# 2. RELEVANCE TO PUBLIC HEALTH

2.1 BACKGROUND AND ENVIRONMENTAL EXPOSURES TO PBDEs IN THE UNITED STATES

PBDEs are classes of structurally similar brominated hydrocarbons in which 2–10 bromine atoms are attached to the molecular structure (i.e., diphenyl ether). Monobrominated structures (i.e., one bromine atom attached to the molecule) are often included when describing PBDEs. There are 209 different molecular combinations, or congeners, that are possible for PBDEs, although only a limited number exist in commercial mixtures. Based on the number of bromine substituents, there are 10 homologous groups of PBDE congeners (monobrominated through decabrominated), with each homologous group containing one or more isomers. The mono-, di-, tri-, tetra-, penta-, hexa-, hepta-, octa-, nona-, and decabromo-congeners can exist in 3, 12, 24, 42, 46, 42, 24, 12, 3, and 1 isomers, respectively. The general chemical structure of PBDEs is shown below:

where m + n = 1 to 10
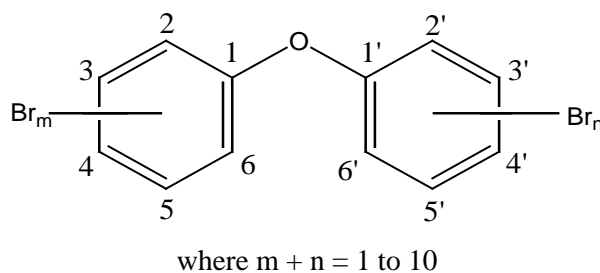
Due to the ether linkage and the position and number of bromine atoms, there are important three-dimensional differences in the structures of PBDEs that can influence the molecules' receptor interactions and toxicological properties as discussed in Section 3.5, Mechanisms of Action. In general, PBDEs are not expected to have the same array of three-dimensional conformations as either polybrominated biphenyls (PBBs) or polychlorinated biphenyls (PCBs).

PBDEs are brominated organic compounds that were used as flame retardant additives in plastics, textiles, and other materials. As additives, they are physically mixed into product applications, rather than chemically bound. Therefore, they have the potential to migrate from the plastic matrix into the environment when conditions are ideal. Production of PBDEs began in the 1970s and has continued until recently. PentaBDE and octaBDE mixtures were voluntarily withdrawn from the U.S. marketplace by their manufacturers at the end of 2004; however, the manufacture and use of decaBDE continued past that date. In December of 2009, the two remaining U.S. producers of decaBDE and the largest U.S. importer

of this product announced commitments to phase out manufacture and importation of decaBDE for most uses in the United States by December 31, 2012, and to end manufacture and import for all uses by the end of 2013.

PBDEs are persistent in the environment and most congeners can be considered bioaccumulative. The fully brominated congener, BDE 209, has a lower tendency to bioconcentrate due to its molecular dimensions; however, numerous studies have detected this substance in tissues of birds, mammals, and fish (see Section 6.4.4).

Monitoring and body-burden data indicate that PBDEs are ubiquitous in the environment and that the general population is exposed to these substances through their past use as flame retardants. A study that examined stored blood samples from 1973 (prior to the use of PBDEs as flame retardants) showed virtually no detections of these substances in human blood; however, varying concentrations of many PBDE congeners were detected in all blood samples collected in 2003 from 39 residents in Mississippi and 10 residents in New York City, illustrating the widespread exposure to PBDEs since their inception. Body burden data have consistently shown the residents of North America have higher concentrations of PBDEs in blood than people residing in Europe, likely due to differences in past production and use of commercial formulas.

House dust was identified as a major source of exposure to PBDEs by a systematic study of American exposure routes. In the United States, concentration levels found in soil, house dust, and air tended to be greater in indoor samples compared to outdoor samples. The EPA calculated the adult intake dose of total PBDEs to be 7.1 ng/kg body weight/day. It estimated children's intakes as 47.2 ng/kg body weight/day for 1–5 year olds, 13.0 ng/kg body weight/day for 6–11 year olds, and 8.3 ng/kg body weight/day for 12–19 year olds. The much higher dose for children aged 1−5 years was largely due to higher soil/dust ingestion in this age group. Exposure to indoor dust was the predominant exposure pathway for PBDEs in these calculations. It was estimated that 90% of the intake resulted from house dust inhalation or dermal exposure. This does not include special populations such as infants that are primarily exposed through breastfeeding. PBDE concentrations were generally lower in house dust samples collected outside of the United States compared with dust samples collected within the United States. The specific PBDE congeners detected in house dust and food vary; BDE 209 is more commonly detected within indoor environments where exposure is more likely to occur through intake of contaminated dust and air. PBDE contamination of food is more likely a result of past emissions or ongoing emissions from dumpsites and older products that still contain pentaBDE, which is mainly composed of the congeners

BDE 47 and BDE 99. Ingestion of PBDEs through the diet appears to be the predominant pathway for European communities. In China, the decaBDE congener, BDE 209, was the most abundant congener detected in both maternal and cord blood samples where industrial production of BDE 209 may result in exposure.

2.2   SUMMARY OF HEALTH EFFECTS

Information is available on the potential health effects of formerly used commercial PBDE mixtures (pentaBDE, octaBDE, and decaBDE) as well as several individual PBDE congeners. As subsequently discussed, the toxicity of decaBDE is generally less pronounced than for lower-brominated PBDEs following acute and repeated-dose exposures. This difference in toxicity may be related to differences in pharmacokinetics, resulting in lower bioavailability of decaBDE (see Sections 2.3 and 3.4 for more details).

The preponderance of health effects information on PBDEs is from studies of orally exposed laboratory animals and human studies in which the main exposure route is unknown, but expected to be oral. As summarized below and detailed in Chapter 3 (Health Effects), the main targets of concern following PBDE exposure in humans are the developing nervous and reproductive systems, the developing and mature endocrine system, the liver, and the male reproductive system. Other potential targets are the female reproductive system, the adult nervous system, and the developing and adult immune system; however, evidence for these end points is limited. In other systems/organs, available data provide no consistent evidence for exposure-related effects (respiratory, cardiovascular, gastrointestinal, hematological, renal, dermal, or ocular effects); therefore, effects in these systems are considered unlikely to occur following PBDE exposure.

Developmental Effects.

Neurodevelopment. Numerous epidemiological studies have reported results suggestive of an effect of PBDE on neurodevelopment in children. PBDE concentrations in cord blood, maternal or infant serum, and/or breast milk have been correlated with cognitive score and adaptive behavior deficits in infants; mental and physical development deficits in infants/toddlers at ages 12, 24, and 36 months; language and social developmental score deficits in toddlers at 24 months; increased impulsivity in toddlers at 24–36 months; poor social competence and attention deficit hyperactivity disorder (ADHD) or increased attention problems in 4-year-old children; impaired fine motor coordination, verbal memory and

comprehension, and sustained attention in 5–7-year-old children; and poor attention and executive function deficits in 9–12-year-old children. In one birth cohort, no associations were observed between maternal serum PBDEs and neonatal behavior in 5-week-old infants or autistic behaviors at 4–5-year-old children; however, children from the same cohort showed associations between maternal serum PBDEs and decreased IQ and increased hyperactivity at 5 years of age and executive function deficits at 5–8 years of age. Pre- and perinatal studies in animals also consistently reported neurodevelopmental effects following exposure to lower-brominated PBDEs and decaBDE at doses ≥0.06 and ≥2.22 mg/kg/day, respectively, including neurobehavioral alterations, delayed ontogeny of reflexes, ultrastructural changes in the hippocampus, altered nicotinic receptor density, altered electrophysiology, and altered gene and protein expression levels.

Based on human and animal data, the developing nervous system is a target of concern for both lower-brominated PBDEs and decaBDE.

Endocrine System Development. In infants, developmental exposure to PBDEs and infant serum or cord blood thyroxine (T_4) levels were either negatively associated or not associated. Associations between developmental PBDE exposure and infant serum or cord blood triiodothyronine (T_3) and thyroid stimulating hormone (TSH) were similarly inconsistent. In animals, numerous studies have reported decreased serum T_4 and/or T_3 levels in pups after gestational and lactational exposure to penta- or tetraBDE at doses as low as 0.3 mg/kg/day in rats and 452 mg/kg/day in mice. Significant reductions in serum T_3 levels were also observed in offspring following gestational and lactational exposure to decaBDE at 146 mg/kg/day in rats and 1,500 mg/kg/day in mice, although no changes were observed in serum T_4 or TSH. A dose-related decrease in serum T_4 was observed in neonatal male mice exposed to decaBDE doses of 6–20 mg/kg/day from postnatal day (PND) 2 to 15, but no change was observed in neonatal females.

While human data are inconsistent, they suggest that PBDEs can interact with thyroid hormone homeostasis in infants and children. These data, along with available animal studies, indicate that the developing thyroid is a target of concern for PBDE exposure, especially lower-brominated PBDEs.

Reproductive System Development. Male reproductive effects significantly associated with PBDE exposure in infants included congenital cryptorchidism (undescended testes), decreased cord serum total testosterone (but not free testosterone, estradiol [E2], aromatase index, sex hormone binding globulin, or Anti-Müllerian hormone), increased serum levels of the sex hormones, E2, free E2, and inhibin B (but not

testosterone, luteinizing hormone [LH], follicle-stimulating hormone [FSH], or sex hormone binding globulin) at 3 months of age, and increased testes volume in boys at 18 months of age. In contrast, no relationships were observed between maternal PBDE exposure and hypospadias (abnormal location of the urinary tract opening) in male offspring, PBDE concentrations in children's adipose tissue and cryptorchidism, or various measures of sexual maturation in female offspring. However, serum PBDE levels in 6–8-year-old females were significantly associated with delayed onset of puberty in a longitudinal cohort of U.S. girls.

In animal studies, reproductive effects were observed in adult F1 offspring of dams exposed to a single dose of pentaBDE at 0.06 mg/kg on gestation day (GD) 6, including reductions in testicular weight, sperm/spermatid number, and daily sperm production in males and a decreased number of secondary follicles and ultrastructural changes in the ovaries in females (although F1 fertility when mated to an unexposed animal was not impaired). In animals exposed pre- or perinatally to decaBDE, reproductive effects were observed in adult male offspring of dams exposed to decaBDE doses of 10–1,500 mg/kg/day from GD 0 to 17, including testicular lesions, decreased anogenital distance (AGD), and altered sperm parameters. In contrast, no exposure-related changes in AGD, onset of puberty, or reproductive organ weight and histology were reported in offspring of dams exposed to decaBDE at doses up to 1,000 mg/kg/day during gestation and lactation or doses up to 20 mg/kg/day from PND 2 to 15. No exposure-related changes in reproductive development were reported in female offspring of dams exposed to decaBDE at doses up to 1,000 mg/kg/day during gestation and lactation or doses up to 20 mg/kg/day from PND 2 to 15.

