHBV_{short\text{-}term}$ of 3 ppb was developed based on developmental effects in a mouse study by Feng et al. (2017).

The Michigan Science Advisory Workgroup set an MCL of 0.42 ppb for PFBS in municipal drinking water in Michigan,[3] based on thyroid effects observed in a developmental toxicity study by Feng et al. (2017) (MSAW, 2019). The workgroup evaluated effects of PFBS on the thyroid and the kidney as the most sensitive toxicological effects in animal studies, and concluded that the thyroid effects had greater functional significance for deriving their health-based value in drinking water.

The US Assistant Secretary of Defense issued a memorandum in 2018 outlining perfluorooctanoic acid (PFOA), PFOS, and PFBS regional screening levels (RSLs) to be used for site specific risk assessments (McMahon, 2019). The RSLs for PFBS were calculated using the US EPA PPRTV (chronic p-RfD) of 0.02 mg/kg-day. The RSLs for residential tap water were 400 ppb using a hazard quotient (HQ) of 1 for PFBS alone, and 40 ppb using an HQ of 0.1 when other PFAS are present (McMahon, 2019).

The European Chemicals Agency (ECHA) also tabulated no-observed-adverse-effect levels (NOAELs) and lowest-observed-adverse-effect levels (LOAELs), using the same studies used by US EPA (2018a), in its overview of PFBS-induced health effects in consideration of identifying PFBS as a substance of very high concern for probable serious effects to the environment and human health (ECHA, 2019).

SYSTEMATIC LITERATURE SEARCH

OEHHA conducted an initial systematic literature search in December 2019 of multiple open literature databases (PubMed, Embase, Scopus, and Toxnet) using a search string intended to identify all studies that mention PFBS in the title or abstract. The search terms used for each database are presented in Appendix I. If PFBS was not named in the title or abstract, it was not captured in the search.

From the initial search, OEHHA identified 684 individual studies that met the search criteria. OEHHA uploaded the identified references into DistillerSR systematic review software and conducted inclusion/exclusion screening for relevant toxicological data against a PECO (populations, exposures, comparators, and outcomes) statement designed to capture relevant toxicological data (Appendix I). Two independent

[3] https://www.michigan.gov/egle/0,9429,7-135--534660--,00.html, accessed August 2020.

reviewers conducted both Tier 1 (title/abstract) and Tier 2 (full-text) reference evaluation against the PECO statement. Tier 1 screening resulted in 112 individual references identified, and Tier 2 resulted in the exclusion of 20 references. References were categorized as animal toxicity studies, human epidemiology and/or biomonitoring studies, toxicokinetics studies, or mechanistic studies. Five studies met the criteria for more than one category. During study evaluation, as studies were identified that were not captured in the original literature search, or as new studies became available after the date of the original literature search, they were added to OEHHA's reference library and evaluated. OEHHA also cross checked search results with the studies listed in HAWC and added any missing studies to OEHHA's reference library.

TOXICOLOGICAL REVIEW

OEHHA conducted a focused review of the toxicological database. The studies and data presented here are not a complete summary of the entire database, but a targeted selection of studies with human relevance and animal toxicity studies with effects in the lowest doses in the database that can be considered for health-protective concentration derivation.

Toxicological Effects in Humans

OEHHA evaluated the epidemiological literature on PFBS and adverse health outcomes in humans. These studies included both adults and children, and several identified some associations with reproductive, developmental, immunological, and cardiovascular endpoints. Several additional epidemiological studies related to PFBS, summarized in the HAWC[4] database, were largely negative. For example, no associations were observed between PFBS levels and premature ovarian insufficiency in women (Zhang et al., 2018), fetal testosterone or estradiol concentration, as measured in cord blood samples at birth (Yao et al., 2019) or later in adolescents (Zhou et al., 2016), or measures of hyperuricemia in children (Qin et al., 2016). The HAWC database, last updated in October of 2019, provides summarized data for these studies. Here, several epidemiological studies showing significant associations with PFBS exposures are summarized.

In a study examining male reproductive endpoints, Song et al. (2018) collected semen and blood samples from male volunteers at an infertility clinic in Guangdong, China. Samples were assayed for PFAS levels and semen quality. PFBS was detected in more than 90% of samples. Detected levels ranged from 0.056 to 0.43 ng/ml in blood, and in semen, levels ranged from undetectable to 0.28 ng/ml. No correlations between blood and semen samples were found for PFBS, however. In this study, PFBS levels in blood were negatively associated with body mass index (BMI; $r=-0.33$, $p<0.01$), while PFBS levels in semen were positively associated with BMI ($r=0.328$, $p<0.01$). PFBS

[4] https://hawcprd.epa.gov/study/assessment/100000037/, accessed August 2020

levels in semen were positively associated with progressive motility in sperm (r=0.195, p=0.048).

Wang et al. (2017) recruited a total of 157 women, 20-45 years of age, to participate in a study based on a diagnosis of endometriosis related infertility. An additional 178 women experiencing non-endocrine related infertility, with no history of endometriosis, were recruited as controls. PFBS was detected in 98.5% of plasma samples collected from both study groups, with detected levels ranging from <0.006 to 0.086 ng/ml in the first (reference) tertile, >0.086 to 0.094 ng/ml in the second tertile, and >0.094 to 1.25 ng/ml in the third tertile. Women with endometriosis related infertility had higher median levels of PFBS ($p<0.001$) as compared to controls. PFBS plasma levels were significantly associated with an increased risk of endometriosis related infertility (odds ratio (OR)=3.74, 95% confidence interval (CI): 2.04-6.84 for the 2nd tertile and OR=3.04, 95% CI: 1.65-5.57 for the 3rd tertile, as compared to the 1st tertile reference values, p=0.001; adjusted for age, BMI, household income, and education). In this study, the control group had no self-reported prior history of endometriosis related infertility; however, examinations were not performed at the time of sample collection, thus it is uncertain if any of the control subjects had asymptomatic endometriosis as a contributing factor to their infertility. Additionally, PFBS plasma levels were collected after onset and diagnosis of endometriosis in the study group, so PFBS levels at the onset of symptoms are unknown. Finally, the measured levels of PFBS in plasma, while above the limit of detection (LOD), were very low, requiring the study authors to run an additional calibration curve.

In a study examining risks for hypertensive disorders during pregnancy, cord blood samples collected from 674 women in a Shanghai Hospital in 2011-2012 were assayed for total PFAS content (Huang et al., 2019b). A previous study by this group found that PFBS levels in cord blood were an accurate representation for PFBS levels in maternal blood. In this study, PFBS concentrations in blood ranged from 0.037 ng/ml in the 25th percentile, to 0.061 ng/ml in the 75th percentile. Samples below the LOD (0.0045 ng/ml) were assigned values of half the LOD. After adjusting for age, education, pre-pregnancy BMI, and parity, one unit increase in standardized PFBS concentration in cord blood was associated with a higher risk of hypertensive disorders of pregnancy (OR=1.64, 95% CI: 1.09-2.47, p-value for linear trend=0.03) and preeclampsia (OR=1.81, 95% CI: 1.03-3.17, p-value for linear trend=0.05). However, participants in this study had a prior diagnosis of maternal hypertensive disorders, and PFBS levels in cord blood at birth may not be representative of PFBS levels earlier in pregnancy, at the onset of symptoms.

A positive correlation between PFAS levels in cord blood and adiposity measurements at age 5 years was observed in a cohort of 404 children (Chen et al., 2019). The mean cord plasma PFBS level was 0.05 ng/ml (±0.03 standard deviation or SD), the median was 0.05 ng/ml, and the range was <0.009-0.39 ng/ml. In linear regression models (adjusted for maternal age, pre-pregnancy BMI, gestational week at delivery, maternal education, paternal smoking during pregnancy, and parity) associations between cord

plasma concentrations of PFBS and waist circumference (β=2.06, 95% CI: 0.43-3.68), fat mass (β=0.79, 95% CI: 0.08-1.51), body fat percentage (β=2.84, 95% CI: 0.29-5.39), and waist to height ratios (β=0.01, 95% CI: 0.0008-0.03) were seen in females only between tertiles 3 and 1. These associations between prenatal exposure and adiposity at age 5 years were statistically significant. It is not possible to determine whether postnatal exposure affected the measurements in the children because the study did not evaluate post-natal PFAS exposure in these children. A prior study in the same cohort of children found no association between PFBS levels in cord plasma and atopic dermatitis in either females or males at 24 months of age (Chen et al., 2018).

Several studies were based on a cohort of 231 asthmatic children, 10-15 years of age, recruited in 2009-2010 to examine the association between PFAS levels in blood and immunological biomarkers (Dong et al., 2013; Zhu et al., 2016). A control group of 225 non-asthmatic children was included in the study. The study participants were recruited from a hospital setting while the control subjects were recruited from public schools from the same region. Mean serum levels of PFBS were significantly higher in children with asthma as compared to children without asthma (0.53±0.20 SD and 0.48±0.20 SD ng/ml, respectively; p=0.022); however median values were the same for both groups (0.48 ng/ml). In the Zhu et al. (2016) study, asthma was associated with higher PFBS serum concentrations, with a stronger association in males versus females, with an adjusted OR for the highest versus lowest quartiles in males of 2.59 (95% CI, 1.14-5.87). Overall, levels of interferon-γ (IFN-γ; a T-helper 1-type cytokine) were lower in children with asthma, and a significant positive association was detected in male asthmatics between PFBS and interleukin-4 (IL-4)/IFN-γ ratio (Spearman correlation coefficient r=0.196, p=0.013). While males appeared more sensitive in this study, the female children with asthma numbered less than half of the males (73 and 158, respectively). Dong et al. (2013) found increased absolute eosinophil counts between the first and fourth quartiles of serum PFBS concentrations within the asthmatic group (p=0.009).

A follow-up paper (Zhou et al., 2017), using the same population of children with asthma, found an interaction effect between asthmatic status and PFBS levels in serum on urinary levels of CC16 (Clara cell secretory protein 16), a surfactant protein and biomarker for asthma (β=-0.127, 95% CI: -0.236, -0.018; interaction p-value=0.023). However, the study authors did not correct for additional environmental factors such as rural versus urban residences, or residential region and corresponding air pollution level for urban dwelling children. The control cohort was used for an additional study examining lipid levels in children. A marginally positive association (p=0.04) between serum levels of PFBS and total cholesterol was observed (Zeng et al., 2015).

A single epidemiological study was identified that examined the relationship between PFBS and cardiovascular disease (CVD). Huang et al. (2018) evaluated data collected from 10,859 participants from the NHANES (National Health and Nutritional Examination Survey) surveys from 1999 to 2014 for associations between total perfluoroalkyl chemicals (PFCs) and CVD. Only 30.05% of the study population had

PFBS serum levels above the LOD, with 5th-95th percentile values ranging from 0.07 to 0.30 ng/ml. PFBS was statistically significantly associated with total incidence of CVD (OR=1.19, 95% CI: 1.06-1.32 for quartile 2 versus quartile 1), but was not significantly associated with any individual CVD such as congestive heart failure, heart attack, or stroke. The association of PFBS with total CVD outcomes remained statistically significant after adjusting for additional factors including BMI, diabetes, family history of CVD, and cholesterol levels (OR=1.34, 95% CI: 1.05 – 1.72 for quartile 2 versus quartile 1).

The epidemiological database for PFBS presents some evidence of reproductive, developmental, immunotoxic, and cardiovascular effects in humans associated with PFBS exposure; however, the data were not sufficient to conduct a quantitative dose-response assessment for health-protective concentration derivation.

Toxicological Effects in Animals

OEHHA conducted an initial review of the animal toxicity database from the 2018 draft assessment of PFBS by US EPA and its supporting documentation available in HAWC. OEHHA determined that the toxicity review completed by US EPA for its draft human health toxicity values assessment for PFBS (US EPA, 2018a) was of high quality, complete, and would aid in conducting an expedited review of PFBS for an NL recommendation. OEHHA also performed a systematic literature search as detailed in Appendix I to capture any potential studies that may have been missed by the search done by US EPA, or published after its draft assessment. It should be noted that the data from the NTP 28-day toxicity study in rats, which was not peer reviewed at the time of release of the US EPA draft assessment (cited as NTP, 2018 in US EPA, 2018a), has since been published in a completed report and is described here as NTP (2019). For the purpose of deriving a health-protective concentration, OEHHA focused on studies that would provide reliable dose-response information in the low-dose region. From the available animal toxicity studies, OEHHA identified four candidate critical studies and conducted an in-depth review and assessment of these studies for candidate points of departure (PODs).[5] These four studies were determined to be of acceptable quality, adequate data reporting, and sufficient sensitivity for health-protective concentration derivation and are included in Table 1. They included two subchronic oral studies, a two-generation reproductive toxicity study in rats, and a developmental toxicity study conducted in female mice (NTP, 2019; Lieder et al., 2009a; Lieder et al., 2009b; Feng et al., 2017). A detailed summary of each study is included in the following sections. Extracted data from all animal toxicity studies and modeling for all data are also publically available in US EPA's evaluation in HAWC. While OEHHA used some of the data available in HAWC, all interpretation of results and modeling of dose-response data presented in this report for the candidate critical studies were performed by OEHHA using health-protective assumptions and approaches.

[5] Detailed summaries of studies not described in this document can be found in US EPA (2018a).

Table 1. Summary of candidate critical studies and endpoints in animals

Species/Sex	Exposure	Endpoints	NOAEL/LOAEL/BMDL (mg/kg-day)	Reference
Sprague Dawley rats (10/sex/dose)	0, 62.6, 125, 250, 500, or 1,000 mg/kg-day by gavage (½ dose twice daily) for 28 days	↑ absolute and relative liver and kidney weights; changes in clinical chemistry; ↓ thyroid hormone levels; treatment related mortality at the highest dose	LOAEL: 62.6 for ↓ thyroid hormone levels in both sexes; ↑ liver weight in males; ↑ kidney weight in females $BMDL_{1SD}$: 6.9 for ↓ tT4 in females	NTP, 2019
Crl:CD rats (10/sex/dose)	0, 60, 200, or 600 mg/kg-day by gavage for 90 days	↑ incidence of hyperplasia in the kidney	NOAEL: 200 $BMDL_{05}$: 38.5 for kidney hyperplasia in females	Lieder et al., 2009a
Crl:CD rats (30/sex/dose)	0, 30, 100, 300, or 1,000 mg/kg-day by gavage[1]	Non-reproductive endpoints **P and F1 (both sexes):** kidney papillary epithelial hyperplasia; **P and F1 (males):** hepatocellular hypertrophy Reproductive endpoints (males) **P:** ↓ sperm count at highest dose; **F1:** abnormal sperm morphology at highest dose; ↑ days to preputial separation at lowest and highest dose	LOAEL: 300 for kidney hyperplasia and hepatocellular hypertrophy[2] $BMDL_{05}$: 37.4 for kidney hyperplasia in P generation males	Lieder et al., 2009b
IRC mice 30 dams/dose; see Table 5 for numbers of animals examined for each endpoint	0, 50, 200 or 500 mg/kg-day from GD1 to GD20 by gavage	**Dams:** ↓thyroid hormone levels **Pups:** ↓ thyroid hormone levels at PND1, PND30, and PND60; delays in eye opening, vaginal opening, and time to first estrus	NOAEL: 50 for ↓ thyroid hormone levels in dams and pups $BMDL_{1SD}$: 22.1 for ↓ tT4 in GD20 dams	Feng et al., 2017

Abbreviations: NOAEL, no-observed-adverse-effect level; LOAEL, lowest-observed-adverse-effect level; GD, gestation day; PND, postnatal day. All BMD results modeled by OEHHA.

[1] Both sexes were exposed from 10 weeks prior to mating; females were exposed during gestation and

lactation; F1 pups were exposed beginning at weaning, similar to parental (P) generation; F2 pups were not dosed directly.

[2] The study authors noted a NOAEL of 100 mg/kg-day for these endpoints in the Discussion section of the study; however, the incidence and severity of these endpoints at 30 and 100 mg/kg-day were not reported in their data table (Table 10 of the publication). Without data for these lower dose groups, a NOAEL cannot be determined with certainty, thus OEHHA identified 300 mg/kg-day as the LOAEL.

Short-Term and Subchronic Toxicity

In a short-term oral toxicity study, ten male and ten female Sprague Dawley rats per dose were gavaged daily for 28 days with 0, 62.6, 125, 250, 500 or 1,000 mg/kg-day PFBS (NTP, 2019). Treatment had no effect on mortality or significant effects on body weight up to 500 mg/kg-day. Most of the animals in the 1,000 mg/kg-day group died as a result of treatment, and were not included in the analysis. Relative liver weight was increased at all dose groups in males, and at 125 mg/kg-day in females. Relative kidney weight was also significantly increased in females at all dose groups, and at 500 mg/kg-day in males. Various clinical chemistry measurements related to kidney and liver toxicity were changed in the 250 and 500 mg/kg-day groups. The most significant effects of PFBS treatment were decreased total thyroxine (tT4), free thyroxine (fT4), and total triiodothyronine (T3) concentrations in all male and female dose groups (Table 2). Thyroid stimulating hormone (TSH) levels were unchanged. Because the lowest dose in the study caused statistically significant changes in thyroid hormone levels, OEHHA determined 62.6 mg/kg-day to be the study LOAEL. Applying a factor of 10 to extrapolate from the LOAEL to a NOAEL would result in a NOAEL of 6.3 mg/kg-day.

Table 2. Thyroid hormone levels in rats following 28-day oral exposure (NTP, 2019)[a]

Dose PFBS (mg/kg-day)	**0**	**62.6**	**125**	**250**	**500**
Males (n)	**9**	**10**	**10**	**10**	**9**
Total T4 (µg/dl)	3.34 ± 0.54	0.90 ± 0.28**	0.22 ± 0.16**	0.10 ± 0.09**	0.29 ± 0.21*
Free T4 (ng/dl)	2.09 ± 0.27	0.64 ± 0.13**	0.32 ± 0.03**	0.30 ± 0.00**	0.30 ± 0.00*
Total T3 (ng/dl)	117.76 ± 24.93	87.85 ± 15.81*	64.48 ± 9.74**	60.20 ± 12.97**	50.44 ± 1.32**
TSH (ng/ml)	23.27 ± 8.37	26.64 ± 12.43	25.51 ± 8.00	23.76 ± 8.16	32.19 ± 9.45
Females (n)	**10**	**10**	**10**	**9**	**9**
Total T4 (µg/dl)	3.10 ± 0.47	1.48 ± 0.28**	1.12 ± 0.16**	0.90 ± 0.24**	0.97 ± 0.27*
Free T4 (ng/dl)	1.54 ± 0.25	0.72 ± 0.16**	0.55 ± 0.09**	0.48 ± 0.12**	0.36 ± 0.09*
Total T3 (ng/dl)	89.29 ± 17.61	61.81 ± 10.56**	61.53 ± 10.53**	52.37 ± 5.61**	51.28 ± 2.61*

Dose PFBS (mg/kg-day)	0	62.6	125	250	500
TSH (ng/ml)	11.92 ± 4.08	14.56 ± 3.35	12.55 ± 3.35	14.40 ± 3.45	13.76 ± 2.97

[a] Presented as mean ± standard deviation (standard deviation calculated by OEHHA from standard error reported by study authors)
* p ≤0.05; ** p ≤0.01, significantly different from the vehicle control group by Dunn's or Shirley's test as reported by study authors

It is OEHHA's policy to determine the point of departure (POD) from a toxicity study by fitting a dose-response model to the data using the US EPA Benchmark Dose Software[6] (BMDS version 2.7) when possible. BMDS uses mathematical models to fit data and determines the dose (benchmark dose or BMD) that corresponds to a pre-determined level of response (benchmark response or BMR). Typically OEHHA uses a BMR of 5% above the background or the response of the control group for dichotomous data. For continuous data, a BMR of 1 standard deviation (SD) from the control mean is typically used when there are no data to indicate what level of response is biologically significant. For thyroid hormone level changes in utero, US EPA used a BMR of 20% relative deviation (RD) from the control mean, citing a body of literature supporting neurodevelopmental effects of thyroid hormone deficiency during gestation in animal models at 10-30% depression of maternal tT4 (US EPA, 2018a). The rationale for not choosing a lower BMR was that a 10% change often fell within normal experiment-to-experiment variation between controls. However, OEHHA chose not to use 20% as a BMR, as developmental effects were noted in some studies cited by US EPA at less than 20% depression of tT4. Since there is uncertainty in what level of tT4 decrease is considered biologically adverse in animal models, OEHHA modeled the data for thyroid hormones using a BMR of 1 SD from the control mean. To account for uncertainty in the data, the model also calculates the 95% lower confidence limit of the BMD, known as the BMDL (L stands for lower confidence limit). While most endpoints from the NTP study were not amenable to BMD modeling, the data for tT4 in female rats were modeled. The model with the best fit (visual fit, lowest Akaike information criterion (AIC), and significant p-values for tests 1-4) was exponential M4, which returned a $BMDL_{1SD}$ of 6.9 mg/kg-day. BMD results for the selected model can be found in Appendix II (Figure A1). The $BMDL_{1SD}$ of 6.9 mg/kg-day is similar to the estimated study NOAEL of 6.3 mg/kg-day, and was considered as a candidate POD for health-protective concentration derivation.

In a subchronic oral toxicity study, Crl:CD rats (10/sex/dose) were gavaged daily with the potassium salt of PFBS (K^+PFBS) at doses of 0, 60, 200 and 600 mg/kg-day for 90 days (Lieder et al., 2009a). No treatment related mortality, body weight or neurotoxic effects were noted. Red blood cell counts, hemoglobin, and hematocrit values were reduced in males receiving ≥200 mg/kg-day. Total protein and albumin were lower in females at 600 mg/kg-day. Microscopic changes, consisting of hyperplasia with some necrosis of the mucosa and some squamous metaplasia, were observed in the stomach of animals in the highest dose group. Histopathological changes in the kidney were

[6] Available at: https://www.epa.gov/bmds

minimal to mild hyperplasia of the epithelial cells in the medullary and papillary tubules and ducts in the inner medullary region. The study authors also noted, "Microscopic changes of an equivocal and uncertain nature were observed in the nasal mucosa and were likely attributable to the route of dosing (oral gavage)."

Statistically significant findings for this 90-day study are summarized in Table 3. In male rats, the absolute and relative spleen weight were significantly reduced at ≥60 mg/kg-day compared to the control group; however, there was no dose-response trend in this reduction across the dose range. Clinical chemistry measurements showed that the average value of chloride was significantly increased in male rats at 600 mg/kg-day. The average total protein and albumin values were significantly reduced in female rats at the same dose. Hematology results showed average red blood cell numbers and hemoglobin values at 600 mg/kg-day were statistically significantly reduced and hematocrit was reduced at ≥200 mg/kg-day. Microscopic changes were observed in the kidneys and stomach of male and female rats at 600 mg/kg-day with increased incidence of hyperplasia of the epithelial cells of the medullary and papillary tubules and ducts in the inner medullary regions compared to the control group. The effects observed in the stomach were likely due to local irritation from the repeated gavage treatment. The kidney effects were considered treatment related. The NOAEL for females and males was 200 mg/kg-day based on the observed histological effects in the kidney. OEHHA conducted BMD modeling of this endpoint using a BMR of 5%. The $BMDL_{05}$ values calculated were 38.5 mg/kg-day for females and 48.3 mg/kg-day for males. BMD modeling results can be found in Appendix II (Figures A2 and A3, respectively). These effects were not the most sensitive endpoints from the available animal toxicity studies, and were therefore not considered for health-protective concentration derivation.

Table 3. Statistically significant findings in rats following 90-day oral exposure to PFBS (Lieder et al., 2009a)

Dose (mg/kg-day)	**0**	**60**	**200**	**600**
Male Results (# animals/group)	**10**	**10**	**10**	**9**
Absolute spleen weight (g)	0.93 ± 0.13	0.77 ± 0.10**	0.83 ± 0.06*	0.80 ± 0.11**
Relative spleen weight (g)	0.181 ± 0.018	0.158 ± 0.015**	0.172 ± 0.017	0.163 ± 0.020*
Chloride (mmol/L)	98 ± 2.0	100 ± 1.2	100 ± 1.4	101 ± 1.7**
Red blood cell (10^6/mm^3)	7.76 ± 0.469	7.62 ± 0.443	7.55 ± 0.282	7.19 ± 0.481*
Hemoglobin (g/dl)	16.4 ± 0.96	16.0 ± 0.41	15.6 ± 0.48	15.5 ± 0.78*
Hematocrit (%)	44.2 ± 2.32	42.7 ± 1.44	41.9 ± 1.50*	40.9 ± 2.24**
Kidney, hyperplasia, tubular/ductular epithelium papilla	1/10	0/10	1/10	8/10^^

Dose (mg/kg-day)	0	60	200	600
Kidney, edema, focal papillary (both kidney)	0/10	0/10	0/10	3/10
Stomach, necrosis, individual cells in limiting ridge	0/10	2/10	2/10	8/10^^
Stomach, hyperplasia/hyperkeratosis, limiting ridge	0/10	0/10	0/10	5/10^
Dose (mg/kg-day)	**0**	**60**	**200**	**600**
Female Results (# animals/group)	**10**	**10**	**10**	**9**
Total Protein (g/dl)	7.2 ± 0.40	7.2 ± 0.34	7.1 ± 0.40	6.7 ± 0.23*
Albumin (g/dl)	4.9 ± 0.38	4.8 ± 0.29	4.7 ± 0.31	4.4 ± 0.23*
Kidney, hyperplasia, tubular/ductular epithelium papilla	0/10	0/10	1/10	6/10^^
Kidney, edema, focal papillary (both kidneys)	0/10	0/10	0/10	3/10
Stomach, necrosis, individual cells in limiting ridge	1/10	0/10	1/10	9/10^^
Stomach, hyperplasia/hyperkeratosis, limiting ridge	0/10	0/10	0/10	7/10^^

Mean ± standard deviation (SD);
Significantly different from control (* p≤0.05; ** p≤0.01) as reported by study authors
Significantly different from control by Fisher Exact test (^ p≤0.05; ^^ p≤0.01) as calculated by OEHHA

Reproductive and Developmental Toxicity

In a two-generation oral reproductive toxicity study, parental (P) generation Crl:CD rats (30/sex/group) were dosed with K^+PFBS at 0, 30, 100, 300 or 1,000 mg/kg-day by oral gavage for 10 weeks prior to and through mating; females were dosed through gestation and lactation (Lieder et al., 2009b). The F1 generation pups were dosed beginning at weaning. The study ended 3 weeks after the birth of the F2 generation pups, which were not dosed. At 300 and 1,000 mg/kg-day, increased absolute and relative liver weight and increased incidence of hepatocellular hypertrophy in males were observed. Increased incidence and severity of histological changes were also noted in the kidney of both sexes in the P and F1 generations at ≥300 mg/kg-day (Table 4). These changes consisted of hyperplasia of the medullary/papillary tubular and ductular epithelium. Low incidences of focal necrosis of the papilla were also observed in the treated males and females, however they did not follow a clear dose-response pattern. Similar effects were also noted in the rat in the 90-day oral study (Lieder et al., 2009a) described above, supporting the kidney as a target organ of PFBS. The study authors identified a NOAEL of 100 mg/kg-day for P and F1 generations, based on liver and kidney effects at higher doses. However, the liver and kidney histopathological data for the 30 and 100 mg/kg-day dose groups were not presented in the study. Thus, OEHHA could only determine a LOAEL of 300 mg/kg-day for histopathological changes in the liver and kidney.