Based on limited human and adequate animal data, it is possible that oral PBDE exposure during development may adversely affect the developing reproductive system, particularly the male reproductive system. However, data are too limited to adequately determine whether or not PBDE exposure in infants and children will lead to altered reproductive performance as adults.

Immune System Development. In offspring of rat dams exposed to pentaBDE at doses up to 25 mg/kg/day via gavage for 70 days prior to mating through PND 21, a significant dose-related trend was observed in the incidence of apoptotic lymphocytes and tingible macrophages in the thymus of PND 43 males, but not females. In PND 28 offspring of mouse dams exposed to decaBDE at doses ≥260 mg/kg/day from GD 10 to PND 21, pulmonary viral titers of respiratory syncytial virus (RSV) (measured 5 days post-infection) were significantly increased. These animal data suggest that oral PBDE

exposure during development may lead to immunosuppression; however, data are too limited to adequately assess the immunotoxic potential of PBDE exposure in infants and children.

Embryotoxicity, Fetotoxicity, and Physical Growth and Development. No human studies have evaluated associations between embryotoxicity or fetotoxicity and PBDE exposure. One study reported a significant increased risk for preterm birth in women with high serum PBDE concentrations, compared with low PBDE concentrations; however, other studies did not observe a significant association between gestational length and PBDE concentrations. Evidence for altered physical growth and development from human studies is inconsistent, with some studies reporting associations between PBDE concentrations in maternal/cord serum, breast milk, or placental tissue and decreased birth weight, length, chest circumference, head circumference, and/or body mass index (BMI), some reporting no associations, and a limited number reporting increased birth weight, length, and/or head circumference. Available data from animal studies indicate that PBDEs are not embryotoxic or fetotoxic at PBDE doses below doses that elicited maternal toxicity, although occasional observations of reduced pup weight were reported.

Based on animal studies, it is unlikely that oral PBDE exposure will cause embryotoxicity or fetotoxicity in humans; however, human and animal data indicate that PBDE exposure could potentially lead to low birth weight or other impacts on physical development.

Endocrine Effects.

Thyroid. Numerous studies have been performed to evaluate the relationship between concentrations of PBDE in body tissues and circulating thyroid hormone levels in human populations. While these studies have demonstrated that PBDE can perturb the human endocrine system and affect hormone levels, the specific findings are not consistent across studies. For example, even limiting the discussion to studies that evaluated both PBDE concentrations and thyroid hormone levels in serum samples collected only from adult men, studies have reported positive associations with T_4, negative associations with T_4, and no association with T_4. Similar inconsistencies were found in studies in pregnant women, with studies reporting a positive associations with T_4, no association with T_4, or negative associations with T_4. Results were similarly inconsistent regarding the association between PBDE concentrations and serum T_3 and TSH.

In contrast to inconsistencies observed in human studies, altered serum thyroid hormone levels have been consistently reported in laboratory animals exposed to lower-brominated PBDEs. Reduced serum T_4 has

been reported in animals following acute or intermediate exposure to lower-brominated PBDEs at doses as low as 0.8 mg/kg/day. At higher doses ($\geq$30 mg/kg/day), some studies also report reduced serum T_3 and/or increased serum TSH; however, other studies reported no significant changes in serum T_3 and/or TSH levels in rats exposed to doses up to 300 mg/kg/day. In rat dams, reduced serum T_4 has been observed following exposure to lower-brominated PBDEs at doses as low as 0.06 mg/kg/day during gestation or gestation plus lactation. In mouse dams, no exposure-related changes were observed following exposure to pentaBDE at 452 mg/kg/day from GD 4 to PND 17. Exposure to pentaBDE at doses up to 120 mg/kg/day during gestation or gestation plus lactation did not significantly alter maternal serum T_3 and/or TSH in most studies; however, a study reported reduced maternal serum T_3 after exposure to pentaBDE at 30 mg/kg/day via dosed cookies from GD 1 to PND 21.

Histopathological changes in the thyroid (e.g., follicular cell hyperplasia, increased epithelial thickness/height, altered morphology of epithelium, cellular debris, degeneration) have been observed in intermediate-duration studies of lower-brominated BDEs at doses as low as 20 mg/kg/day in rats, 0.45 mg/kg/day in mice, and 0.06 mg/kg/day in mink. Increased thyroid weights were reported in rats exposed to doses of penta- or octaBDE as low as 50 mg/kg/day for 90 days; however, no exposure-related changes in thyroid weights were observed in rats exposed to doses of penta- or octaBDE up to 200 mg/kg/day for 15–28 days or in F0 or F1 mink exposed to dietary pentaBDE at doses up to 0.31 mg/kg/day in one-generation studies (4 weeks pre-mating through postnatal week [PNW] 6 or 33). In acute studies, no exposure-related changes in thyroid weight or histology were observed in rats exposed to penta- or tetraBDE doses up to 36 mg/kg/day for 14 days

Unlike the lower-brominated PBDEs, serum T_4 levels were not altered in rats exposed to decaBDE at doses up to 600 mg/kg/day for 4–90 days. Some studies reported reduced serum T_3 levels in rats exposed to decaBDE at doses as low as 50 mg/kg/day for 28–90 days, but another 90-day study reported no change in serum T_3 levels in rats exposed to decaBDE at doses up to 100 mg/kg/day. Serum TSH was reduced in male rats exposed to decaBDE at doses $\geq$300 mg/kg/day for 33 days. At higher doses ($\geq$950 mg/kg/day), significant reductions in serum T_4 and T_3 were observed in male mice exposed for 35 days and pregnant mice exposed from GD 7 to 9.

In chronic studies of decaBDE, thyroid follicular cell hyperplasia was observed in male mice exposed to $\geq$3,200 mg/kg/day for 103 weeks; no histopathological changes in the thyroid were observed at doses up to 7,780 mg/kg/day in female mice, 2,240 mg/kg/day in male rats, or 2,550 mg/kg/day in female rats. In intermediate-duration studies, dose-related increases in thyroid hyperplasia were reported for male rats

exposed to a low-purity decaBDE compound at ≥80 mg/kg/day for 30 days, but hyperplasia was not observed in rats or mice exposed to high-purity decaBDE at doses up 8,000 or 9,500 mg/kg/day, respectively, for 13 weeks or in rat dams exposed to doses up to 146 mg/kg/day from GD 10 to PND 21. However, multiple areas of degenerated follicular epithelium and slight attenuation of the follicular epithelium were observed in the thyroid glands of young male rats exposed to decaBDE at doses ≥300 mg/kg/day for 33 days. No changes in thyroid weight were observed in rats exposed to decaBDE at doses up to 90 mg/kg/day for 28 days, but increased thyroid weights were reported in rat dams exposed to ≥2 mg/kg/day from GD 10 to PND 21 and young male rats exposed to 600 mg/kg/day for 33 days.

While human data are inconsistent, they suggest that PBDEs can interact with thyroid hormone homeostasis. These data, along with available animal studies, indicate that the thyroid is a target of concern for PBDE exposure, especially lower-brominated PBDEs.

Pancreas. An analysis of cross-sectional National Health and Nutrition Examination Survey (NHANES) data showed a significant increase in the risk of diabetes associated with serum concentrations of BDE 153 (but not BDE 28, BDE 47, BDE 99, or BDE 100), although the risk was higher with exposure to 50–75th percentile BDE 153 concentrations than >75th percentile BDE 153 concentrations. Serum BDE 153 concentrations (but not BDE 28, BDE 47, BDE 85, BDE 99, BDE 100, or BDE 154) were also shown to be significantly associated with increased odds of developing gestational diabetes in a cohort of 258 pregnant women. However, other cross-sectional and prospective studies found no relationship between serum PBDE concentrations and diabetes in an adult cohort from Wisconsin, an elderly cohort in Finland, or an elderly cohort in Sweden.

An intermediate-duration study evaluated insulin-regulation and pancreatic morphology in rats following exposure to decaBDE at 0, 0.05, 1, or 20 mg/kg/day daily via gavage in corn oil for 8 weeks. Serum insulin was significantly decreased by 50–60% at 1 and 20 mg/kg/day, and glucose levels were concomitantly increased by 12, 18, and 21% at 0.05, 1, and 20 mg/kg/day. Consistent with the insulin findings, morphological changes were seen in the pancreas at 1 and 20 mg/kg/day (blurred boundaries among pancreatic islet cells; quantitative data not reported). Additionally, microarray analysis indicated that type I diabetes mellitus (T1DM) canonical pathways were significantly enriched following decaBDE exposure. Subsequently, gene act network and gene coexpression network found that some major histocompatibility complex molecules and TNF-α were involved in the T1DM pathway. Only one other animal study evaluated the pancreas following decaBDE exposure. In rats exposed to decaBDE via gavage for 28 days at doses of 0, 0.27, 0.82, 2.47, 7.4, 22.2, 66.7, or 200 mg/kg/day, slight or moderate

insulitis was observed in the Langerhan's islets of the "majority of samples," but findings were not exposure-related. Similarly, no exposure-related effects were observed for serum glucose levels. The only other study evaluating serum glucose levels after decaBDE exposure instead reported reduced serum glucose levels in male rats exposed to 20 mg/kg/day of a dietary PBDE mixture containing 52.1% pentaBDE (DE-71), 44.2% decaBDE (BDE 209), and 0.4% octaBDE (DE-79) for 70 days. The observed decreased glucose levels could be due to the pentaBDE component, as male rats exposed to pentaBDE at doses of 0.27–200 mg/kg/day for 28 days also showed decreased glucose levels; the study authors did not report the lowest dose at which glucose levels were significantly lower in male rats, but they reported a BMD_{10RD} of 179.55 mg/kg/day and a $BMDL_{10RD}$ of 66.7 mg/kg/day.

Limited human evidence is inconclusive regarding potential associations between diabetes and PBDE exposure; however, considering the animal data, the pancreas may be a target of concern for oral PBDE exposure.