Statistically significant increases in abnormal sperm morphology and decreased testicular sperm count were observed in the P generation males at 1,000 mg/kg-day, but similar effects were not observed in the F1 generation. Study authors also cited the changes were within the historical control range, and unlikely to be treatment related. There were no additional treatment-related effects on reproduction, fertility, or delivery outcomes among the P or the F1 rats. The reproductive NOAEL was 1,000 mg/kg-day. In the F1-generation, terminal body weight and relative liver weight were reduced, and preputial separation was delayed approximately 2 days in males at 1,000 mg/kg-day. There were no effects observed in the F1 females or F2 pups of either sex. The developmental NOAEL in the F1 generation is 300 mg/kg-day based on body weight and liver weight effects at 1,000 mg/kg-day.

Statistically significant effects were modeled by OEHHA in BMDS using a BMR of 5%. Most endpoints returned questionable or unusable results. The lowest $BMDL_{05}$ from a viable model using the published data was 35.6 mg/kg-day for hyperplasia in the kidney of P generation males. BMD results for the selected model can be found in Appendix II (Figure A4). It should be noted that Lieder et al. (2009b) only published histopathological data for the 0, 300 and 1,000 mg/kg-day dose groups. US EPA (2018a) has the histopathological data from the two lower dose groups not found in the Lieder et al. (2009b) publication, as it was originally reported in the 3M sponsored toxicological study (cited as York, 2003c in US EPA, 2018a), which was the basis for the published study. OEHHA extracted the additional information from HAWC and modeled kidney hyperplasia in the P generation using all dose groups. For males, the $BMDL_{05}$ was 37.4 mg/kg-day, higher than what was calculated from modeling only 3 dose groups from the published study. For females, the $BMDL_{05}$ was 26.2 mg/kg-day when modeled with all dose groups, whereas the models using only the 3 dose groups in the published study were not viable. Regardless, the kidney effects from Lieder et al. (2009b) were not the most sensitive in the animal toxicity database and were not further evaluated for health-protective concentration derivation.

Table 4. Incidence of histomorphological changes in liver and kidney of P and F1 generation rats (Lieder at el., 2009b)

Dose PFBS (mg/kg-day)[a]	**0**	**300**	**1,000**	**0**	**300**	**1,000**
Sex	**Male**	**Male**	**Male**	**Female**	**Female**	**Female**
Hepatocellular hypertrophy P generation	0/30	3/30	26/30**	0/30	0/30	0/30
Hepatocellular hypertrophy F1 generation	0/30	3/30	14/30**	0/30	0/30	0/30
Papillary epithelial tubular/ductal hyperplasia P generation	0/30	9/30**	19/30**	3/30	16/30**	21/30**
Papillary epithelial tubular/ductal hyperplasia F1 generation	3/30	5/30	21/30**	2/30	13/30**	15/30**
Focal papillary edema P generation	1/30	2/30	6/30	1/30	8/30*	7/30

Dose PFBS (mg/kg-day)[a]	0	300	1,000	0	300	1,000
Sex	**Male**	**Male**	**Male**	**Female**	**Female**	**Female**
Focal papillary edema F1 generation	1/30	0/30	9/30*	0/30	7/30*	4/30

* p<0.05, ** p<0.01, Fisher Exact test, significant from control, calculated by OEHHA

In a developmental toxicity study, thirty female ICR mice were orally dosed with 0, 50, 200 or 500 mg/kg-day K^+PFBS from gestation days (GD) 1 to 20 (Feng et al., 2017). The dams were randomly assigned into 3 groups measuring either reproductive and developmental endpoints, gonadal and thyroid hormones, or serum levels of PFBS.

Female offspring experienced significant effects on numerous developmental and reproductive parameters at ≥200 mg/kg-day; results for male offspring were not reported as they were used for a different purpose. These effects included statistically significant decreases in perinatal body weight, delayed eye opening, delayed vaginal opening, and delayed time to first estrous when compared to control. Postnatal day (PND) 60 offspring also exhibited decreased ovarian and uterine sizes and relative weights, and reduced follicle and corpus luteum numbers per ovary. Pubertal (PND30) and adult (PND60) offspring exhibited statistically significant decreases in serum estrogen (E2). Progesterone (P4) levels also were significantly decreased at PND60 and luteinizing hormone (LH) levels were significantly increased at PND30 at the same doses. Most notably, serum T4 and T3 levels were significantly decreased in PND1, PND30, and PND60 offspring. Slight increases in TSH were observed at PND30 and PND60, but the effect was only statistically significant at PND30. The changes in thyroid hormone levels were especially noteworthy as the effects of gestational exposure to PFBS persisted in the offspring in the postnatal period up to 60 days (the termination of the study).

There was no effect of PFBS on maternal body weight or general health. Maternal thyroid hormone levels were examined on GD20. Dams treated with 200 and 500 mg/kg-day PFBS exhibited statistically significant decreased serum levels of tT4 and T3, fT4, and increased TSH. There was no change in E2 and P4 levels. The study NOAELs were 50 mg/kg-day for both developmental and maternal effects. Data for statistically significant endpoints are presented in Table 5.

Table 5. Statistically significant developmental and thyroid hormone effects of gestational exposure to PFBS (Feng et al., 2017)[a]

PFBS (mg/kg-day)	Number of animals (n)	0	50	200	500
Eye opening (PND)	50/10[b]	14.8 ± 0.71	15.1 ± 1.2	16.3 ± 1.4**	16.5 ± 2.8**
Vaginal opening (PND)	30/10[b]	27.2 ± 2.2	28.4 ± 2.7	30.7 ± 3.3**	32.5 ± 3.3**
First estrous (PND)	30/10[b]	28.4 ± 2.7	28.2 ± 2.2	33.8 ± 3.3**	34.1 ± 2.2**
Relative ovarian weight[c] (PND60)	30/10[b]	0.052 ± 0.009	0.048 ± 0.011	0.046 ± 0.007*	0.047 ± 0.009*
Relative uterine weight (PND60)	30/10[b]	0.343 ± 0.096	0.308 ± 0.068	0.258 ± 0.055**	0.273 ± 0.055**

PFBS (mg/kg-day)	Number of animals (n)	0	50	200	500
Total T4 (µg/dl)[c] PND1 offspring	30/10[b]	1.4 ± 0.3	1.3 ± 0.7	0.9 ± 0.5**	0.7 ± 0.7**
T3 (ng/dl) PND1 offspring	30/10[b]	20.1 ± 5.8	21.2 ± 2.9	14.3 ± 2.9**	12.7 ± 2.9**
TSH (ng/ml) PND1 offspring	30/10[b]	112.9 ± 25.5	91.4 ± 25.5	94.1 ± 17.0	86.0 ± 34.0
Total T4 (µg/dl) PND30 offspring	10/10[b]	4.2 ± 0.44	4.0 ± 0.44	2.5 ± 0.38**	2.7 ± 0.54**
T3 (ng/dl) PND30 offspring	10/10[b]	75.1 ± 5.0	72.5 ± 8.4	51.3 ± 8.4*	49.7 ± 10.0*
TSH (ng/ml) PND30 offspring	10/10[b]	343.4 ± 31.4	328.5 ± 23.6	393.1 ± 39.3*	393.1 ± 31.4*
Total T4 (µg/dl) PND60 offspring	10/10[b]	2.6 ± 0.2	2.5 ± 0.6	2.0 ± 0.4*	2.0 ± 0.5**
T3 (ng/dl) PND60 offspring	10/10[b]	67.2 ± 6.7	72.0 ± 8.4	56.1 ± 8.4*	60.3 ± 6.7*
TSH (ng/ml) PND60 offspring	10/10[b]	279.6 ± 42.5	274.2 ± 34.0	325.3 ± 51.0	317.2 ± 51.0
Free T4 (pg/ml) GD20 dams	8	16.8 ± 2.0	17.6 ± 1.8	14.7 ± 1.4*	15.0 ± 1.3*
Total T4 (µg/dl) GD20 dams	8	2.4 ± 0.3	2.2 ± 0.3	1.9 ± 0.4*	1.9 ± 0.4*
T3 (ng/dl) GD20 dams	8	90.6 ± 9.1	84.5 ± 11.7	75.6 ± 11.3*	76.2 ± 12.1*
TSH (ng/ml) GD20 dams	8	271.0± 65.0	250.9 ± 53.4	329.0 ± 39.1*	331.6 ± 37.0*

* p <0.05; ** p <0.01 significantly different from controls (1-way ANOVA) as reported by study authors.
[a] Group data were reported as mean +/- standard error, converted to standard deviation by OEHHA.
[b] Numerator is number of pups, denominator is number of litters.
[c] Organ weights and hormone levels for offspring were presented graphically and not tabulated in the publication. Group response data were extracted from US EPA's analysis in HAWC: https://hawcprd.epa.gov/assessment/100000037/, accessed August 2020.

OEHHA conducted BMD modeling of endpoints for the Feng et al. (2017) study using a BMR of 1 SD. Numerous endpoints, particularly for PND30 and PND60 pups, were not amenable to BMD modeling and resulted in poorly fitted models. Of the valid models, $BMDL_{1SD}$ values ranged from 22.1 mg/kg-day (tT4 in GD20 dams) to 59.1 mg/kg-day (tT4 in PND1 pups). OEHHA considered the lowest $BMDL_{1SD}$ of 22.1 mg/kg-day for decreased tT4 from the GD20 dams as a candidate POD for health-protective concentration derivation. BMD model output from the selected model for this endpoint can be found in Appendix II (Figure A5). Additional discussion of significant results and modeling of thyroid hormone levels are provided in the Critical Effect Determination section.

Pharmacokinetics

OEHHA identified numerous studies from the database that provided information on the pharmacokinetics of PFBS in both humans and animal models. These studies are summarized here.

A recently published study (Xu et al., 2020) investigated serum half-life in a cohort of airport employees in Sweden found to be exposed to high levels of PFAS due to groundwater contamination from firefighting foam. Subsequent to the detection of PFAS contamination in the groundwater, subjects were supplied clean drinking water and within 2 weeks, blood samples were taken from 26 employees. Seventeen employees were followed for five months and paired serum and urine samples were collected. Only results for PFBS are discussed here. Airport drinking water was found to contain 0.2 ppb PFBS, while the municipal drinking water serving the surrounding area contained <0.0003 ppb. The average serum level in the first sampling of all subjects was 0.33 ng/ml. The average half-life calculated for PFBS in the subjects was 0.12 years, or 44 days.

There is evidence that the pharmacokinetic behavior of PFBS varies by species and sex, although serum half-life estimates vary across studies. Chengelis et al. (2009) looked at serum half-life in both monkey and rat following a single intravenous (i.v.) exposure. The serum half-life for PFBS in cynomolgus monkey was 8-15 hours, with females having a shorter half-life than males. However, only three animals per sex were used in this study, and one male had high serum levels. In rats, the serum half-life was 2.1 hours in males, and was much shorter in females, at 0.64 hours. Female rats appeared to have a higher rate of clearance, higher volume of distribution, and a 3-fold shorter half-life, as compared to males. Urine was the major excretion pathway for PFBS in rats, with 70% of the dose recovered within the first 24 hours. Urinary excretion half-life was 2-3 hours in rats, with no sex differences (Chengelis et al., 2009).

Olsen et al. (2009) compared pharmacokinetics of PFBS across three species (rats, monkeys, and humans). In male and female rats exposed via oral gavage or i.v. injection to 30 mg/kg PFBS, the main route of excretion was urine, with up to 74% excreted in the first 24 hours. By 96 hours post-dosing, less than 1% of the administered dose was still detectible in urine. The terminal serum half-life was 4.5 hours in male and 4.0 hours in female rats following i.v. dosing, but males had a lower initial serum elimination half-life and larger mean area under the concentration-time curve (AUC). Following oral gavage dosing, the serum half-life was shorter in males, at 4.7 hours, as compared to 7.4 hours in females; however, mean serum concentrations were higher in males at 24 hours post-exposure. The authors attribute this to more rapid initial serum elimination in females. In cynomolgus monkeys, 34% to 84% of administered PFBS was excreted in urine within the first 24 hours of a single i.v. dose. Serum half-life for PFBS was 95 and 83 hours in male and female monkeys, respectively. In humans, Olsen et al. (2009) measured urine and serum levels periodically over a six-month period following occupational exposure to PFBS. The

geometric mean serum half-life across six participants was 25.8 days. The mean half-life among five male workers was 24 days, and a single female employee had a serum half-life of 45 days. There was some correlation between paired serum and urine concentrations, suggesting that urinary excretion is an important pathway in humans as well. The elimination rates for PFBS are much faster than for PFOS and, as with other perfluorinated compounds, the serum half-life is much longer in humans relative to other species.

Gomis et al. (2018) constructed a one-compartment toxicokinetic model to predict serum concentrations of six PFAS based on external dose. Previously published subchronic studies in male rats, administered a single dose via oral gavage, with time-dependent serum concentration data reported were included. Data from female rats were excluded, based on evidence of sex-specific differences in elimination for some PFAS, including PFBS. The endpoint of concern used to develop a dose-response relationship incorporating the kinetic model was increased relative liver weight. The AUC was used as the best measure of internal dose, and changes in serum concentration were assumed to be equivalent to changes in liver concentration. The model showed PFBS reaching steady state levels within 24 hours following a simulated 10-day exposure using a dose of 1 mg/kg-day. Administered doses from published studies were converted to steady state serum and liver levels using the model, then compared to observed changes in liver weight. PFBS showed no effect on liver weight up to 600 mg/kg-day. In contrast, the model predicted a lowest-observed-effect level (LOEL) of 1.34 mg/kg-day for increased liver weight following exposure to PFOS. However, uncertainty analysis showed that the predicted liver AUC for PFBS was associated with high uncertainty.

A more recent toxicokinetic model was constructed (Huang et al., 2019a) using data generated by the authors from a single i.v. or a single oral gavage dose, in both male and female rats. A two-compartment model fit the data for male rats, with a plasma half-life of 2 hours following i.v. dosing at 4 mg/kg, or 2.7-4.4 hours following oral gavage dosing at 4, 20, or 100 mg/kg. In female rats, the corresponding half-lives were shorter, at 0.36 hours and 1.1-1.5 hours following i.v. and oral gavage dosing, respectively. The female i.v. dosing data fit a two compartment model, while the female oral gavage data fit a one compartment model. Liver, kidney, and brain tissue were collected at varying time points up to 12 hours following a single 20 mg/kg oral gavage dose. Tissue clearance differed between males and females, with PFBS levels declining but still detectable in liver and kidney at 12 hours post-dosing in males, whereas in females PFBS was no longer detected in the liver and kidney at 12 hours post-dosing. Levels in brain tissue declined over the initial post-dosing period and were undetectable by 12 hours post-dosing in males. In females, however, low levels of PFBS were detected in brain tissue only during the initial hour following dosing. Significant sex-specific differences were seen in this study, with plasma half-lives of 1-2 hours in female rats and 3-4 hours in male rats following oral gavage exposure. In addition, females exhibited faster tissue clearance of PFBS.

Similar to rats, female mice were also shown to have a shorter half-life of PFBS than males in a recent study published by Lau et al. (2020). Female mice had an average half-life of 4.5 hours following a 30 or 300 mg oral gavage dose of PFBS, while the half-life for male mice was significantly longer at 5.8 hours. As expected, the AUC was also significantly higher for males than females for serum, liver, and kidney compartments, and clearance rate was lower for male mice.

Bogdanska et al. (2014) exposed adult mice to radiolabeled PFBS via diet. Following 5 days of exposure, PFBS was detected in liver, stomach, kidney, bone, cartilage, lung, and spleen. Of these compartments, the highest levels were detected in liver, with steady state levels reached after 3 days. High levels were also seen in bone and cartilage; interestingly, PFBS-exposed animals had elevated levels of hemoglobin at the end of the exposure period. Low levels of PFBS were detected in fat, muscle, and brain. Compared to PFOS, PFBS was found in similar tissue compartments, but at levels that were 4-50 times lower than corresponding PFOS levels, likely due to more rapid elimination.

High levels of PFAS are seen in the liver following oral exposure, and a possible mechanism for transport via bile salt transporters was examined (Zhao et al., 2015). The enterohepatic system moves bile salts from the liver to the intestines, where they are absorbed into the blood stream and sent back to the liver. To determine if sodium-dependent bile salt transporters are involved in the disposition of PFAS, human and rat hepatocytes were incubated with PFBS, PFOS, and perfluorohexane sulfonic acid (PFHxS) for 2 minutes, and uptake was quantified. While sodium dependent uptake was observed for all three compounds, uptake levels of PFBS were much lower than that of both PFOS and PFHxS. Chinese hamster ovary (CHO) cells and human embryonic kidney (HEK) cells expressing both rat and human bile salt transporters were used to determine individual transporter affinity for PFAS. The sodium/taurocholate co-transporting polypeptide (NTCP) transporter and apical sodium-dependent bile salt transporter (ASBT) both play important roles in bile salt transport in the intestine. In human NTCP expressed in CHO cells, PFAS exposure inhibited uptake of the model substrate [^{3}H]-taurocholate in a chain-length dependent manner, with PFBS having only a mild effect on transport activity as the shortest of the three compounds. Rat NTCP expressed in HEK293 cells, in contrast, did not show the same chain-length dependent effects, with PFHxS having the strongest effect and PFOS and PFBS having equivocal, milder effects on transport. Human ASBT exhibited sodium dependent uptake of only PFOS; neither human nor rat ASBT showed sodium dependent uptake of PFBS. All three PFAS tested are substrates for human and rat NTCP, but only PFOS is a substrate for ASBT, suggesting that NTCP is likely the major of the two uptake transporters.

Lactation is not a significant pathway for excretion of PFBS in dairy cows (Kowalczyk et al., 2013). The main pathway for clearance was urine in lactating cows fed PFBS, PFHxS, PFOA, and PFOS contaminated feed for 28 days, followed by a 21-day exposure-free period. PFBS was undetectable in serum by 4 days post-exposure, and

urine levels were not detectible by 10 days post-exposure. PFBS was not secreted in milk; only one of six cows had PFBS levels above the limit of detection (LOD) in milk, at 0.12 µg/L. PFOS, however, was detected at high levels in milk; indeed, the levels of PFOS continued to increase in milk after exposure ended, with peak concentration in milk occurring on day 35. Immediately following the 28-day exposure period, low levels of PFBS were found in liver and kidney samples; however, PFBS levels were below the LOD by 21 days post-exposure, suggesting that PFBS does not bioaccumulate in dairy cows. Interestingly, there was no correlation between PFAS levels in milk and fat or protein content of the milk.

In Vitro Studies

OEHHA identified 27 studies from its literature search that met the PECO criteria for mechanistic studies. These were in vitro studies in a variety of human and animal cell types. The key findings of studies providing relevant mechanistic data are described here. No in vitro studies evaluated had data useful for dose-response assessment.

At concentrations up to 100 µM, PFBS did not affect cell viability or TSH-induced cAMP production in cultured rat thyroid cells (Croce et al., 2019). Production of reactive oxygen species (ROS) and oxidative DNA damage were not observed in PFBS-treated human HEPG-2 liver cells at concentrations of up to 2,000 µM (Eriksen et al., 2010) and PFBS had no effect on membrane fluidity or mitochondrial membrane potential in fish leukocytes at concentrations up to 50 mg/L (Hu et al., 2003). A more recent study (Liu et al., 2020) showed a significant increase in ROS production but no cytotoxicity in human mesenchymal stem cells (hMSCs). In a study by Slotkin et al. (2008), lipid peroxidation was induced in differentiating neuronotypic PC12 cells following PFBS exposures ranging from 50 to 250 µM. Interestingly, PFBS also displayed a concentration-dependent decrease in tyrosine hydroxylase and choline acetyltransferase activities, which are indicative of the differentiation of PC12 cells into the dopamine and acetylcholine neurotransmitter phenotypes; however, possible effects on neuronal differentiation are still unclear (Slotkin et al., 2008).

A luciferase-based assay showed PFBS had no effect on activation of nine human nuclear receptors, including peroxisome proliferator-activated receptor alpha (PPARα) (Behr et al., 2020). An earlier study by (Wolf et al., 2008) showed mild activation of PPARα in both mouse and human cell reporter systems, although at higher concentrations than Behr et al. (2020). PFBS treatment upregulated expression of common adipogenic gene markers in hMSCs induced to enter adipogenesis. Increased lipid accumulation was observed in embryonic mouse 3T3-L1 preadipocyte cells following exposure, along with increased levels of PPARγ, a regulator of fatty acid storage (Qi et al., 2018).

In a study to assess possible toxicity to the respiratory system, no effects on inflammatory responses in human bronchial epithelial cells were observed, and no effects on lung surfactant were seen with PFBS (Sørli et al., 2020). However, PFBS did

induce changes in interleukin-10 (IL-10) release, with more pronounced effects in female whole blood leukocytes as compared to males, and interfered with NF-κB (nuclear factor kappa B) activation by inhibiting I-κB (inhibitor of NF- κB) degradation, thereby highlighting possible immunomodulatory effects via inhibition of cytokine production (Corsini et al., 2012).

Despite low levels of cellular uptake, PFBS significantly inhibited Cyp19 aromatase, an enzyme important in maintaining hormonal balance of androgens and estrogens, in a human placental cell line (Gorrochategui et al., 2014). No effects on the activity of 3β- or 17β-hydroxysteroid dehydrogenase enzymes in human or rat microsomes derived from testis cells were observed following exposure (Zhao et al., 2010).

CRITICAL EFFECT DETERMINATION AND REFERENCE LEVEL CALCULATIONS

OEHHA develops health-protective concentrations using the general approach of the Public Health Goal (PHG) program for developing health-protective concentrations of chemicals in drinking water that are expected to result in no adverse effects from exposure over a lifetime. For noncancer effects, the derivation of the health-protective concentration starts with the PODs derived from animal or human studies. This dose is converted to an acceptable daily dose (ADD), which is then back calculated to a health-protective concentration in drinking water. Because there were no studies of the carcinogenicity of PFBS, only a noncancer health-protective concentration was derived.

OEHHA evaluated the health outcomes of the most sensitive animal toxicity studies available in the literature for health-protective concentration derivation. In the four studies selected as candidate critical studies (Table 1), the health outcomes included effects on the thyroid, reproductive organs and tissues, developing offspring, liver, and kidney following oral exposure to PFBS. From these identified targets of PFBS, the kidney and thyroid were found to be the most sensitive to PFBS-induced toxicity. These target organs are the same as those identified by US EPA (2018a) and MDH (2017). As discussed in the following sections, OEHHA identified the thyroid as the most sensitive for health-protective concentration derivation.

Renal effects were observed in both the 90-day oral and two-generation reproductive toxicity studies in rats (Lieder et al., 2009a and 2009b). Rats of both sexes and in both studies had significant focal papillary edema and papillary tubular/ductal epithelial hyperplasia at doses of 300 mg/kg-day and greater. Kidney hyperplasia and edema were not reported in the other animal toxicity studies of PFBS, but the studies were shorter in duration, and significant increases in relative kidney weight and blood urea nitrogen (serum marker of renal injury) were observed in the 28-day oral study in rats (NTP, 2019). Overall, the animal toxicity data support the kidney being a target organ of PFBS toxicity. However, the potential PODs based on kidney effects are not the most sensitive in the database. Therefore, kidney effects were not selected as the critical endpoint for health-protective concentration derivation.

Thyroid hormone disruption was a key effect identified in animal toxicity studies of PFBS in two species, and in both adult and developing animals. Significant, dose dependent, and consistent reductions in thyroid hormones T3 and T4 (total and free) were observed in adult male and female rats following 28 days of oral exposure to PFBS (NTP, 2019). Pregnant female mice and gestationally-exposed female mouse offspring had similar reductions in thyroid hormones, which also persisted in offspring through the postnatal period and into adulthood (Feng et al., 2017). Pregnant mice and their female offspring during the postnatal period also exhibited increases in TSH, although the increase was not always statistically significant. Neither of these studies detected significant effects on thyroid weight or histopathology, though the sensitivity to detect changes may have been limited by the length of exposure. The study by Feng et al. (2017) also reported various significant effects on developmental outcomes, including effects on body weight and delays in eye opening, vaginal opening, and time to first estrus.

OEHHA has extensively reviewed thyroid physiology and adverse effects of thyroid hormone disruption and deficiency in humans and animals in previous assessments, most notably for the perchlorate PHG (OEHHA, 2004; OEHHA, 2015). Perchlorate causes thyroid perturbation by blocking iodide uptake in the thyroid gland, potentially decreasing the production of T3 and T4.

A brief background on thyroid hormone regulation and the effects of thyroid hormone deficiency, particularly as they relate to developmental outcomes, was taken from the most recent perchlorate PHG document (OEHHA, 2015) and is presented here. The principal hormones secreted by the thyroid are T4 and T3. Iodide is a key component of both. While T4 is produced only by the thyroid gland, about 80% of T3 is formed outside the thyroid by deiodination of T4. T4 and T3 influence the growth and maturation of tissues, cell respiration and total energy expenditure, and the turnover of essentially all substrates (including carbohydrates, cholesterol, and proteins), vitamins, and hormones (including the thyroid hormones themselves). Control of T4 and T3 concentrations in blood is mainly regulated by a negative feedback loop involving three organs: the thyroid gland, which produces thyroid hormones; and the pituitary gland and hypothalamus, which respond to and help maintain optimal levels of thyroid hormones. When levels of thyroid hormone decline, the hypothalamus secretes thyrotropin-releasing hormone (TRH), which stimulates the pituitary to produce TSH, which then prompts the thyroid gland to produce T4 and T3. Circulating T4, T3, and TSH can readily be measured in the serum of experimental animals and humans and serve as biomarkers of exposure and effect of agents that disrupt thyroid-pituitary status (US EPA, 1998a, and 1998b; Hill et al., 1989).

Thyroid hormones are critical determinants of growth and development in fetuses, infants and young children. There are data suggesting that certain adverse fetal effects during development are inversely related to maternal serum T4 levels (Pop et al., 2003; Kooistra et al., 2006). Maternal serum fT4 is able to pass through the placenta and is converted to T3 in the fetal brain. The T3 generated in the fetal brain is believed to be necessary for the development of the brain, specifically the cerebral cortex, the

extrapyramidal system, and the cochlea (Porterfield, 2000). The availability of a minimum level of maternal fT4 is crucial for proper fetal brain development in the first and second trimesters, as the fetal thyroid is not fully mature and functional during that time period.

Evidence suggests that even small decreases in thyroid hormone levels may be associated with significant adverse effects, including altered cognitive development in children and increased cardiovascular risk factors in adults. Importantly, these changes have been seen at thyroid hormone levels that are within what have been traditionally defined as normal reference ranges, and have occurred in people without any other evidence of overt thyroid disease. These findings suggest that small changes in thyroid hormone levels may be associated with some increased risk of thyroid-related adverse outcomes (OEHHA, 2015). While the mode of action (MOA) by which PFBS disrupts thyroid hormones is unknown at this time, the resulting reduction of T3 and T4 in animal models supports a thyroid hazard.

Critical Effect Determination

Table 1 lists the PODs (both NOAELs and BMDLs) derived from the studies OEHHA identified as suitable for quantitative dose-response analysis and health-protective concentration derivation. The results from animal toxicity studies of PFBS in mice and rats support the thyroid as a potential target organ of PFBS, and the measured effects in the thyroid were found to be more sensitive than in the kidney. Thyroid hormone disruption from the Feng et al. (2017) and NTP (2019) studies were the most sensitive endpoints in the PFBS animal toxicity database, and both were considered for health-protective concentration derivation.