Hepatic Effects. Liver effects have been reported in adult, pregnant, and developing animals exposed to lower-brominated PBDEs. Histopathological effects in the liver (hepatocellular hypertrophy, necrosis, and vacuolation) were consistently observed in animals exposed to lower-brominated PBDEs for 15–90 days at doses ≥0.45 mg/kg/day, regardless of life-stage. In acute exposure studies, fatty degeneration of the liver was observed following a single pentaBDE dose of 2,000 mg/kg or repeated pentaBDE doses of 200 mg/kg/day for 7 or 14 days. Increased liver weight was also observed in animals exposed to lower-brominated PBDEs for 1–14 days at doses ≥8 mg/kg/day and 15–90 days at doses ≥1.2 mg/kg/day. In studies that evaluated hepatic enzyme induction (e.g., elevated ethoxyresorufin o-deethylase [EROD], methoxyresorufin o-deethylase [MROD], pentoxy-resorufin o-deethylase [PROD], and uridine diphosphoglucuronyl transferase [UDPGT] activity), significantly increased enzyme activities were observed in animals following acute or intermediate-duration exposure to doses ≥3 or ≥0.06 mg/kg/day, respectively, and were always observed at doses at or below the dose causing elevated liver weights in the same study. No studies evaluating liver effects following chronic exposure to lower-brominated PBDEs were located.

Evidence for hepatic toxicity following exposure to decaBDE is less consistent than evidence for lower-brominated PBDEs. There is no evidence of hepatic toxicity following acute exposure to decaBDE at doses up to 1,000 mg/kg/day for 4–14 days. In intermediate-duration studies, slight to moderate hepatocellular hypertrophy was observed in rats exposed to decaBDE at 60 mg/kg/day for 28 days, in pregnant rats exposed to 300 mg/kg/day for 21 days, and in mice exposed to 9,400 mg/kg/day for

28 days; however, other studies did not report exposure-related changes in liver histology following exposure to decaBDE at doses up to 9,500 mg/kg/day for 3–13 weeks. In an older study using an impure decaBDE compound containing lower-brominated congeners (77% decaBDE, 22% nonaBDE, 0.8% octaBDE), centrilobular cytoplasmic enlargement and vacuolation, as well as increased liver weight, were observed in male rats exposed to dietary doses of ≥80 and 800 mg/kg/day, respectively, for 30 days. Increased liver weights were also reported in rats exposed to doses ≥1 mg/kg/day for 8 weeks and mice exposed to 9,400 mg/kg/day for 28 days, but other intermediate-duration studies reported no exposure-related changes in liver weights at doses of 1,000 mg/kg/day for 21–90 days. In chronic studies, exposure to decaBDE for 103 weeks caused liver lesions that included neoplastic nodules in rats at ≥1,120 mg/kg/day, thrombosis and degeneration in rats at 2,240 mg/kg/day, and centrilobular hypertrophy and granulomas in mice at ≥3,200 mg/kg/day.

Developing animals appear to be more susceptible to liver damage following exposure to decaBDE than adult animals. Transient histopathological changes (diffuse liver cell hypertrophy with increased cytoplasmic eosinophilia) and elevated liver weights were observed in male and female rat offspring exposed to decaBDE from GD 10 to PND 21 at ≥2 and 146 mg/kg/day, respectively. Fatty degeneration, elevated liver weights, and elevated liver enzymes were observed in young male rats exposed to decaBDE at ≥300 mg/kg/day from PND 10 to 42. In mice, decaBDE exposure from GD 0 to 17 caused acute cell swelling of hepatocytes associated with pressure occlusion of hepatic sinusoids and elevated liver enzymes in PND 71 male offspring at ≥10 mg/kg/day; however, liver weight was unaltered at doses up to 600 mg/kg/day.

No studies are available on hepatic effects of PBDEs in humans. Based on the evidence in animals, PBDEs are potentially hepatotoxic in humans, especially lower-brominated PBDEs.

Male Reproductive Effects. Several studies have found results suggestive of reproductive effects in men associated with exposure to PBDE, including significant inverse correlations between serum concentrations of BDE 153 (hexaBDE) and sperm concentration and testis size in young adult Japanese males, significantly reduced sperm mobility in association with increased serum PBDE concentrations (BDE 47, BDE 100, and total) in Canadian men recruited at a fertility clinic, and altered parameters of semen quality associated with selected BDEs in men participating in a prospective cohort study in Michigan and Texas. Although a number of studies have evaluated the potential effects of PBDE exposure on male reproductive hormone levels, these studies collectively do not show consistent effects associated with PBDE exposure.

2. RELEVANCE TO PUBLIC HEALTH

Reproductive effects have been reported in male rodents following intermediate-duration exposure to the lower-brominated PBDE congener tetraBDE, including decreased serum testosterone at ≥0.001 mg/kg/day, histopathological changes in rat or mouse testes (increased epithelial thickness, multinucleated giant cells, vacuolar spaces, apoptosis, germ cell loss) at doses ≥0.03 or ≥0.045 mg/kg/day, and decreased sperm production in rats at 1 mg/kg/day. No dose-related changes were observed in testicular weight or sperm morphology, motility, or capacitation at doses up to 30 mg/kg/day. For other lower-brominated PBDEs (pentaBDE, octaBDE), no exposure-related effects were observed in serum testosterone levels at doses up to 60 mg/kg/day for 15–70 days, sperm parameters at doses up to 20 mg/kg/day for 20 days, or male reproductive histology or organ weight at doses up to 750 mg/kg/day for 38–90 days. However, in acute studies, serum testosterone levels were significantly decreased by ~40–45% in male rats 45 days after a single gavage exposure to pentaBDE at doses ≥0.6 mg/kg and dose-related decreases in androgen-dependent tissue weights (prostate, seminal vesicle, Cowper's gland, gland penis, levator ani bulbo cavernosus) were observed in castrated rats exposed to pentaBDE at doses ≥30 mg/kg/day for 9 days (Herschberger assay).

Studies of decaBDE have been more limited. Decreased relative testes and epididymides weights, degenerative changes in the seminiferous tubules, reduced sperm count and viability, and reduced serum testosterone were reported in male mice exposed to decaBDE at 950 mg/kg/day via gavage for 35 days, compared with controls; no exposure-related effects were observed at 750 mg/kg/day. In contrast, no changes in sperm count, motility, or morphology were observed in rats or mice exposed to decaBDE at doses up to 60 or 1,500 mg/kg/day for 28–50 days; however, exposure-related decreases were observed in one sperm velocity measure (lateral head amplitude) in mice exposed to ≥500 mg/kg/day. A dose-related decrease in epididymis weight and a dose-related increase in seminal vesicle/coagulation gland weight were observed in rats exposed to 1.7–60 mg/kg/day for 28 days; however, the lowest doses at which the effects were observed were not reported. No testicular weight changes were observed at doses up to 60 mg/kg/day. In other intermediate-duration studies, no exposure-related changes in organ weight were reported for male reproductive organs in rats or mice exposed to decaBDE doses up to 800 or 1,500 mg/kg/day. Histopathological changes in male reproductive tissues have not been reported in rats or mice exposed to decaBDE at doses up to 2,550 or 7,780 mg/kg/day, respectively, for 103 weeks. Findings were negative in a one-generation study that exposed male and female rats to an impure decaBDE compound containing lower-brominated congeners (77% decaBDE, 22% nonaBDE, 0.8% octaBDE) for 60 days prior to mating through PND 21.

Based on the evidence in humans and animals, lower-brominated PBDEs are potentially toxic to the male reproductive system in humans. Available data for decaBDE provide very limited evidence of male reproductive damage.

Female Reproductive Effects.   Evidence for reproductive effects in women associated with exposure to PBDEs is inconsistent. Increased length of menstrual periods (prior to pregnancy) was associated with increased breast milk concentrations of PBDEs in a study of 46 Taiwanese women, and with plasma levels of BDE 47 and BDE 153 in a study of 42 Cree women of James Bay, Canada, but not in a smaller study with 20 Taiwanese women or in 223 Californian women. Age at menarche was not associated with PBDE concentrations in breast milk; however, an analysis of cross-sectional data from a sample of 271 adolescent girls (NHANES) found that higher serum PBDE concentrations were associated with younger age of menarche. Decreased fecundability (i.e., increases in time to pregnancy between stopping contraception and becoming pregnant) was significantly associated with increased serum concentrations of BDE 47, BDE 99, BDE 100, and BDE 153 (and their sum) in 223 Californian women; however, fecundability was not significantly related to serum PBDE concentrations in a cohort of 501 Michigan and Texas couples followed prospectively for 1 year after discontinuing contraception for the purpose of becoming pregnant. A study of 65 women from Boston undergoing *in vitro* fertilization found no association between serum PBDE concentrations and risk of implantation failure, but did find a significantly increased risk of failure associated with increased (i.e., above median) concentrations of BDE 153 (but not other congeners or total PBDE) in follicular fluid. PBDEs were not associated with Polycystic Ovary Syndrome or with diagnosis of uterine fibroids.

In one-generation animal studies, no exposure-related changes were observed in reproductive end points (number of pregnancies, gestation length, number, size, or sex ratio of litters) in rats or mice exposed to lower-brominated PBDEs at doses up to 25 or 1 mg/kg/day, respectively. Similarly, in gestation plus lactation studies, no exposure-related effects on litter parameters (successful delivery of litters, gestation length, litter size, sex ratio, number of live pups) were observed in rats or mice exposed to lower-brominated PBDEs at doses up to 32 or 10 mg/kg/day, respectively, during gestation and lactation only. The number of litters surviving until PND 8 was significantly decreased following exposure to tetraBDE at 0.1 mg/kg/day from pre-mating day 28 through PND 21 in one study; however, reduced pup survival was not reported in other studies. In a one-generation study in mink, females exposed to pentaBDE at doses ≥0.25 mg/kg/day from pre-mating day 28 through PNW 6 did not whelp. It is not clear in one study whether mink exposed to 0.25 mg/kg/day never became pregnant or had complete litter loss. However, another study reported that female mink exposed to 0.31 mg/kg/day had no exposure-related changes in

mating success; rather, sows showed complete litter loss with 70% showing clear postimplantation loss.
In one-generation and intermediate-duration studies, no changes in reproductive organ weight or
histology were observed in female rats, mice, or mink exposed to lower-brominated PBDEs at doses up to
750, 0.45, or 0.31 mg/kg/day, respectively. One acute study reported increased paired ovary weight after
exposure to tetraBDE at 0.14 mg/kg on GD 6; however, no changes in reproductive organ weight or
histology were observed in female rats acutely exposed to pentaBDE at doses up to 300 mg/kg/day.

In female mice exposed to dietary tetraBDE for 28 days, serum testosterone and E2 were significantly
increased at 0.45 mg/kg/day; no other study reported altered reproductive hormones in females.