The most sensitive endpoint in the 28-day oral gavage study in adult rats (NTP, 2019) was decreased tT4 in non-pregnant female rats. As previously stated, BMD modeling for other hormone levels (fT4, T3) was not successful based on fit statistics, and no statistically significant effects on TSH were measured in the study. Female rats were also more sensitive to thyroid hormone perturbation than males. BMD modeling of the female rat tT4 data with a BMR of 1 SD returned a $BMDL_{1SD}$ of 6.9 mg/kg-day from the exponential M4 model, which had the best statistical and visual dose-response fit. Similar effects have been noted for other PFAS, such as PFHxS, where studies in the rat showed marked reduction of circulating thyroid hormones but a lack of an accompanying rise in TSH, suggesting there was no activation of the hypothalamic-pituitary-thyroid (HPT) axis (Ramhøj et al., 2020). Studies in humans have demonstrated potential consequences of reductions in tT4 hormone levels without a compensatory response in the HPT axis. Maternal hypothyroxinemia (defined as maternal fT4 level below the 2.5^{th} percentile, and in the absence of TSH elevation) has been linked to developmental and cognitive delays in offspring (Negro et al., 2011). However, the lack of developmental effects following PFBS exposure in pregnant rats at doses as high as 300 mg/kg-day (Lieder et al, 2009b; York, 2002) suggests the level of fT4 depression in the rat due to PFBS treatment was not sufficient to cause

developmental effects in the rat fetus. Alternatively, the assays themselves may not have been sensitive enough to detect fetal effects, if they were in fact present. Either scenario suggests that, for PFBS, the rat may not be the best model for impacts of lowered maternal thyroid hormone on the developing human fetus.

Thyroid hormone disruption in the Feng et al. (2017) developmental toxicity study was observed in GD20 dams, and PND1, PND30, and PND60 offspring. OEHHA considered decreased T4 levels in PND1 mice as a critical endpoint, consistent with US EPA's approach in its draft human health toxicity assessment for PFBS (US EPA, 2018a). However, there were uncertainties in the dataset that questioned the reliability of using the data for BMD modeling. First, the data itself were presented graphically and required the data to be digitized for analysis. While that in itself would not be sufficient reason to preclude use of the dataset, there was also uncertainty in the number of animals used in the analysis. It is unclear if the dose group analyses were based on the number of litters or the number of fetuses as the statistical unit when analyzing PND1 offspring. Thus, the appropriate SD cannot be derived from the standard error (SE) bars on the graph, nor can OEHHA determine if potential litter effects or litter covariates were accounted for in their analysis. Like US EPA, OEHHA attempted to contact study authors for additional information but did not receive a response. Due to these uncertainties in the dataset, OEHHA determined that it would not be appropriate to perform BMD modeling on the thyroid hormone endpoints for PND1 mice. Developmental delays in female offspring from Feng et al. (2017) were also considered for potential PODs for health-protective concentration derivation, but the PODs were slightly higher and thus, the endpoints were less sensitive than thyroid hormone perturbation. Regardless, the Feng et al. (2017) study clearly demonstrated that thyroid hormone disruption from PFBS exposure during the gestational period was significant, dose dependent, and the effects persisted into adulthood in the mouse test species. This hazard from the animal toxicity studies suggests that in humans, pregnant women and their offspring are likely the most susceptible subpopulations to adverse effects resulting from thyroid hormone perturbation due to PFBS exposure during a critical window of development. Developmental delays in mouse offspring were also significant, indicating that deriving the health-protective concentration from thyroid hormone changes would be protective of both potential thyroid and developmental effects.

Due to the uncertainties in the PND1 dataset, OEHHA modeled the serum thyroid hormone levels of GD20 dams instead. Modeling of tT4 in the serum of GD20 dams, using a BMR of 1 SD, returned a $BMDL_{1SD}$ of 22 mg/kg-day. Although this is higher than the candidate POD of 6.9 mg/kg-day derived from the adult female rats in the NTP (2019) study, OEHHA is selecting 22 mg/kg-day, derived from the Feng et al. (2017) mouse study, as the POD. While the pregnant mouse was slightly less sensitive to the thyroid hormone perturbation caused by PFBS than the adult rat, the adverse effects in mouse offspring observed in Feng et al. (2017) supports the hazard in humans, namely that adverse fetal outcomes can be caused by even small changes in maternal thyroid hormone. The lack of adverse developmental outcomes in the offspring of gestationally

exposed rats in both reproductive (Lieder et al., 2009b) and developmental toxicity studies (York, 2002) suggest that at least for PFBS, the mouse is a more sensitive and/or more representative model of the in utero effects of thyroid hormone disruption in humans.

Health-Protective Concentration Calculation

Human Equivalent Dose (HED)

To derive a human equivalent dose (HED) from the animal POD, OEHHA utilized pharmacokinetic data to calculate a human clearance factor (CL) from Olsen et al. (2009) and applied the human CL to the average serum concentration for mouse derived from Lau et al. (2020).

To calculate the human CL for PFBS, OEHHA used matched serum and urine concentrations for PFBS from Olsen et al. (2009) and the following equation:

$$CL = \frac{amount\ in\ urine}{C_{ss} \times BW}$$

where C_{ss} is the steady state serum concentration of PFBS and BW is body weight.

The study included 6 individuals (5 male, 1 female), with 3 time points per individual where matched serum and urine concentrations were evaluated. By the third time point, all urine samples were below the limit of quantitation (LOQ). Assuming urine volume of 1.3 L/day and BW of 70 kg, the available data points (with matched values >LOQ) are presented in Table 6. The mean clearance calculated from the available data was 3.90 ml/kg-day.

Table 6: Renal clearance calculated from paired serum and urine concentrations (ml/kg-day) from 6 human subjects exposed to PFBS (Olsen et al., 2009)

	First pair	Second pair	Average per subject
Subject 1	9.08	3.66	6.37
Subject 2	2.53	ND	2.53
Subject 3	12.31	6.96	9.64
Subject 4	0.30	1.55	0.93
Subject 5	2.61	1.17	1.89
Subject 6*	2.01	ND	2.01
Arithmetic mean			**3.90**

ND, not determined because urine concentration was below the limit of quantitation.
*, denotes the sole female subject

Assuming that renal clearance is the dominant elimination route for PFBS, the relationship between the applied dose (AD) and average serum concentration ($C_{average}$) can be described with the formula:

$$AD = C_{average} \times CL$$

Therefore, the extrapolation from animal AD (AD_{animal}) to human AD (AD_{human}) can be ultimately expressed through the ratio of animal and human clearances, or dose adjustment factor (DAF), assuming that $C_{average}$ is similar at applied doses in either species:

$$AD_{human} = \frac{AD_{animal}}{\left({CL_{animal}}/{CL_{human}}\right)} = \frac{AD_{animal}}{DAF}$$

While CL_{human} can be calculated from experimentally measured values (Table 6), CL_{animal} for female mice, given 30 or 300 mg/kg PFBS by gavage as a single dose, was reported as 0.056 or 0.064 L/kg-h, respectively (Lau et al., 2020). The clearance values were fairly close at the two different doses. Moreover, the 30 mg/kg dose in this study was close to the selected POD of 22 mg/kg-day and the lowest dose tested of 50 mg/kg-day from the mouse study by Feng et al. (2017). Therefore, inputting CL_{animal} for the 30 mg/kg dose from Lau et al. (2020) into the formula for DAF:

$$DAF = \frac{CL_{animal}}{CL_{human}} = \frac{0.056\frac{L}{kg-h} \times 1000\frac{ml}{L} \times 24\ \frac{h}{day}}{3.9\frac{ml}{kg-day}} = 345.$$

This correction adjusts for pharmacokinetic differences between mouse and human using the metric of averaged serum concentration. Furthermore, although Feng et al. (2017) administered K^+PFBS to mice, a molecular weight adjustment for PFBS is not necessary because it is assumed that the salt would be completely dissociated in the neutral pH environment of the blood and what is measured is the free acid (PFBS).

To derive the POD_{human}, divide the animal POD by the DAF:

$$POD_{human} = \frac{22\ mg/kg\text{-}day}{345} = 0.06\ mg/kg\text{-}day$$

For comparative purposes, a DAF was also derived for the rat and an alternative POD_{human} ($POD_{alt/human}$) was calculated from the POD of 6.9 mg/kg-day for decreased tT4 from NTP (2019). OEHHA evaluated the available rat kinetic studies and identified a study conducted by the NTP, published as Huang et al. (2019), as the best quality study because multiple exposure routes were studied (oral gavage and intravenous), low doses were included (4 mg/kg), several kinetic models were tested, and quality control procedures and transparency of reporting were adequate. One additional study (Chengelis et al., 2009) reported clearance values with i.v. dosing that were consistent with the Huang (2019) study. The last of the available studies, Olsen et al. (2009), reported higher CL values, likely due to higher applied dose (30 mg/kg). In all three studies, female rats demonstrated higher clearance rates than males (4- to 8-fold),

unlike humans that do not have known PFBS kinetic differences between males and females.

For female rat, the reported clearance value from the lowest oral gavage dose (152 ml/h-kg) in Huang et al. (2019) was chosen for DAF calculation. This method is consistent with the calculation of the mouse CL, described above, and most closely reflects the conditions of the candidate critical study, which was conducted via oral gavage at comparable doses. To calculate the DAF, the human CL from Table 6 is used:

$$DAF = \frac{CL_{animal}}{CL_{human}} = \frac{152\frac{ml}{kg-h} \times 24\ \frac{h}{day}}{3.9\frac{ml}{kg-day}} = 935.$$

The large DAF is due to dramatic species differences in CLs between rats and humans. Although in the derivation of this formula it was assumed that the average serum concentration in rat would be similar to the steady state serum concentration in human, the overall kinetic profiles of serum concentrations are not quite comparable due to large differences in half-lives and serum elimination. Thus, in the rat, the initial spike in serum concentration would quickly decrease, whereas in the human the serum concentration would demonstrate less variation throughout the treatment period. In this case, assuming the equipotency of averaged serum concentrations would allow for interspecies extrapolation but at the same time contributes to increased uncertainty when using the DAF for interspecies extrapolation.

To derive the alternative $POD_{alt/human}$, the animal POD from the rat study (NTP, 2019) is divided by the DAF calculated for rat:

$$POD_{alt/human} = \frac{6.9\frac{mg}{kg-day}}{935} = 0.007\frac{mg}{kg-day}$$

As noted above, there are several uncertainties associated with this derivation. While the decrease in tT4 was chosen as the critical endpoint in the rat, the underlying mechanism of PFBS thyroid toxicity is not known. The large DAF reflects dramatic differences in elimination between the rat and the human. In addition to the need for kinetic adjustment, such large differences in elimination can contribute to species differences in the toxicodynamics of PFBS. Thus, with the currently available data on kinetics in the rat, there would likely be greater uncertainty in deriving a health-protective concentration from a rat study than from a mouse study.

Acceptable Daily Dose (ADD)

An ADD is the estimated maximum daily dose of a chemical (in mg/kg-day) that can be consumed by a human for an entire lifetime without adverse effects. To determine the

ADD, the POD is adjusted by factors to account for uncertainties in the risk assessment, such as differences between animals and humans (interspecies extrapolation), and differences among humans (intraspecies variation, including sensitive subgroups) in response to a chemical exposure. Additionally, factors may be applied to extrapolate from subchronic to chronic exposure duration, from LOAEL to NOAEL when a NOAEL or BMDL is not available, and also when the toxicity database is incomplete. These factors combined are referred to as the composite uncertainty factor (UF).

When developing health-protective concentrations for noncancer effects based on animal toxicity studies, OEHHA generally applies a composite UF of 300, consisting of 10 for interspecies extrapolation (√10 for pharmacokinetics and √10 for pharmacodynamics) and 30 for intraspecies variability (10 for pharmacokinetics and √10 for pharmacodynamics) (OEHHA, 2008). A detailed description of these factors is presented in Appendix III.

When calculating the ADD for PFBS using the POD from Feng et al. (2017), OEHHA applied an interspecies UF of √10 to account for potential differences in pharmacodynamics when extrapolating data from animal studies to humans. Because PFBS is not known to be metabolized in animals or humans, and because a pharmacokinetic adjustment was applied to the animal POD to derive a human equivalent dose, the pharmacokinetic components of the interspecies and intraspecies UFs were reduced by √10 each. Therefore, the intraspecies UF was reduced from OEHHA's default of 30 to 10 to account for human variability. Additionally, an additional UF of √10 was applied for database deficiencies, most notably the absence of a chronic toxicity study. This resulted in a composite UF of 100.

A subchronic UF of 10 is typically applied when the study duration is <8% of the animal's lifetime in order to account for the potential exacerbation of toxicity following chronic exposure (OEHHA, 2008). However, this factor was not applied because the critical effect of decreased tT4 in GD20 dams occurred during a critical window of susceptibility (GD1-20), which apparently led to the decrease in tT4 in the offspring (since the developing rodent thyroid is not functional and capable of producing its own thyroid hormone until late in gestation, at GD17 (US EPA, 2018a)). Furthermore, gestational exposure to PFBS resulted in a decrease in tT4 in pups that lasted into adulthood, without continuation of exposure in the postnatal period. It is unknown if cumulative prenatal and postnatal exposures would exacerbate the effects observed. Therefore, a database deficiency UF of √10 was added, as a chronic exposure study may result in more severe effects than what was detected in Feng et al. (2017). In addition, it is not known whether chronic exposure would result in other effects that occur at lower doses than the observed changes in thyroid, kidney, and liver in adult animals. Additionally, other deficiencies in the database besides chronic exposure studies exist for PFBS. While it is known that thyroid hormones are critical for cognitive development, there is no developmental neurotoxicity study for PFBS. There are also no studies of potential immunotoxicity or carcinogenicity, which is a concern as effects

on immunotoxicity and positive results in cancer bioassays have been observed for other PFAS such as PFOS and PFOA (NTP, 2016; OEHHA, 2019).

As mentioned in the toxicological review section, despite having a lower POD, OEHHA decided against using the NTP (2019) study to derive an ADD because of large toxicokinetic differences between female rats and humans, and uncertainty around the utility of the rat model for effects in humans of maternal thyroid hormone disruption on fetal development. Thus, the Feng et al. (2017) study was chosen to calculate an ADD with the highest level of confidence and human relevance.