Information on reproductive effects of decaBDE is limited. Findings were negative in a one-generation
study that exposed male and female rats to an impure decaBDE compound contaminated with lower-
brominated congeners (77% decaBDE, 22% nonaBDE, 0.8% octaBDE) for 60 days prior to mating
through PND 21. In a gestational exposure study in mice (GDs 7–9), significant increases were observed
in the percentage of postimplantation loss per litter and resorptions per litter in dams exposed to ≥750 and
≥1,500 mg/kg/day, respectively. Additionally, the percentage of live fetuses per litter was significantly
decreased by 10% in dams exposed to 2,000 mg/kg/day. Histological changes in the ovaries (atrophic
changes, decreased number of follicles, and increased fibrotic tissue) were observe in female rats exposed
to decaBDE at 300 mg/kg/day from 3 weeks of age, through mating to untreated males, gestation, and
lactation (~11 weeks); F0 reproductive success was not reported. In other studies, no histopathological
changes in female reproductive organs were observed in rats or mice following intermediate- or chronic-
duration exposure to decaBDE at doses up to 8,000 or 9,500 mg/kg/day, respectively.

Based on inconsistent data in humans and animals, it is unclear whether PBDEs affect the female
reproductive system in adults.

Adult Neurological Effects.   While neurobehavioral development is a potential effect of concern for
PBDE exposure in humans, available human data are too limited to determine if PBDE exposure is
neurotoxic in adults or adolescents. No association was found between serum PBDE concentrations and
neuropsychological function assessed by 34 tests of cognitive and motor function, affective state, and
olfactory function in a study population of 144 volunteers (67 males and 77 females) between the ages of
55 and 74 years who lived for at least 25 years in the upper Hudson valley of New York State. In
515 secondary students from Belgium (mean age 14.9 years), serum PBDE concentrations were not
associated with most aspects of neurological performance measured in a battery of neurological tests;

however, there was a significant deterioration in performance in the finger tapping test with increasing PBDE level, suggesting an effect of PBDE on motor activity.

Evidence for neurological effects of lower-brominated PBDEs in adult animals is limited. In repeat-exposure neurobehavioral studies, impaired learning and memory were observed in male rats exposed to tetraBDE at ≥0.1 mg/kg/day for 30 days and impaired attention and inhibitory control were observed in male mice exposed to pentaBDE at 26.2 mg/kg/day for 125 days. No exposure-related neurobehavioral changes were observed in rats exposed to pentaBDE at doses ≤17.5 mg/kg/day for 90–125 days or male rats exposed once to pentaBDE doses up to 1.2 mg/kg/day. No exposure-related changes in brain weight and/or histology were observed in animals exposed to lower-brominated PBDEs at doses up to 750 mg/kg/day.

Evidence for neurological effects of decaBDE in adult animals is extremely limited. Decreased anxiety behavior in the elevated-plus maze was observed in male mice exposed to decaBDE for 15 days; however, no exposure-related changes were observed in anxiety behaviors in the light/dark test, in learning or memory in the Morris water maze, or in general neurological behaviors assessed using a functional observation battery. In another study, no changes were observed in open-field behavior of male rats exposed to decaBDE at doses up to 50 mg/kg/day via gavage for 90 days. No changes in brain weight were observed in rats or mice exposed to decaBDE at doses up to 90 or 160 mg/kg/day, respectively, for 15–60 days. No overt signs of neurotoxicity were observed in rats and mice exposed to decaBDE in estimated dietary doses as high as 16,000–19,000 mg/kg/day for 14 days, 8,000–9,000 mg/kg/day for 13 weeks, or 2,550–7,780 mg/kg/day for 103 weeks. Although the high doses and extended exposure durations provided opportunities for the induction and/or development of clinical signs, the study is limited by lack of testing for subtle behavioral changes and neurodevelopmental effects.

Based on available data in humans and animals, it is unclear whether PBDEs affect the adult nervous system.

Immunological and Lymphoreticular Effects.   Limited human data regarding potential immunotoxic effects of PBDEs are available. A significant negative association was found between serum concentrations of lower-brominated PBDE and number of circulating lymphocytes in a subset of a cohort of 33 adolescent children from the Netherlands. No effects on pokeweed mitogen-stimulated DNA proliferation or IgG immunoglobulin synthesis were found in human lymphocytes exposed to lower-

brominated PBDEs *in vitro*. Studies of Swedish subjects reported a significantly negative association between serum levels of BDE 47 and levels of protein complement 3, but not with levels of multiple inflammatory markers. In a study of Chinese children, serum levels of BDE 28 and 209 were associated with an increased risk of asthma.

There is limited evidence for impaired immune function in animals following exposure to lower-brominated PBDEs; however, comprehensive immunological evaluations have not been performed on any congener or previously used commercial mixture. The plaque-forming splenic cell antibody response to injected sheep red blood cells was significantly reduced in mice exposed to 72 mg/kg/day pentaBDE for 14 days; single doses as high as 500 mg/kg had no effect. In the same study, exposure to up to 72 mg/kg/day had no effect on natural killer cell (NKC) activity. *In vitro* production of IgG immunoglobulin from pokeweed mitogen-stimulated splenocytes was reduced in mice exposed to 36 mg/kg/day pentaBDE for 14 days. Other 14-day studies in mice found no changes in NKC activity to murine YAC-1 target cells at pentaBDE doses up to 72 mg/kg/day or numbers of splenic and thymic lymphocyte subsets at pentaBDE doses up to 36 mg/kg/day, although 18 mg/kg/day of tetraBDE caused significantly reduced numbers of total lymphocytes and CD4+, CD8+, and CD45R+ subtypes in spleen. In the only intermediate-duration study evaluating immune function, no dose-related changes were observed in antibody-mediated immunity to keyhole limpet hemocyanin (KLH) or phytohemagglutinin (PHA) skin response in mink exposed to pentaBDE at doses up to 0.78 mg/kg/day for 9 weeks.

Histopathological changes in the spleen (hyperplasia, germinal center development) were reported in some studies following intermediate-duration exposure to lower-brominated PBDEs at doses as low as 0.63 mg/kg/day in mink and at 0.45 mg/kg/day in mice. In other studies, no exposure-related changes were observed in spleen, thymus, lymph node, and/or bone marrow tissue histology in rats exposed to lower-brominated PBDEs at doses up to 750 mg/kg/day for 28–90 days. Additionally, no exposure-related changes were observed in the histology of the spleen, thymus, Peyer's patches, or mesenteric lymph nodes in rat dams exposed to pentaBDE at doses up to 25 mg/kg/day via gavage for 70 days prior to mating through PND 21 (~21 weeks).

Exposure to decaBDE at 1,800 mg/kg/day for 28 days did not cause increased pulmonary viral titers of RSV (measured 5 days post-infection) in mice. In rat dams exposed to 300 mg/kg/day from 21 days prior to mating through PND 21, altered T-lymphocyte cell population distribution in the thymus and a significantly reduced response to *in vitro* PHA exposure in cultured lymphocytes were observed. In another study, no dose-related changes were reported for T-cell, B-cell, or macrophage population

distribution in the spleen of rats exposed to decaBDE at doses up to 60 mg/kg/day via gavage for 28 days.
In a high-dose study, female mice exposed to decaBDE at 800 mg/kg every other day showed impaired
CD4 T-cell function from 4 to 10 months of exposure, compared with controls.

Chronic ingestion of decaBDE caused splenic lesions (hematopoiesis, fibrosis, lymphoid hyperplasia) in
rats exposed to ≥1,200 mg/kg/day for 103 weeks. After exposure for 13 weeks, histopathological
examinations of spleen, thymus, lymph node, and/or bone marrow tissues showed no effects in rats or
mice exposed to decaBDE at doses up 8,000 or 9,500 mg/kg/day. In rat dams exposed to decaBDE at
300 mg/kg/day for 21 days prior to mating through PND 21, lesions of the thymus (thickened thymus
capsule, decreased lymphoid tissue in the cortex with adipose tissue replacement, increased medulla size,
and obscured corticomedullary junction) and spleen (decreased size and number of lymphoid nodules,
thinner lymphatic sheath around arteries, and fibrotic tissue with macrophages in the medulla) were
observed.

Evidence from animals suggests that PBDE exposure may cause immune suppression, but data are limited
and inconsistent. Additionally, comprehensive immunological evaluations have not been performed and
human data are extremely limited. Therefore, currently available information is insufficient to adequately
characterize the human immunotoxic potential of PBDEs.

Cancer.   In human case-control epidemiological studies, no clear associations have been found between
non-Hodgkin's lymphoma risk and exposure to BDE 47 in a group of Swedish men and women including
19 cases and 27 controls, testicular cancer risk and serum PBDE (sum of BDE 47, BDE 99, and
BDE 153) in a small group of Swedish men and women including 58 cases and 58 controls, breast cancer
risk and adipose concentrations of PBDE (BDE 47, BDE 99, BDE 100, BDE 153, BDE 154, and their
sum) in a group of women from the San Francisco Bay area of California including 78 cases and
56 controls, breast cancer and BDE 47 in serum in native Alaskan women, thyroid cancer and serum
PBDE (BDE 47, BDE 99, BDE 100, BDE 153, and their sum) in participants in a large multicenter
clinical trial in the United States that included 104 cases and 208 controls, or prostate cancer and serum
levels of BDE 47 in a study involving 208 prostate cancer incident cases and 268 controls in Singaporean
males. In a study examining the association between exocrine pancreatic cancer risk and PBDE
concentrations in adipose tissue (sum of BDE 28, BDE 47, BDE 66, BDE 100, BDE 99, BDE 85,
BDE 154, BDE 153, BDE 138, and BDE 183) in a group of Swedish men and women, PBDE
concentrations were significantly higher in the 21 cases compared with the 59 controls. Case-control
analysis found that the risk of pancreatic cancer was not significantly increased with lipid PBDE using

median concentration in controls as a cut-off after adjustment for age, sex, and BMI at tissue sampling; however, the increase in risk was significant when the BMI adjustment was performed for the year before tissue sampling (body weight 1 year before tissue sampling obtained by questionnaire).

For most PBDEs, including pentaBDE and octaBDE, animal studies of carcinogenic effects are not available; cancer data on PBDEs in animals are limited to results of studies on commercial decaBDE products. In a bioassay conducted by the National Toxicology Program (NTP), male and female rats were exposed to high purity commercial decaBDE (lots that were 96 or 94–97% pure) in the diet in low doses of 1,120 and 1,200 mg/kg/day, respectively, and high doses of 2,240 and 2,550 mg/kg/day, respectively, for 103 weeks. Male and female mice were similarly exposed to low doses of 3,200 and 3,760 mg/kg/day, respectively, and high doses of 6,650 and 7,780 mg/kg/day, respectively. Incidences of neoplastic nodules in the liver were significantly increased in the male and female rats, although the term neoplastic nodule is poorly defined and understood, and is no longer used by NTP to characterize hepatoproliferative lesions in rats. Incidences of hepatocellular adenoma or carcinoma (combined) were significantly increased in the male mice. Slightly elevated incidences of thyroid gland follicular cell adenoma or carcinoma (combined) were additionally observed in exposed male mice, although the increases were not statistically significant. Carcinogenicity was also evaluated in rats that were exposed to 0.01, 0.1, or 1.0 mg/kg/day dietary doses of a 77.4% decaBDE mixture (containing 21.8% nonaBDE and 0.8% octaBDE) for approximately 2 years. No exposure-related neoplastic changes were found, but the power of this study to detect carcinogenic effects is limited by the very low dose levels in comparison to those tested in the NTP bioassay.

The EPA hazard descriptor for decaBDE is "suggestive evidence of carcinogenic potential" based on: (1) no studies of cancer in humans exposed to decaBDE; (2) a statistically significant increase in incidence of neoplastic nodules and a slight increase in incidence of carcinomas (not statistically significant) in the liver of low- and high-dose male rats and high-dose female rats; (3) a significantly increased incidence of hepatocellular adenoma or carcinoma (combined) in male mice at the low dose and marginally increased incidence at the high dose; (4) a nonsignificantly increased incidence of hepatocellular adenoma or carcinoma (combined) in female mice; (5) a slightly greater (but statistically not significant) incidence of thyroid gland adenomas or carcinomas (combined) in dosed male and female mice; (6) a significantly increased incidence in male mice, at both doses, of follicular cell hyperplasia, considered by many as a precursor to thyroid tumors; and (7) an apparent absence of genotoxic potential. DecaBDE has been classified as a Group 3 carcinogen (*not classifiable as to its carcinogenicity to humans*) by the International Agency for Research on Cancer (IARC) based on inadequate evidence of

carcinogenicity in humans and inadequate or limited evidence in experimental animals. The EPA assigns the cancer category Group D (*not classifiable as to human carcinogenicity*) to mono-, di-, tri-, tetra-, penta-, hexa-, octa-, and nonaBDEs and reports "*inadequate information*" to classify the specific congeners 2,2',4,4'-tetraBDE, 2,2',4,4',5-pentaBDE, and 2,2',4,4',5,5'-hexaBDE. The Department of Health and Human Services has not evaluated PBDEs for carcinogenicity. ACGIH has no data regarding cancer classifications for PBDEs.

2.3 MINIMAL RISK LEVELS (MRLs)

Estimates of exposure levels posing minimal risk to humans (MRLs) have been made for PBDEs. An MRL is defined as an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects (noncarcinogenic) over a specified duration of exposure. MRLs are derived when reliable and sufficient data exist to identify the target organ(s) of effect or the most sensitive health effect(s) for a specific duration within a given route of exposure. MRLs are based on noncancerous health effects only and do not consider carcinogenic effects. MRLs can be derived for acute, intermediate, and chronic duration exposures for inhalation and oral routes. Appropriate methodology does not exist to develop MRLs for dermal exposure.

Although methods have been established to derive these levels (Barnes and Dourson 1988; EPA 1990), uncertainties are associated with these techniques. Furthermore, ATSDR acknowledges additional uncertainties inherent in the application of the procedures to derive less than lifetime MRLs. As an example, acute inhalation MRLs may not be protective for health effects that are delayed in development or are acquired following repeated acute insults, such as hypersensitivity reactions, asthma, or chronic bronchitis. As these kinds of health effects data become available and methods to assess levels of significant human exposure improve, these MRLs will be revised.

People are environmentally exposed to PBDE mixtures of different congeneric composition than previously used commercial PBDE products. Although the toxicity or potency of environmental mixtures of congeners consequently may be greater or less than that of the commercial PBDE mixtures or individual congeners, there are insufficient mixture toxicity data on which to directly base MRLs for environmental PBDEs. Due to the likelihoods that (1) multiple mechanisms (aryl hydrocarbon receptor [AhR]-receptor-dependent mechanisms, AhR-independent mechanisms, or both) may be involved in health effects induced by PBDEs, (2) different PBDE congeners may produce effects by different mechanisms, and (3) humans are exposed to complex mixtures of interacting PBDEs with differing

biological activities, as well as to the lack of a suitable approach for quantitatively evaluating joint toxic action from concurrent exposures to PBDEs, PBBs, PCBs, chlorinated dibenzo-p-dioxins (CDDs), and/or chlorinated dibenzofurans (CDFs) in the environment, data from previously used commercial PBDE mixtures and individual congeners were reviewed to develop MRLs for assessing health risks from environmental exposures to PBDEs.

Separate MRLs were derived for lower-brominated PBDEs and decaBDE based on important differences in pharmacokinetics and toxicity of decaBDE compared to lower-brominated PBDEs. The most recent and best available estimates of oral absorption efficiencies for PBDE congeners indicate a range of 10–26% for decaBDE (BDE 209) and 70–85% for tetraBDE (BDE 47), pentaBDE (BDE 99, BDE 100), and hexaBDE (BDE 153, BDE 154) (Chen et al. 2006; Hakk et al. 2002a, 2002b, 2009; Morck and Klasson Wehler 2001; Morck et al. 2003; Örn and Klasson-Wehler 1998; Riu et al. 2008; Sandholm et al. 2003; Sanders et al. 2006a, 2006b; Staskal et al. 2005). Consistent with the higher absorption efficiencies of the lower-brominated congers, the animal toxicity database indicates that toxic effects occur at lower doses following exposure to lower-brominated PBDEs than following exposure to decaBDE. For example, the acute exposure levels required to cause neurobehavioral effects in mice are higher for decaBDE (≥ 2.22 mg/kg) than penta-, tetra-, and hexaBDE (≥ 0.8, ≥ 1, and ≥ 0.45 mg/kg, respectively) (Eriksson et al. 2001; Gee and Moser 2008; Johansson et al. 2008; Sand et al. 2004; Viberg et al. 2003a, 2003b, 2004a, 2004b).

Inhalation MRLs

Lower-brominated BDEs. Derivation of an acute-duration MRL for lower-brominated BDEs is not recommended at this time due to insufficient information. The inhalation database for acute-duration exposure to PBDEs is essentially limited to a single 14-day unpublished industry-sponsored study of octaBDE in rats (Great Lakes Chemical Corporation 1978). In this study, groups of five male and five female Charles River CD rats were whole-body exposed to dust of an unspecified commercial octaBDE mixture in mean analytical concentrations of 0, 0.6, 3.7, 23.9, or 165.2 mg/m^3 for 8 hours/day for 14 consecutive days. The average mass median aerodynamic diameter (MMAD) and geometric standard deviation (GSD) of the particles were 3.5 µm and 2, respectively. Study end points included clinical signs (including observations for respiratory distress and nasal and ocular irritation), body weight and food consumption, hematology (5 indices), blood chemistry (5 indices, thyroid hormones not assessed), urinalysis (10 indices), organ weights (5 organs including thyroid/parathyroid), gross pathology, and histology (21 tissues including nasal turbinates, trachea, lungs, and thyroid). The clinical laboratory tests

were limited to rats in the control and two highest dose groups. The histological exams were limited to
the control and highest dose groups, except for the liver, which was examined in all groups. Signs of
increased respiration rate (rapid breathing) were observed by the end of each exposure period in rats
exposed to ≥ 24 mg/m^3; this effect always disappeared by the following morning. Liver weight was
significantly increased and hepatic lesions occurred in rats exposed to ≥ 3.7 mg/m^3. At 3.7 mg/m^3, the
liver lesions consisted of very slight to slight, focal to multifocal cytoplasmic enlargement of the
hepatocytes, accompanied by focal acidophilic degeneration of individual to small groups of cells. The
liver lesions were similar in the higher dose groups except that the hepatocyte enlargement was multifocal
to diffuse in distribution, and there were focal, small to large areas of hepatocellular necrosis present to a
very slight to marked degree. There were no exposure-related histological changes in other tissues.

As detailed above, hepatocellular hypertrophy accompanied by some degenerative hepatocellular changes
was found following exposure to octaBDE at concentrations ≥ 3.7 mg/m^3 for 14 days (Great Lakes
Chemical Corporation 1978). However, this study is limited by small animal numbers and incomplete
evaluation of other end points at lower doses. Additionally, a well-designed 13-week study (Great Lakes
Chemical Corporation 2000) found hepatocellular hypertrophy at a higher minimum effect level
(16 mg/m^3) than the 14-day study, but no degenerative liver changes. The available information indicates
that there is insufficient evidence for considering the hepatic changes as adverse acute effects. More
importantly, exposure to ≥ 16 mg/m^3 caused changes in serum levels of thyroid hormones (decreased T$_3$,
increased TSH) in the 13-week study. Thyroid hormone levels were not determined in the 14-day study.
Therefore, due to the lack thyroid hormone data in the 14-day study, as well as the lack of any clear
lowest-observed-adverse-effect levels (LOAELs) for the other end points in the 14-day study, particularly
at exposures levels below the LOAEL for thyroid effects in the 13-week study, the data are inadequate to
derive an MRL for acute-duration exposure.

- An MRL of 0.006 mg/m^3 has been derived for intermediate-duration inhalation exposure (15–
  364 days) to lower-brominated BDEs.

The intermediate-duration inhalation MRL is based on a no-observed-adverse-effect level (NOAEL) of
1.1 mg/m^3 for changes in thyroid hormones in rats that were intermittently exposed to octaBDE for
13 weeks (Great Lakes Chemical Corporation 2000). Calculation of the MRL is detailed below.

The inhalation database for intermediate-duration exposure to PBDEs consists of one well-conducted
13-week study (Great Lakes Chemical Corporation 2000). This is an unpublished industry-sponsored
study in which a commercial octaBDE product (bromine content 78.7%) was administered to groups of

10 male and 10 female Crl:CD(SD)IGS BR rats, via nose-only inhalation as a dust aerosol, in measured concentrations of 0 (air only), 1.1, 16, or 202 mg/m^3 for 6 hours/day, 5 days/week, for 13 weeks. The mean MMADs in the low to high exposure groups were 2.0, 2.7, and 2.8 μm, and the corresponding mean GSDs were 3.37, 3.72, and 3.01. Clinical and physical signs, body weight, food consumption, and survival were evaluated throughout the study. Ophthalmic, hematology (11 indices), serum chemistry (18 indices), and serum thyroid hormone (TSH, total T$_3$, and total T$_4$) evaluations were performed near the end of the exposure period. Urine analyses were not conducted. Comprehensive necropsies, organ weight measurements, and histological examinations (including respiratory tract and thyroid) were performed following exposure termination.