To calculate the ADD, divide the POD_{human} by the composite UF:

$$ADD = \frac{0.06 \text{ mg/kg-day}}{100} = 0.0006 \text{ mg/kg-day}.$$

Relative Source Contribution (RSC)

In estimating health-protective levels of chemicals in drinking water for noncancer endpoints, OEHHA considers the relative source contribution (RSC), which is the proportion of exposures to a chemical attributed to tap water as part of total exposure from all sources, including food and ambient air. When developing an appropriate RSC value for a chemical, OEHHA follows US EPA's Exposure Decision Tree Approach (US EPA, 2000). This approach takes into account the availability of exposure data, including the levels and relevant sources of exposure, and any other non-water regulatory standards for the chemical. In addition, any specific subpopulations of concern are identified and considered in the process. A chemical-specific RSC value can be calculated when adequate data are available for all sources of exposure, including exposures from drinking water. If data are not adequate for determining the contribution of one or more of these non-water exposure sources, then default values may be used (US EPA, 2000). The default estimates include a low estimate of 20% and a high estimate of 80%, used when data are not otherwise available to better characterize sources of exposure.

For PFBS, the population of concern is all residents of California. Relevant exposure sources to Californians include environmental media and consumer products. However, the availability of data to quantify PFBS exposure from these sources is limited. PFBS has been detected in household dust, consumer products such as carpets, textiles, and upholstery protectors, fast food packaging, and various food and drink items consumed on a daily basis (US EPA, 2018a). Frequency of detection and concentrations measured varied widely by study and products tested. The European Food Safety Authority (EFSA) estimated dietary exposures to PFBS ranged from 0.03−1.89 nanograms per kilogram per day (ng/kg-day) (minimum) to 0.10−3.72 ng/kg-day (maximum) (EFSA, 2012).

While there is limited data quantifying the levels of PFBS in some items, PFBS is a ubiquitous environmental contaminant and the potential exposure sources are extensive. Because there is insufficient human data to assess the population's exposure to PFBS from all sources other than tap water, a default RSC of 20% was selected and is consistent with the US EPA (2000) guidance.

Drinking Water Intake (DWI)

To calculate a health-protective concentration for a chemical, the ADD is converted to a concentration in drinking water that accounts for the total exposure to the chemical that people receive from using tap water. It includes intake from ingestion, inhalation, and dermal contact with contaminants in tap water from household uses (e.g., drinking, cooking, bathing, and showering). Inhalation exposure can take place when a chemical volatilizes out of the water during cooking or showering. Dermal absorption of the chemical can occur during bathing and other household uses of tap water.

The daily water intake equivalent (DWI) is expressed in the units of liters or liter equivalents per kilogram of body weight per day (L/kg-day or L_{eq}/kg-day, respectively). Liter equivalents represent the equivalent of the amount of tap water one would have to drink to account for the exposure to a chemical in tap water through oral, inhalation, and dermal routes. For oral intake rates, the PHG program uses age-specific water ingestion estimates (OEHHA, 2012) derived from a nationwide survey of food and beverage intake from approximately 20,000 people (US Department of Agriculture's Continuing Survey of Food Intake of Individuals 1994-1996, 1998 dataset). These age-specific intake rates are normalized to body weight and expressed as L/kg-day.

PFBS exposure from tap water is expected to be predominantly from oral exposure. According to the Norwegian Geotechnical Institute (NGI, 2018), the volatilization of PFBS and K^+PFBS from water is negligible, and the air-phase presence is due to direct emissions into the air or contaminated water droplets or particles. Although no studies were found that evaluated the absorption of PFBS following dermal exposure, based on typical household uses of tap water, like showering and bathing, dermal absorption is not anticipated to be a significant route of exposure.[7] Thus, inhalation and dermal exposures to PFBS during household uses of tap water are expected to be negligible.

Although the critical effect for PFBS was measured in gestationally-exposed GD20 mice, OEHHA selected the 95th percentile consumer-only water intake rate of 0.237 L/kg-day for infants 0-6 months of age (OEHHA, 2012) for health-protective concentration derivation because infants are particularly susceptible to the effects of thyroid hormone perturbation and its sequelae, especially its effects on cognitive development. For instance, infants are less able to tolerate decreases in T4 because they have less than one day's worth of T4 stores compared to adults, who have several weeks' worth (van den Hove et al., 1999). Also, infants have higher exposure to

[7] The estimate for PFBS octanol-water partition coefficiet (log Kow) is 1.82 (https://pubchem.ncbi.nlm.nih.gov/)

drinking water contaminants because they consume more water (when fed reconstituted formula) on a body weight basis than adults. Furthermore, gestational exposure in the Feng et al. (2017) study resulted in pups exhibiting decreased T4 levels into adulthood. This underscores the need to account for greater exposure during the neonatal and infancy periods of life. Use of the 0-6 month infant drinking water intake rate is consistent with OEHHA's approach for another thyroid toxicant, perchlorate (OEHHA, 2015).

Health-Protective Concentration (C)

C = ADD × RSC ÷ DWI, where:

ADD = acceptable daily dose of 0.0006 mg/kg-day,

RSC = relative source contribution of 0.2, and

DWI = daily water intake rate of 0.237 L/kg-day

C = (0.0006 mg/kg-day × 0.2) ÷ 0.237 L/kg-day = 0.5 µg/L (or 0.5 ppb).

OEHHA recommends that SWRCB set the NL for PFBS in drinking water at 0.5 ppb.

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APPENDIX I. LITERATURE SEARCH TERMS AND PECO STATEMENT

PubMed – Search executed 12.4.2019

Search Terms	Results
(375-73-5[rn] OR nonafluorobutane-1-sulfonic acid [nm] OR PFBS[tiab] OR PFBuS[tiab] OR perfluorobutanesulfonic[tiab] OR perfluorobutanesulfonate[tiab] OR perfluorobutanesulphonic[tiab] OR perfluorobutanesulphonate[tiab] OR 1-perfluorobutanesulfonic[tiab] OR 1-perfluorobutanesulfonate[tiab] OR 1-perfluorobutanesulphonic[tiab] OR 1-perfluorobutanesulphonate[tiab] OR nonafluorobutanesulfonic[tiab] OR nonafluorobutanesulfonate[tiab] OR nonafluorobutanesulphonic[tiab] OR nonafluorobutanesulphonate[tiab] OR nonafluroro-1-butanesulfonic[tiab] OR nonafluoro-1-butanesulfonate[tiab] OR nonafluroro-1-butanesulphonic[tiab] OR nonafluoro-1-butanesulphonate[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluorobutane-1-sulfonic"[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluorobutane-1-sulfonate"[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluorobutane-1-sulphonic"[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluorobutane-1-sulphonate"[tiab] OR nonafluorobutane-1-sulfonic[tiab] OR nonafluorobutane-1-sulfonate[tiab] OR nonafluorobutane-1-sulphonic[tiab] OR nonafluorobutane-1-sulphonate[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluoro-1-butanesulfonic"[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluoro-1-butanesulfonate"[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluoro-1-butanesulphonic"[tiab] OR "1,1,2,2,3,3,4,4,4-nonafluoro-1-butanesulphonate"[tiab] OR (perfluorobutane [Tiab] AND sulfonic [Tiab]) OR (perfluorobutane [Tiab] AND sulfonate [Tiab]) OR (perfluorobutane [Tiab] AND sulphonic [Tiab]) OR (perfluorobutane [Tiab] AND sulphonate [Tiab]) OR perfluoro-1-butanesulfonic[tiab] OR perfluoro-1-butanesulfonate[tiab] OR perfluoro-1-butanesulphonic[tiab] OR perfluoro-1-butanesulphonate[tiab] OR "pentyl perfluorobutanoate"[tiab] OR nonaflate[tiab] OR (perfluorobutane [Tiab] AND sulfonic [Tiab]) OR (perfluorobutane [Tiab] AND sulfonate [Tiab]) OR (perfluorobutane [Tiab] AND sulphonic [Tiab]) OR (perfluorobutane [Tiab] AND sulphonate [Tiab]) OR nonafluorobutane sulfonic[tiab] OR nonafluorobutane sulfonate[tiab] OR nonafluorobutane sulphonic[tiab] OR nonafluorobutane sulphonate[tiab] OR perfluorobutane-1-sulfonic[tiab] OR perfluorobutane-1-sulfonate[tiab] OR perfluorobutane-1-sulphonic[tiab] OR perfluorobutane-1-sulphonate[tiab] OR TCR-282[tiab])	358

EMBASE – Search executed 12.4.2019

Search Terms	Results
('375 73 5':ti,ab OR 'perfluorobutane sulfonate'/exp OR 'PFBS':ti,ab OR 'PFBuS':ti,ab OR 'perfluorobutanesulfonic':ti,ab OR 'perfluorobutanesulfonate':ti,ab OR 'perfluorobutanesulphonic':ti,ab OR 'perfluorobutanesulphonate':ti,ab OR '1 perfluorobutanesulfonic':ti,ab OR '1 perfluorobutanesulfonate':ti,ab OR '1 perfluorobutanesulphonic':ti,ab OR '1 perfluorobutanesulphonate':ti,ab OR 'nonafluorobutanesulfonic':ti,ab OR 'nonafluorobutanesulfonate':ti,ab OR 'nonafluorobutanesulphonic':ti,ab OR 'nonafluorobutanesulphonate':ti,ab OR 'nonafluroro 1 butanesulfonic':ti,ab OR 'nonafluoro 1 butanesulfonate':ti,ab OR 'nonafluroro 1 butanesulphonic':ti,ab OR 'nonafluoro 1 butanesulphonate':ti,ab OR '1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulfonic':ti,ab OR '1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulfonate':ti,ab OR '1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulphonic':ti,ab OR '1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulphonate':ti,ab OR 'nonafluorobutane 1 sulfonic':ti,ab OR 'nonafluorobutane 1 sulfonate':ti,ab OR 'nonafluorobutane 1 sulphonic':ti,ab OR	410

‘nonafluorobutane 1 sulphonate’:ti,ab OR ‘1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulfonic‘:ti,ab OR ‘1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulfonate‘:ti,ab OR ‘1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulphonic‘:ti,ab OR ‘1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulphonate‘:ti,ab OR (‘perfluorobutane’ NEAR ‘sulfonic‘) OR (‘perfluorobutane‘ NEAR ‘sulfonate‘) OR (‘perfluorobutane‘ NEAR ‘sulphonic‘) OR (‘perfluorobutane‘ NEAR ‘sulphonate‘) OR ‘perfluoro 1 butanesulfonic’:ti,ab OR ‘perfluoro 1 butanesulfonate’:ti,ab OR ‘perfluoro 1 butanesulphonic’:ti,ab OR ‘perfluoro 1 butanesulphonate’:ti,ab OR ‘pentyl perfluorobutanoate‘:ti,ab OR ‘nonaflate’:ti,ab OR (‘perfluorobutane‘ NEAR ‘sulfonic‘) OR (‘perfluorobutane‘ NEAR ‘sulfonate‘) OR (‘perfluorobutane‘ NEAR ‘sulphonic‘) OR (‘perfluorobutane‘ NEAR ‘sulphonate‘) OR ‘nonafluorobutane sulfonic’:ti,ab OR ‘nonafluorobutane sulfonate’:ti,ab OR ‘nonafluorobutane sulphonic’:ti,ab OR ‘nonafluorobutane sulphonate’:ti,ab OR ‘perfluorobutane 1 sulfonic’:ti,ab OR ‘perfluorobutane 1 sulfonate’:ti,ab OR ‘perfluorobutane 1 sulphonic’:ti,ab OR ‘perfluorobutane 1 sulphonate’:ti,ab OR TCR-282:ti,ab)	

SCOPUS – Search executed 12.4.2019

Search Terms	Results
CASREGNUMBER(“375 73 5”) OR TITLE-ABS(“perfluorobutane sulfonate” OR [PFBS] OR [PFBuS] OR “perfluorobutanesulfonic” OR “perfluorobutanesulfonate” OR “perfluorobutanesulphonic” OR “perfluorobutanesulphonate” OR “1 perfluorobutanesulfonic” OR “1 perfluorobutanesulfonate” OR “1 perfluorobutanesulphonic” OR “1 perfluorobutanesulphonate” OR “nonafluorobutanesulfonic” OR “nonafluorobutanesulfonate” OR “nonafluorobutanesulphonic” OR “nonafluorobutanesulphonate” OR “nonafluroro 1 butanesulfonic” OR “nonafluoro 1 butanesulfonate” OR “nonafluroro 1 butanesulphonic” OR “nonafluoro 1 butanesulphonate” OR “1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulfonic” OR “1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulfonate” OR “1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulphonic” OR “1 1 2 2 3 3 4 4 4 nonafluorobutane 1 sulphonate” OR “nonafluorobutane 1 sulfonic” OR “nonafluorobutane 1 sulfonate” OR “nonafluorobutane 1 sulphonic” OR “nonafluorobutane 1 sulphonate” OR “1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulfonic” OR “1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulfonate” OR “1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulphonic” OR “1 1 2 2 3 3 4 4 4 nonafluoro 1 butanesulphonate” OR “perfluorobutane sulfonic” OR “perfluorobutane sulfonate” OR “perfluorobutane sulphonic” OR “perfluorobutane sulphonate” OR “perfluoro 1 butanesulfonic” OR “perfluoro 1 butanesulfonate” OR “perfluoro 1 butanesulphonic” OR “perfluoro 1 butanesulphonate” OR “pentyl perfluorobutanoate” OR “nonaflate” OR “perfluorobutane sulfonic” OR “perfluorobutane sulfonate” OR “perfluorobutane sulphonic” OR “perfluorobutane sulphonate” OR “nonafluorobutane sulfonic” OR “nonafluorobutane sulfonate” OR “nonafluorobutane sulphonic” OR “nonafluorobutane sulphonate” OR “perfluorobutane 1 sulfonic” OR “perfluorobutane 1 sulfonate” OR “perfluorobutane 1 sulphonic” OR “perfluorobutane 1 sulphonate” OR "TCR-282")	613