Hepatic, nasal, lung, thyroid, and ovarian effects were observed (Great Lakes Chemical Corporation 2000). The liver was affected in both sexes as shown by dose-related increases in centrilobular hepatocellular hypertrophy at ≥16 mg/m^3 and liver weight (absolute and relative) at 202 mg/m^3. Total incidences of centrilobular hepatocellular hypertrophy in the 0, 1.1, 16, and 202 mg/m^3 groups were 1/10, 0/10, 3/10, and 10/10, respectively, in males and 0/10, 0/10, 3/10, and 6/10, respectively, in females; severity was predominantly minimal in affected animals from all groups. The incidence of nasal goblet cell lesions was increased at 202 mg/m^3, but showed no clear dose-related trends for increasing incidence or severity. Total incidences of nasal goblet cell hypertrophy were slightly increased in nasal level II of both sexes at ≥1.1 mg/m^3; respective incidences in the 0, 1.1, 16, and 202 mg/m^3 exposure groups were 4/10 (all minimal), 9/10 (7 minimal, 2 mild), 6/10 (all minimal), and 10/10 (9 minimal, 1 mild) in males, and 2/10 (all minimal), 6/10 (all minimal), 4/10 (all minimal), and 8/10 (all minimal) in females. Nasal goblet cell hypertrophy was also slightly increased in nasal level IV in males at 202 mg/m^3 (4/10, 0/10, 1/10, and 8/10, all minimal severity, not increased in females). Histological changes in the lungs included alveolar histiocytosis and chronic active inflammation that were only clearly increased in incidence at 202 mg/m^3. Total incidences of alveolar histiocytosis at 0, 1.1, 16, and 202 mg/m^3 were 3/10, 5/10, 5/10, and 10/10, respectively, in males, and 0/10, 5/10, 2/10, and 10/10, respectively, in females. Corresponding total incidences of chronic active lung inflammation were 0/10, 0/10, 2/10, and 10/10 in males, and 0/10, 1/10, 1/10, and 10/10 in females. The severity of both lesion types tended to increase from minimal at lower doses to mild/moderate at 202 mg/m^3. Gross lung changes also occurred in both sexes at 202 mg/m^3; these included lung firmness and white discoloration and/or enlargement in the bronchial and/or mediastinal lymph nodes. The lymph node effects correlated with the histological finding of granulomatous inflammation. There were no exposure-related gross or histopathological changes in the spleen, bone marrow, thymus, or other tissues, including thyroid. Thyroid hormone assessments, however, showed exposure-related decreases in mean thyroxine (total T$_4$) at ≥16 mg/m^3 in

both sexes, and increases in TSH at ≥16 mg/m^3 in males and 202 mg/m^3 in females. The changes were usually statistically significant (p<0.05 or p<0.01) compared to controls and were considered to be consistent with chemical-induced hypothyroidism. There were no serum T$_3$ changes. Qualitative histological evaluations of step sections of ovaries showed an absence of corpora lutea in 3/10 females exposed to 202 mg/m^3, compared to 0/10 in the control and lower exposure groups. This 30% incidence was interpreted to be a treatment-related effect because an absence of corpora lutea was considered unusual in rats at 20 weeks of age.

Considering the minimal severity of the nasal goblet cell hypertrophy, lack of clear dose-related increasing trends for incidences and severity of this nasal effect, clear identification of both a NOAEL (1.1 mg/m^3) and LOAEL (16 mg/m^3) for changes in serum levels of thyroid hormones, and abundant evidence for thyroid effects of PBDEs in oral studies, the effects on thyroid hormones are the most appropriate basis for estimation of an intermediate-duration inhalation MRL. The MRL of 0.006 mg/m^3 was derived by dividing the NOAEL$_{HEC}$ of 0.53 mg/m^3 by an uncertainty factor of 30 (3 for species to species extrapolation with dosimetric adjustments and 10 for human variability) and a modifying factor of 3 (for an incomplete database reflecting a single study in one species). The NOAEL$_{HEC}$ was calculated using the following equations:

$$\text{NOAEL}_{ADJ} = 1.1 \text{ mg/m}^3 \times 6 \text{ hours/24 hours} \times 5 \text{ days/7 days} = 0.196 \text{ mg/m}^3$$
$$\text{NOAEL}_{HEC} = \text{NOAEL}_{ADJ} \times \text{RDDR} = 0.196 \text{ mg/m}^3 \times 2.7 = 0.53 \text{ mg/m}^3$$

The regional deposited dose ratio (RDDR) for the extrathoracic region was used to extrapolate deposited doses in rats to deposited doses in humans. The following parameters were used to calculate the RDDR: MMAD of 2.0 μm with a mean GSD (sigma g) of 3.37, default human body weight of 70 kg, and a default female F344 rat body weight of 0.18 kg. Additional information on the derivation of the intermediate-duration inhalation MRL for lower-brominated BDEs is provided in Appendix A.

No MRL was derived for chronic-duration inhalation exposure to lower-brominated BDEs due to a lack of chronic studies.

Decabromodiphenyl Ether. No MRLs were derived for acute-, intermediate-, or chronic-duration inhalation exposure to decaBDE due to a lack of inhalation studies on this PBDE congener.

Oral MRLs

Lower-brominated Diphenyl Ethers

- An MRL of 0.00006 mg/kg/day has been derived for acute-duration oral exposure (14 days or less) to lower-brominated diphenyl ethers.

The acute oral MRL is based on a LOAEL of 0.06 mg/kg/day for endocrine effects in rat dams and reproductive and neurobehavioral effects in F1 offspring exposed to 2,2',4,4',5-pentaBDE (BDE 99) on GD 6 via gavage (Kuriyama et al. 2005, 2007; Talsness et al. 2005). The MRL was estimated by dividing the 0.06 mg/kg LOAEL by an uncertainty factor of 1,000 (10 for use of a LOAEL, 10 for animal to human extrapolation, and 10 for human variability).

In a series of reports, pregnant rats were exposed to BDE 99 at 0, 0.06, or 0.3 mg/kg via gavage on GD 6 (Kuriyama et al. 2005, 2007; Talsness et al. 2005). Serum thyroid hormones levels (T_3, free-T_3, T_4, free-T_4) were analyzed in dams and pups on PNDs 1, 14, and 22 (Kuriyama et al. 2007). Male and female offspring were evaluated for emergence of physical landmarks and reflexes and for open-field behavior on PNDs 36 and 71 (Kuriyama et al. 2005). Twelve male offspring per dose group were sacrificed at PND 140, and the thymus, spleen, liver, testis, epididymis, seminal vesicle, and ventral prostate were weighed. The right testis and caudal epididymis were retained for spermatid and sperm counts and morphology, respectively. Additionally, blood was collected for analysis of testosterone and LH levels (Kuriyama et al. 2005). Similarly, 10 F1 females per group were sacrificed ~PND 90 for histological evaluation of the ovary, uterus, and vagina. Ovarian follicles were counted in 10 ovaries from each group, and 1 ovary from 1 female offspring in each group was analyzed by transmission electron microscopy (Talsness et al. 2005). Fertility was assessed in F1 males and females (20/group) mated to unexposed partners. The uterine and F2 fetal weights and the number of implantations, resorptions, and fetuses were determined. The F2 fetuses were examined for external anomalies and when present, the fetuses were stained and examined for skeletal anomalies (Kuriyama et al. 2005; Talsness et al. 2005). In a separate group of F1 males, male sexual behavior was assessed in 20 males/group at ~PND 160 (Kuriyama et al. 2005).

Serum T4 levels were significantly decreased by 23–33% in the 0.06 and 0.3 mg/kg dams, sacrificed on PND 1. No changes were observed in T_3, free-T_3, or free-T_4 at PND 1 or any thyroid hormone levels at PND 22 in dams. In pups, no dose-related changes were observed at PND 1 or 14. At PND 22, serum T_4 was significantly decreased by 19–22% in F1 males and females and serum free-T_4 was significantly

decreased by 24% in F1 females exposed to 0.3 mg/kg (Kuriyama et al. 2007). For F1 development of physical landmarks and reflexes, no exposure-related effects were observed for the age at fur development or eye opening, testes descent, or the ability to master the rotating rod test. However, significant delays in the eruption of incisors in F1 pups and the development of the cliff-drop aversion reflex were observed in F1 males in the 0.3 mg/kg group, compared with controls. During a 24-hour observation of open-field activity, total activity, time spent active, duration of activity per active phase, and total activity per active phase were all significantly increased in F1 offspring on PND 36 in the 0.3 mg/kg group, compared with controls. On PND 71, the increased total activity and time spent active persisted in the 0.3 mg/kg group, and was also significantly increased in the 0.06 mg/kg group.

In F1 males sacrificed on ~PND 140, no exposure-related changes were observed in body weight, liver weight, or thymus weight; however, absolute spleen weight was significantly increased by 9% in the 0.06 and 0.3 mg/kg groups, and relative spleen weight was significantly increased by 12% in the 0.06 mg/kg group. Compared with controls, significantly altered male reproductive organ weights at PND 140 included a 10 and 11% decrease in relative testes and epididymis weight, respectively, in the 0.3 mg/kg group and a 5% decrease in relative epididymis weight in the 0.06 mg/kg group; no significant changes were observed in absolute organ weights. In both dose groups, the number of spermatids and sperm and daily sperm production were significantly decreased, compared with controls. No exposure-related effects were observed for sperm morphology. No changes were observed in serum testosterone or LH levels. Despite sperm alterations, no significant exposure-related effects were observed in male reproductive function or the majority of male sexual behaviors. The only significantly altered male sexual behavior was a 32% decrease in the percent of males with two or more ejaculations.

In F1 females sacrificed on ~PND 90, no statistically significant, exposure-related histological changes were observed at the light microscopic level in the ovary, uterus, or vagina of female offspring, and no exposure-related effects were observed in the number of ovarian follicles. However, multiple ultrastructural changes were noted in the ovaries of PND 90 female offspring from dams exposed to 0.06 or 0.3 mg/kg, including destruction of the surface of the serosal epithelial cells, necrosis, and numerous vesicular structures with dense granular material within the cytoplasm. Additional changes observed in the 0.3 mg/kg group included degenerative changes and aggregates of small and large vesicles filled with homogeneously dense granular material in the cytoplasm and clumped chromatin within the condensed nucleus. No exposure-related changes were found for F1 female pregnancy rate, total implantation sites, implantation sites/dam, F2 fetuses/gravid dam, or total number of live F2 fetuses. However, the resorption rates were 12 and 15% in the 0.06 and 0.03 mg/kg groups, respectively,

compared with the control rate of 9%. Statistics were not reported; however, the resorption rates in the
exposed rats were also reportedly increased compared with historical controls (average control resorption
rate=5.4%, with rates up to 10% considered to be within normal limits). In addition, the percentage of
litters with resorptions was higher in the exposed females, being 47% in the control group and 69% and
72% in the 0.06 and 0.3 mg/kg groups, respectively. In F2 pups, mean fetal weight was significantly
increased (by 5%) in the 0.06 mg/kg group, but not in the 0.3 mg/kg group, compared with controls.
Three fetuses from different litters in the 0.3 mg/kg/day group showed skeletal anomalies (tail, skull,
vertebrae); however, this incidence of anomalies in 3/18 litters is not significantly elevated compared with
the control incidence of 0/19 (Fisher's exact test, performed for this review).

Collectively, these studies indicate a LOAEL of 0.06 mg/kg, the lowest dose tested, for endocrine effects
in F0 dams (decreased serum T_4), reproductive effects in F1 adult offspring (impaired spermatogenesis,
ultrastructural changes in ovaries, increased resorptions in F1 females mated to unexposed males) and
neurobehavioral effects in F1 adult offspring (increased activity in open field). No NOAEL was
identified.

Data from other several acute-duration studies of PBDEs support the selection of the co-critical effects
observed at the LOAEL of 0.06 mg/kg:

- Numerous studies reported reduced serum T_4 levels in adult, nonpregnant mice and rats following
  acute exposure to commercial pentaBDE mixtures (Bromkal 70, Bromkal 70-5 DE, DE-71), the
  commercial octaBDE mixture DE-79, or 2,2',4,4'-tetraBDE (BDE 47). Significant reductions of
  19–92% have been reported following gavage exposure at doses ≥10 and ≥0.8 mg/kg/day in rats
  and mice, respectively, for 1–14 days (Darnerud and Sinjari 1996; Fowles et al. 1994; Hallgren
  and Darnerud 1998, 2002; Hallgren et al. 2001; Hoppe and Carey 2007; Richardson et al. 2008;
  Stoker et al. 2004, 2005; Zhou et al. 2001).

- In a companion study to the critical studies described above, pregnant rats (8/group) were
  administered BDE 47 (98% purity) at 0, 0.14, or 0.7 mg/kg via gavage in peanut oil vehicle on
  GD 6 (Talsness et al. 2008). As observed in pentaBDE-exposed F1 females, ultrastructural
  changes (accumulation of vesicular structures with homogeneously dense granular material in the
  cytoplasm of the stromal cells, large vacuoles) were observed in the ovaries of F1 females from
  both dose groups on PND 100. No exposure-related changes were observed in F1 female fertility
  or F2 litter parameters. F1 males were not evaluated for developmental reproductive effects
  following tetraBDE exposure.

- Alterations in open-field activity have been consistently reported in mice exposed to pentaBDE
  (BDE 99) at doses ≥0.8 mg/kg on PND 3 or 10 and evaluated at 2–8 months of age, characterized
  by decreased activity during the first 20-minute period of a 1-hour session, followed by increased
  activity during the third 20-minute period (Eriksson et al. 2002b, 2006; Fischer et al. 2008; Sand
  et al. 2004; Viberg et al. 2002, 2004a, 2004b). Several other 1-day exposure studies reported

similar findings in rats and mice following exposure to various lower-brominated PBDEs. Decreased spontaneous activity and/or impaired habituation were observed in rats exposed to BDE 99 at 8 mg/kg on PND 10, mice exposed to 2',4,4',5,5'-hexaBDE (BDE 153) at ≥0.45 mg/kg on PND 10, mice exposed to BDE 47 at 10.5 mg/kg on PND 10, mice exposed to 2,2',3,4,4',5',6-heptaBDE (BDE 183) at 15.2 mg/kg on PND 3, and mice exposed 2,2',3,4,4',5,5',6-octaBDE (BDE 203) at 16.8 mg/kg on PND 3 or 10 (Eriksson et al. 2001; Viberg et al. 2003a, 2005, 2006). Increased vertical activity was significantly increased at 4 months, but not at 2 months, in mice exposed to BDE 47 at ≥1 mg/kg on PND 10; no changes were observed in horizontal activity or habituation (Gee and Moser 2008).

Additional information on the derivation of the acute-duration oral MRL for lower-brominated BDEs is provided in Appendix A.

- An MRL of 0.000003 mg/kg/day has been derived for intermediate-duration oral exposure (15–364 days) to lower-brominated BDEs.

The intermediate oral MRL is based on a minimal LOAEL of 0.001 mg/kg/day for a 34% reduction in serum testosterone in male rats exposed to 2,2',4,4'-tetraBDE (BDE 47) for 8 weeks via gavage (Zhang et al. 2013b). The MRL was estimated by dividing the 0.001 mg/kg/day minimal LOAEL by an uncertainty factor of 300 (3 for use of a minimal LOAEL, 10 for animal to human extrapolation, and 10 for human variability).

Groups of 20 male rats were exposed to BDE 47 (≥98.7%) at 0, 0.001, 0.03, or 1 mg/kg/day via gavage in corn oil 6 days/week for 8 weeks (Zhang et al. 2013b). Twenty-four hours after the final treatment, rats were sacrificed. Testes were fixed for histological analysis and labeling of apoptotic cells or prepared for analysis of sperm production. Daily sperm production was estimated by dividing the total number of mature spermatids per testis by 6.1 (i.e., the days of the seminiferous cycle that the spermatids are present in the seminiferous epithelium). Testicular samples were examined for reactive oxygen species (ROS) and mRNA expression of apoptosis related proteins (ser15, ser473, p53, PTEN, AKT, BAD, caspase 3, FAS, FASL). Serum levels of E2, FSH, LH, and testosterone were measured. Histological examination of the testes showed a significant increase in the number of multinucleated giant cells (arising from spermatocytes that aborted meiosis) at ≥0.03 mg/kg/day and abundant vacuolar spaces in the seminiferous epithelium at 1 mg/kg/day (quantitative data not reported). Additionally, the number of apoptotic cells was significantly increased by 1.9- and 3-fold in the testes of rats from the 0.03 and 1 mg/kg/day groups, respectively, and the mRNA levels of several apoptosis genes were elevated in a dose-related manner. Daily sperm production was significantly decreased by 23% in the 1 mg/kg/day group, compared with controls. Serum testosterone was significantly decreased by ~34, 53, and 62% in the 0.001, 0.03, and 1 mg/kg/day groups, respectively, compared with controls. No exposure-related changes were observed

in serum E2, FSH, or LH levels. Testicular ROS levels were significantly elevated at 1 mg/kg/day, compared with controls. A minimal LOAEL of 0.001 mg/kg/day was determined for this study based on the 34% decrease in serum testosterone. The change in testosterone is considered a minimal LOAEL because it is unclear if the magnitude of change represents a biologically adverse effect; however, this statistically significant reduction in serum testosterone is considered an early indication of damage to the male reproductive system, considering the additional effects observed at ≥0.03 mg/kg/day (histological lesions in testes, sperm effects).

One additional rat study and a mouse study report histopathological changes in the testes following intermediate-duration exposure to tetraBDE at ≥0.03 mg/kg/day; neither study evaluated serum testosterone levels (Huang et al. 2015; Wang et al. 2013). In the rat study, a NOAEL of 0.001 mg/kg/day and a LOAEL of 0.03 mg/kg/day were identified for increased epithelial thickness and spermatocyte apoptosis in the testes of males exposed to BDE 47 for 8 weeks via gavage (Huang et al. 2015). In the mouse study, a NOAEL of 0.0015 mg/kg/day and a LOAEL of 0.045 mg/kg/day were identified for germ cell loss and increased apoptosis in the testes of males exposed to BDE 47 for 30 days via gavage (Wang et al. 2013).

No other study evaluated testicular histopathology following exposure to BDE 47. Following intermediate exposure to other congeners, no changes in testicular histology were observed in rats exposed to commercial pentaBDE mixtures (Bromkal 70-5 DE, DE-71) at gavage doses up to 250 mg/kg/day for 15–28 days (Becker et al. 2012; Oberg et al. 2010), commercial penta- or octaBDE mixtures (DE-71, unspecified octa mixture) at dietary doses up to 750 mg/kg/day for 28–90 days (IRDC 1976, 1977; WIL Research Laboratories 1984), or a pentaBDE mixture (52.1% pentBDE, 44.2% decaBDE, 0.4% octaBDE) at dietary doses up to 20 mg/kg/day for 70 days (Ernest et al. 2012). However, testicular apoptosis was not evaluated in any of these studies.

No other study evaluated serum testosterone levels following exposure to BDE 47. However, as observed with exposure to BDE 47, acute exposure to 0.06 or 1.2 mg/kg of 2,2',4,4',5-pentaBDE (BDE 99) also led to a significant 40–45% decrease in serum testosterone levels in rats (Alonso et al. 2010). No other studies evaluated this end point following exposure to single congeners. Other studies evaluating serum testosterone levels after intermediate-duration exposure to lower-brominated PBDEs mixtures (DE-71, dietary PBDE mixture described above) did not report exposure-related decreases (Becker et al. 2012; Ernest et al. 2012; Stoker et al. 2005). These data suggest that the individual congeners, BDE 47 and BDE 99, which have been identified as two of the most abundant congeners for human exposure (Harrad

et al. 2004; Lorber 2008; Wong et al. 2013), may have a greater capacity to alter serum testosterone levels
than PBDE mixtures.

One-generation studies of BDE 47 reported developmental effects at ≥0.03 mg/kg/day, including:

- Impaired spatial learning in the Barnes maze in PNW 8 offspring of mouse dams fed tetraBDE-
  dosed cornflakes from pre-mating day 28 through PND 21 (Koenig et al. 2012).

- Decreased center-field activity in an open field (indicating increased anxiety) in PND 60 female
  offspring from mouse dams fed tetraBDE-dosed cornflakes from pre-mating day 28 through
  PND 21 (Ta et al. 2011).

- Decreased pre-weaning weight, decreased pup vocalizations on PNDs 8–10, and decreased
  sociability on PND 72 in female offspring of mouse dams exposed to tetraBDE via gavage from
  pre-mating day 28 through PND 21 (Woods et al. 2012).

Additional information on the derivation of the intermediate-duration oral MRL for PBDEs is provided in
Appendix A.

A chronic-duration oral MRL was not derived for lower-brominated PBDEs due to insufficient data.
Only one chronic study of PBDEs other than high-purity decaBDE has been conducted (Kociba et al.
1975; Norris et al. 1975a). In this study, Sprague-Dawley rats (25/sex/dose level) were fed a 77.4% pure
commercial decaBDE mixture (containing 21.8% nonaBDE and 0.8% octaBDE) for approximately
2 years. Evaluations that included clinical signs, body weight, food consumption, hematology, clinical
chemistry, urine indices, and comprehensive histological examinations showed no exposure-related
effects. The highest NOAEL is 1 mg/kg/day (highest tested dose), but this NOAEL is not appropriate for
MRL estimation due to insufficient sensitivity of the study. In particular, using the NOAEL of
1 mg/kg/day and an uncertainty factor of 100, a chronic oral MRL based on this study would be 5 times
higher than the 0.002 mg/kg/day intermediate MRL. A similar pattern was observed for thyroid effects in
the study used to derive the acute-duration oral MRL (Zhou et al. 2001) as summarized above. Due to the
insufficiencies of the chronic data for MRL derivation, the intermediate oral MRL could be used as a
value for chronic exposure.