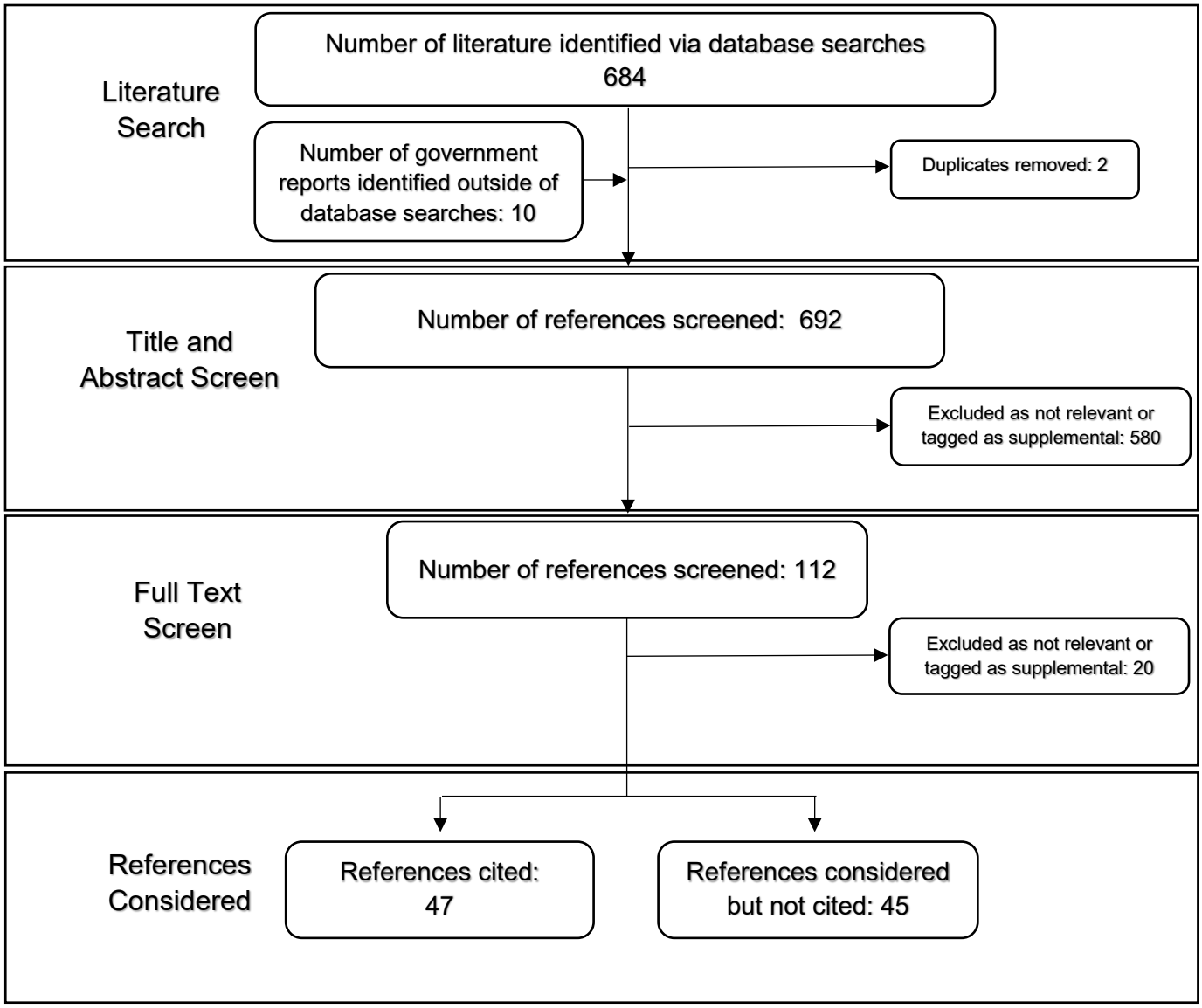
ToxNet – Search executed 12.3.2019

Search Terms	Results
"1 perfluorobutanesulfonic acid" OR "perfluorobutanesulfonic acid" OR 375-73-5 [rn] PubMed records excluded	0

PECO statement used for Tier 1 and Tier 2 literature screening

PECO element	Evidence
Populations	**Human:** Studies of any population and lifestage (occupational or general population, including children and other sensitive populations), including biomonitoring or exposure studies. **Animal:** Non-human mammalian animal species of any lifestage (including preconception, in utero, lactation, peripubertal, and adult stages). Zebrafish studies will be tagged as "potentially relevant supplemental information." **Mechanistic:** Studies of any human or animal (mammalian and non-mammalian) cell type, and mechanistic/genomic/in silico data with any biological significance.
Exposures	Relevant forms: Perfluorobutane Sulfonic Acid (CASRN 375-73-5), K^+PFBS (CASRN 29420-49-3), and any synonyms. If uncertain about chemical identity, please look it up. **Human:** Any exposure to PFBS via any route. **Animal:** Any exposure to PFBS via the oral route. Studies involving intraperitoneal or dermal exposures, or exposure to mixtures will be tagged as "potentially relevant supplemental information." **Mechanistic:** Any cell type exposed to PFBS alone. Studies involving exposures to mixtures will be tagged as "potentially relevant supplemental information."
Comparators	**Human:** A comparison or referent population exposed to lower levels (or no exposure/exposure below detection limits) of PFBS, or exposure to PFBS for shorter periods of time. Case reports and case series will be tracked as "potentially relevant supplemental information." **Animal:** A concurrent control group exposed to vehicle-only treatment or untreated control. **Mechanistic:** A concurrent control group of cells exposed to vehicle-only treatment or untreated control.
Outcomes	All health outcomes (both cancer and noncancer) and toxicokinetics. **Exclude**: ecological studies, animal biomonitoring studies, and reviews.
PBPK models	Studies describing PBPK models for PFBS will be included. Studies describing toxicokinetic data and ADME will also be included.

Flowchart of literature screen

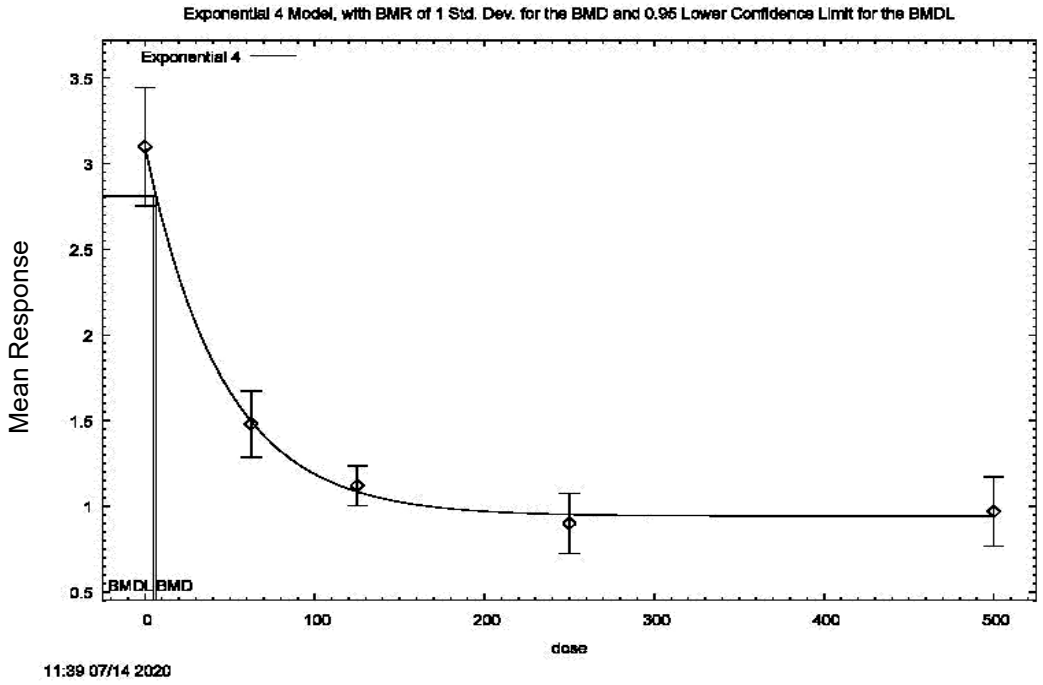


APPENDIX II. BENCHMARK DOSE MODELING

This appendix provides the BMD modeling outputs for PFBS toxicity data that were amenable to dose-response modeling. All models are run with default parameters and a benchmark response of 5% for dichotomous data and one standard deviation from the control mean for continuous data unless otherwise noted.

The model for tT4 in female rats (Figure A1) was run with modeled variance instead of the default constant variance. Model selection criteria when comparing outputs of different models for the same endpoint/dataset are: scaled residual ≤ the absolute value of two, goodness of fit p-value ≥0.05,[8] the Akaike's information criterion (AIC), and visual inspection of the dose-response curve. The lower limit of the 95% confidence interval of the BMD resulting in the benchmark response, the BMDL, is selected as the POD. The model selected for each study to derive candidate PODs is presented below.

Figure A1. Exponential model (M4) output for total T4 in female rats exposed to PFBS for 28 days (NTP, 2019)



Model Run Output for Figure A1: Exponential Model. (Version: 1.11; Date: 03/14/2017)

The form of the response function is: Y[dose] = a * [c-(c-1) * exp(-b * dose)]

[8] US EPA's Benchmark Dose Technical Guidance (2012) suggests using a goodness of fit p-value ≥ 0.1; however, models with less adequate fit (goodness of fit p-value ≥ 0.05) may be used when other criteria are taken into account, such as variability in the endpoint and visual fit.

A modeled variance is fit

Benchmark Dose Computation.

BMR = 1.0000 Estimated standard deviations from control

BMD = 10.2347

BMDL at the 95% confidence level = **6.89341**

Parameter Estimates

Variable	Estimate	Default Initial Parameter Values
lnalpha	-3.08282	-3.06165
rho	1.25696	1.30431
a	3.10137	3.255
b	0.0220902	0.00871856
c	0.30522	0.263331
d	n/a	1

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Resid
0	10	3.1	3.1	0.48	0.44	-0.009948
62.6	10	1.48	1.49	0.27	0.27	-0.08236
125	10	1.12	1.08	0.16	0.23	0.5226
250	9	0.9	0.96	0.23	0.21	-0.7963
500	9	0.97	0.95	0.26	0.21	0.3389

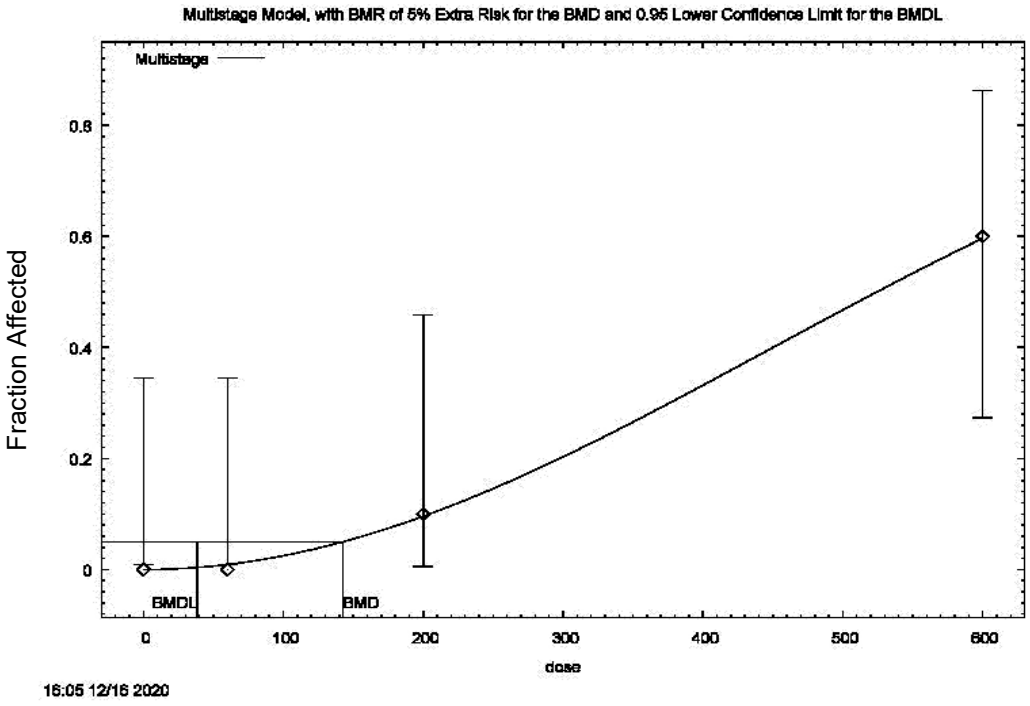
Likelihoods of Interest

Model	Log(likelihood)	# Param's	AIC
A1	35.84093	6	-59.68185
A2	42.58305	10	-65.1661
A3	40.92103	7	-67.84207
R	-17.56606	2	39.13213
4	40.44943	5	-70.89885

Tests of Interest

Test	-2*log(Likelihood Ratio)	Test df	p-value
Test 1	120.3	8	<0.0001
Test 2	13.48	4	0.009137
Test 3	3.324	3	0.3443
Test 6a	0.9432	2	0.624

Figure A2. Multistage 2º model output for incidence of kidney hyperplasia in female rats exposed to PFBS for 90 days (Lieder et al., 2009a)



Model Run Output for Figure A2: Multistage Model. (Version: 3.4; Date: 05/02/2014)

The form of the probability function is: P[response] = background + (1-background)*[1-EXP(-beta1*dose^1-beta2*dose^2...)]

Benchmark Dose Computation.

BMR = 5% Extra risk

BMD = 142.582

BMDL at the 95% confidence level = **38.485**

Parameter Estimates

Variable	Estimate	Default Initial Parameter Values
Background	0	0
Beta(1)	0	0.0000343191
Beta(2)	2.5231E-06	2.5006E-06

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	p-value
Full model	-9.98	4			
Fitted model	-10.07	1	0.183913	3	0.98
Reduced model	-18.55	1	17.1362	3	0

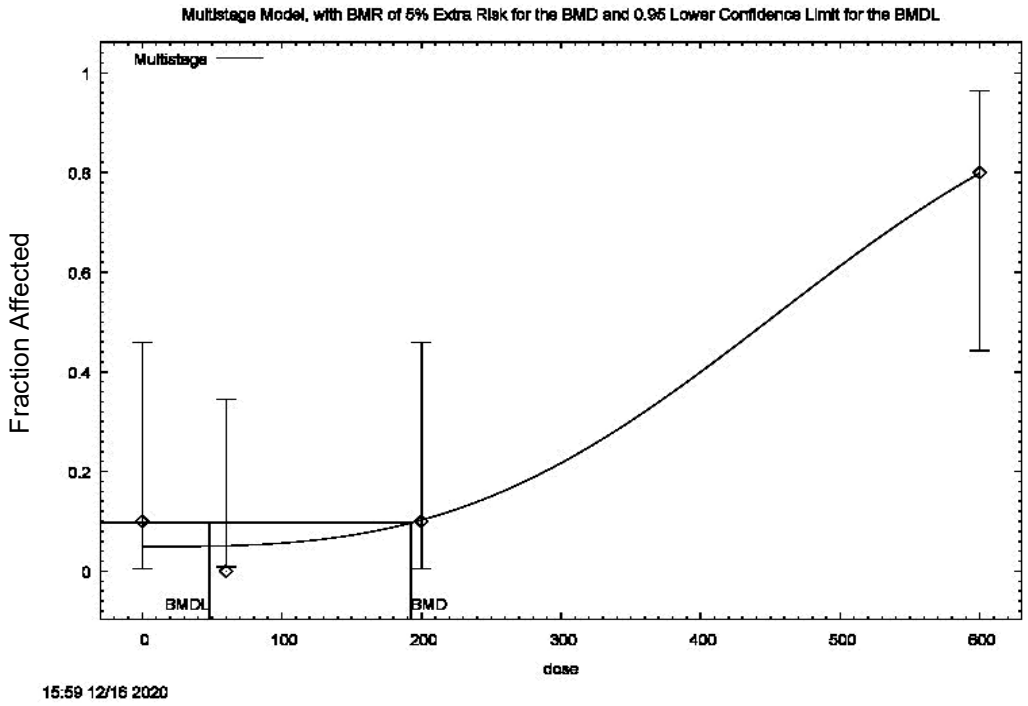
AIC: = 22.1458

Goodness of Fit Table

Dose	Est. Prob.	Expected	Observed	Size	Scaled Resid
0	0	0	0	10	0
60	0.009	0.09	0	10	-0.3
200	0.096	0.96	1	10	0.04
600	0.5968	5.968	6	10	0.02

Chi^2 = 0.09 d.f = 3 P-value = 0.9926

Figure A3. Multistage 3⁰ model output for incidence of kidney hyperplasia in male rats exposed to PFBS for 90 days (Lieder et al., 2009a)



Model Run Output for Figure A3: Multistage Model. (Version: 3.4; Date: 05/02/2014)

The form of the probability function is: P[response] = background + (1-background)*[1-EXP(-beta1*dose^1-beta2*dose^2...)]

Benchmark Dose Computation.

BMR = 5% Extra risk

BMD = 192.584

BMDL at the 95% confidence level = **48.3215**

Parameter Estimates

Variable	Estimate	Default Initial Parameter Values
Background	0.0494416	0.00902802
Beta(1)	0	0
Beta(2)	0	4.4199E-06
Beta(3)	7.1812E-09	0

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	p-value
Full model	-11.51	4			
Fitted model	-12.24	2	1.47094	2	0.48
Reduced model	-22.49	1	21.9754	3	<.0001

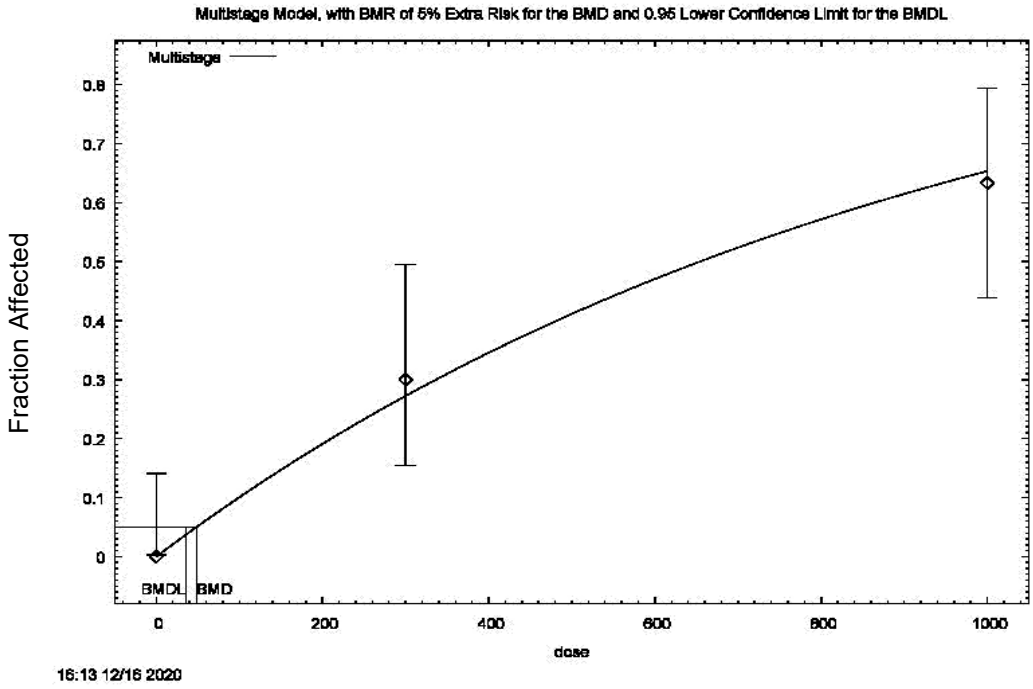
AIC: = 28.4823

Goodness of Fit Table

Dose	Est. Prob.	Expected	Observed	Size	Scaled Resid
0	0.0494	0.494	1	10	0.74
60	0.0509	0.509	0	10	-0.73
200	0.1025	1.025	1	10	-0.03
600	0.7985	7.985	8	10	0.01

Chi^2 = 1.08 d.f = 2 P-value = 0.5824

Figure A4. Multistage 2º model output for kidney hyperplasia in male rats exposed to PFBS 10 weeks prior to and through mating (Lieder et al., 2009b)



Model Run Output for Figure A4: Multistage Model. (Version: 3.4; Date: 05/02/2014)

The form of the probability function is: P[response] = background + (1-background)*[1-EXP(-beta1*dose^1-beta2*dose^2...)]

Benchmark Dose Computation.

BMR = 5% Extra risk

BMD = 48.4207

BMDL at the 95% confidence level = **35.6241**

Parameter Estimates

Variable	Estimate	Default Initial Parameter Values
Background	0	0.0243684
Beta(1)	0.00105933	0.000989205
Beta(2)	0	0

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	p-value
Full model	-38.04	3			
Fitted model	-38.12	1	0.166671	2	0.92
Reduced model	-55.8	1	35.5163	2	<.0001

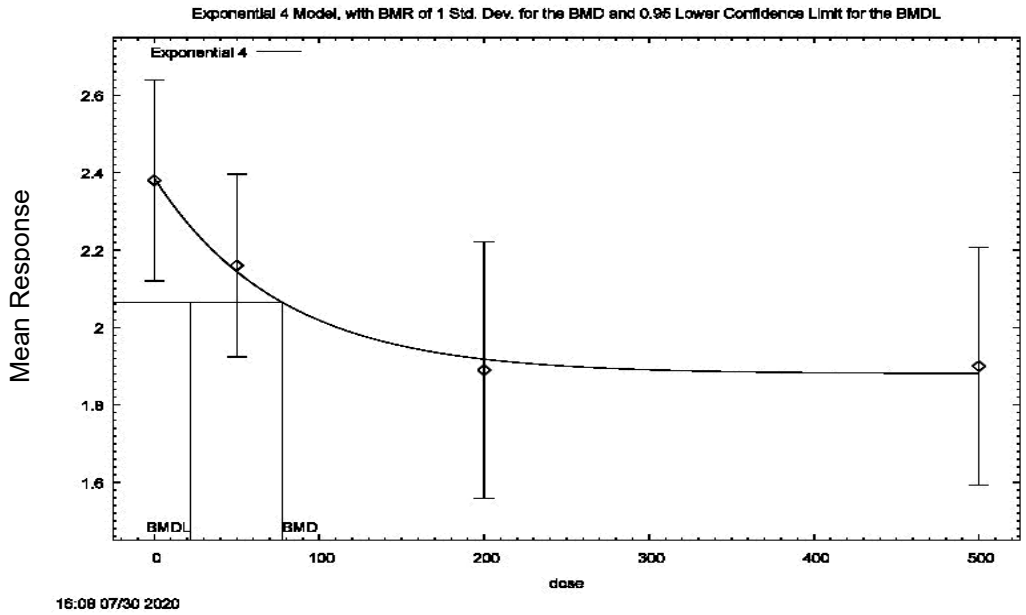
AIC: = 78.248

Goodness of Fit Table

Dose	Est. Prob.	Expected	Observed	Size	Scaled Resid
0	0	0	0	30	0
300	0.2722	8.167	9	30	0.34
1000	0.6533	19.599	19	30	-0.23

Chi^2 = 0.17 d.f = 2 P-value = 0.9188

Figure A5. Exponential model (M4) output for total T4 in GD20 mice exposed to PFBS (Feng et al., 2017)



Model Run Output for Figure A5: Exponential Model. (Version: 1.11; Date: 03/14/2017)

The form of the response function is: Y[dose] = a * [c-(c-1) * exp(-b * dose)]

A constant variance model is fit

Benchmark Dose Computation.

BMR = 1.0000 Estimated standard deviations from control

BMD = 77.5199

BMDL at the 95% confidence level = **22.057**

Parameter Estimates

Variable	Estimate	Default Initial Parameter Values
lnalpha	-2.27342	-2.27687
rho	n/a	0
a	2.38602	2.499

Variable	Estimate	Default Initial Parameter Values
b	0.0129944	0.00483893
c	0.788156	0.720288
d	n/a	1

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Resid
0	8	2.38	2.39	0.31	0.32	-0.05311
50	8	2.16	2.14	0.28	0.32	0.1365
200	8	1.89	1.92	0.4	0.32	-0.2481
500	8	1.9	1.88	0.37	0.32	0.1646

Likelihoods of Interest

Model	Log(likelihood)	# Param's	AIC
A1	20.42995	5	-30.85989
A2	20.98678	8	-25.97355
A3	20.42995	5	-30.85989
R	15.0267	2	-26.0534
4	20.37479	4	-32.74958

Tests of Interest

Test	-2*log(Likelihood Ratio)	Test df	p-value
Test 1	11.92	6	0.06377
Test 2	1.114	3	0.7738
Test 3	1.114	3	0.7738
Test 6a	0.1103	1	0.7398

APPENDIX III. DEFAULT UNCERTAINTY FACTORS FOR PHG DERIVATION

This appendix describes the default uncertainty factors OEHHA generally uses to calculate the Acceptable Daily Dose when deriving PHGs. When scientific evidence is compelling these defaults are supplanted by alternative factors or modeled results. Table A1 below is adapted from OEHHA's "Technical Support Document for the Development of Noncancer Reference Exposure Levels" (OEHHA, 2008).

Table A1. Default uncertainty factors for PHG derivation, adapted from OEHHA (2008)

Uncertainty Factor	Value
Interspecies uncertainty factor (UF_A)	
Combined interspecies uncertainty factor (UF_A):	1 human observation √10 animal observation in nonhuman primates 10 where no data are available on toxicokinetic or toxicodynamic differences between humans and a non-primate test species
Toxicokinetic component (UF_{A-k}) of UF_A:	1 where animal and human PBPK models are used to describe interspecies differences √10 non-primate studies with no chemical- or species-specific kinetic data
Toxicodynamic component (UF_{A-d}) of UF_A:	1 where animal and human mechanistic data fully describe interspecies differences. (*This is unlikely to be the case.)* 2 for residual susceptibility differences where there are some toxicodynamic data √10 non-primate studies with no data on toxicodynamic interspecies differences
Intraspecies uncertainty factor (UF_H)	
Toxicokinetic component (UF_{H-k}) of UF_H:	1 human study including sensitive subpopulations (e.g., infants and children), or where a PBPK model is used and accounts for measured inter-individual variability √10 for residual susceptibility differences where there are some toxicokinetic data (e.g., PBPK models for adults only) 10 to allow for diversity, including infants and children, with no human kinetic data
Toxicodynamic component (UF_{H-d}) of UF_H:	1 Human study including sensitive subpopulations (e.g., infants and children) √10 Studies including human studies with normal adult subjects only, but no reason to suspect additional susceptibility of children 10 Suspect additional susceptibility of children (e.g., exacerbation of asthma, neurotoxicity)

Uncertainty Factor	Value
LOAEL uncertainty factor (UF_L)	
Values used:	10 LOAEL, any effect 1 NOAEL or BMDL used
Subchronic uncertainty factor (UF_S)[1]	
Values used:	1 Study duration >12% of estimated lifetime √10 Study duration 8-12% of estimated lifetime 10 Study duration <8% of estimated lifetime
Database deficiency factor (UF_D)	
Values used:	1 No substantial data gaps √10 Substantial data gaps including, but not limited to, developmental toxicity

[1]Exposure durations of 13 weeks or less are subchronic regardless of species (OEHHA, 2008)

References

OEHHA (2008). Air Toxics Hot Spots Risk Assessment Guidelines: Technical Support Document for the Derivation of Noncancer Reference Exposure Levels. Office of Environmental Health Hazard Assessment, California Environmental Protection Agency, Sacramento, California.