Decabromodiphenyl Ether

- An MRL of 0.01 mg/kg/day has been derived for acute-duration oral exposure (14 days or less) to
  decabromodiphenyl ether

2. RELEVANCE TO PUBLIC HEALTH

The MRL was derived based on a NOAEL of 1.34 mg/kg for neurobehavioral effects in 2–4-month-old mice following a single exposure to 2,2',3,3',4,4',5,5',6,6'-decaBDE (BDE 209) on PND 3 (Buratovic et al. 2014; Johansson et al. 2008). The MRL was estimated by dividing the 1.34 mg/kg NOAEL by an uncertainty factor of 100 (10 for animal to human extrapolation and 10 for human variability).

In the first study (Johansson et al. 2008), neonatal male mice (3–4 litters/group) were exposed to a single dose of BDE 209 (98% purity) at 0, 1.34, 2.22, 13.4, or 20.1 mg/kg via gavage in a 20% fat emulsion vehicle (1:10 mixture egg lecithin and peanut oil) on PND 3 (Johansson et al. 2008). Mice were observed for clinical signs of toxicity and body weight was measured at PND 3 and PNW 4. Spontaneous motor behavior (locomotion, rearing, total activity) was evaluated in an open field test at 2 months (10 mice/group) and at 4 months (16 mice/group). Motor activity was measured during a 60-minute period, divided into three 20-minute intervals. Nicotine-induced behavior was evaluated at 4 months following single subcutaneous injections of 80 µg nicotine/kg (8/group) or 10 mL 0.9% NaCl/kg (8/group). Anxiety was assessed at 4 months using the elevated plus maze. No clinical signs of toxicity or body weight effects were observed. At 2 months, significantly decreased locomotion, rearing, and total activity were observed during the first 20-minute interval of the open field assessment in mice exposed to ≥2.22 mg/kg, compared with controls. However, during the third 20-minute interval, when activity should decrease due to habituation, locomotion, rearing, and total activity were significantly increased in mice exposed to ≥13.4 mg/kg. None of the end points measured were significantly altered in mice exposed to 1.34 mg/kg. At 4 months, significantly decreased locomotion, rearing, and total activity were observed during the first interval of the open field assessment in mice exposed to ≥2.22 mg/kg, compared with controls. During the third interval, significantly increased locomotion, rearing, and total activity were observed in mice exposed to ≥2.22 mg/kg. Additionally, total activity, but not rearing or locomotion, was significantly decreased during the first 20-minute interval in the 1.34 mg/kg group; no significant changes were observed during the third interval in the 1.34 mg/kg group. Statistical analysis shows that habituation ability declined in mice exposed to ≥2.22 mg/kg/day when tested at 4 months of age, compared with 2 months of age. At 4 months, nicotine exposure caused significantly decreased activity during the first interval in mice exposed to ≥13.4 mg/kg, compared with saline-injected mice from the same decaBDE exposure group. This finding is the opposite of the expected increase in activity due to nicotine exposure, which was observed in controls and lower dose decaBDE groups. During the third interval, mice exposed to ≥13.4 mg/kg and nicotine showed impaired habituation. No exposure-related effects were observed in the elevated plus maze assessment. A NOAEL of 1.34 mg/kg and a LOAEL of 2.22 mg/kg were determined for the nonhabituating profile (i.e., decreased activity early in the test period and increased activity late in the test period). The singular finding of decreased total activity

during the first 20-minute interval at 4 months in the 1.34 mg/kg group was not considered sufficient to establish a LOAEL of 1.34 mg/kg. The nonhabituating profile, which is a common effect observed with developmental PBDE exposure (Eriksson et al. 2002b, 2006; Fischer et al. 2008; Sand et al. 2004; Viberg et al. 2002, 2003a, 2004a, 2004b), was considered to be a stronger basis for a NOAEL/LOAEL determination.

In the second study (Buratovic et al. 2014), neonatal male mice (6 litters/group; 31–40 males and 23–34 females per group) were administered 2,2',3,3',4,4',5,5',6,6'-decaBDE (BDE 209, >95% purity) at doses of 0, 1.34, 5.76, or 13.4 mg/kg via gavage in a 20% fat emulsion vehicle (1:10 mixture egg lecithin and peanut oil) on PND 3. Mice were observed for clinical signs of toxicity and body weight changes throughout the study (no further details were provided). Spontaneous motor behavior (locomotion, rearing, total activity) was evaluated in an open field at 2 months (18/sex/group). Motor activity was measured during a 60-minute period, divided into three 20-minute intervals. Directly after spontaneous motor evaluation, 9/sex/group were injected with a cholinergic agent (0.25 mg/kg paraoxon in males, 80 µg/kg nicotine in females), while the other 9/sex/group were injected with 0.9% saline, for evaluation of cholinergic-induced locomotion. At 4 months, spontaneous behavior was assessed again in the saline-injected animals only (9 males/group at all doses and 9 females/group in the control and high-dose group only). Learning and memory was assessed using the Morris water maze at 5 and 7 months in 13–15 males from the 0, 5.76, and 13.4 mg/kg groups only (the same mice were evaluated at each time point). In the spontaneous activity assessment, a dose-related decrease in locomotion, rearing, and total activity was observed during the first 20 minutes of open field testing in a novel environment at 2 months. Decreases were significant at all doses tested in both sexes; however, findings were only dose-related for total activity. However, during the third 20-minute interval, when activity should decrease due to habituation, locomotion, rearing, and total activity were significantly increased in males and females at ≥5.76 mg/kg. At 2 months, cholinergic agents caused decreased activity during the first interval in mice exposed to ≥5.76 mg/kg, compared with saline-injected mice from the same decaBDE exposure group. This finding is the opposite of the expected increase in activity due to paraoxon or nicotine exposure, which was observed in controls and low-dose decaBDE groups. During the third interval, mice exposed to ≥5.76 mg/kg and cholinergic agent showed impaired habituation. At 4 months, total activity during the first 20 minutes was still significantly decreased at all doses in males, and locomotion and rearing were significantly decreased in males in the mid- and high-dose groups only; all three parameters were significantly decreased in high-dose females (other doses not evaluated). All three parameters were significantly increased in high-dose males and females during the third 20-minute period, indicating decreased habituation; locomotion and rearing were also slightly, but significantly, increased in mid-dose

males. In the Morris water maze, initial learning was comparable between exposed and control mice at
5 and 7 months. However, latencies to find the escape platform during the reversal learning phase
(learning to find the escape platform in a new location after initial training) were significantly longer in
mid- and high-dose males at 5 and 7 months (other exposure groups not assessed). A NOAEL of
1.34 mg/kg and a LOAEL of 5.76 mg/kg were determined for the nonhabituating profile (i.e., decreased
activity early in the test period and increased activity late in the test period). Similar to the Johansson et
al. (2008) study, the finding of decreased total activity during the first 20-minute interval at 2 and
4 months in the 1.34 mg/kg group was not considered sufficient to establish a LOAEL of 1.34 mg/kg.
The nonhabituating profile was considered to be a stronger basis for a NOAEL/LOAEL determination,
and additional neurological effects (impaired learning, altered response to cholinergic agents) support a
LOAEL of 5.76 mg/kg.

A similar study supports the LOAEL of 2.22 mg/kg for altered neurobehavior in developmentally
exposed mice. Decreased spontaneous activity and impaired habituation were observed in 2- and
6-month-old mice exposed to BDE 209 at doses $\geq$2.22 mg/kg on PND 3, which was the lowest dose tested
(Viberg et al. 2003b). These effects were not observed if exposure was on PND 10 or 19 at doses up to
20.1 mg/kg (Viberg et al. 2003b). Additionally, decreased spontaneous activity was observed in
2-month-old rats following exposure to BDE 209 doses $\geq$6.7 mg/kg on PND 3 (lowest dose tested)
(Viberg et al. 2007). At 20.1 mg/kg, impaired habituation and decreased nicotine-induced behavior were
also observed. This nonhabituating behavior profile (i.e., decreased activity early in the test period and
increased activity late in the test period) is consistent with neurobehavioral alterations observed following
early postnatal exposure to lower-brominated PBDEs and has been reported in adult mice neonatally
exposed to certain PCB congeners (see the Acute MRL Worksheet for lower-brominated PBDEs for more
details).

Additional neurodevelopmental effects observed in mice following acute exposure to BDE 209 from
PND 2 to 15 at 20 mg/kg/day via micropipette include delayed ontogeny of reflexes, increased
locomotion in males at PND 70, and learning impairment and impulsivity at 16 months, but not at
3 months (Rice et al. 2007, 2009). In rats, impaired learning was observed in Morris water maze in
PND 25 rat offspring of dams exposed to BDE 209 from GD 1 to 14 at doses $\geq$30 mg/kg/day via gavage
(Chen et al. 2014).

Additional information on the derivation of the acute-duration oral MRL for BDE 209 is provided in
Appendix A.

- An MRL of 0.0002 mg/kg/day has been derived for intermediate-duration oral exposure (15–
  364 days) to decabromodiphenyl ether.

The MRL was derived based on a minimal LOAEL of 0.05 mg/kg/day for a 12% increase in serum

glucose in adult rats exposed to 2,2′,3,3′,4,4′,5,5′,6,6′-decabromodiphenyl ether (BDE 209) for 8 weeks

via gavage (Zhang et al. 2013a). The MRL was estimated by dividing the 0.05 mg/kg/day LOAEL by an

uncertainty factor of 300 (3 for use of a minimal LOAEL, 10 for animal to human extrapolation, and 10

for human variability).

Groups of 10 male rats were exposed to BDE 209 at 0, 0.05, 1, or 20 mg/kg/day daily via gavage in corn

oil for 8 weeks (Zhang et al. 2013a). Rats were observed for clinical signs of toxicity and body weights

were measured every 3 days. Rats were fasted for 24 hours after the final gavage treatment, and then

sacrificed. Body weights and heart, spleen, lung, kidney, and liver weights were recorded. Blood was

collected for clinical chemistry analysis (serum total cholesterol, triglycerides, glucose, insulin, and

TNF-α) and determination of plasma markers of oxidative stress (malondialdehyde [MDE], reduced

glutathione [GSH], and superoxidase dismutatase [SOD]). Liver samples from three rats in the control

and low-dose (0.05 mg/kg/day) groups were collected for microarray analysis (Affymetrix GeneChip),

and gene ontogeny category, pathway, gene-act-network, and gene co-expression analyses were

conducted. Quantitative real-time-polymerase chain reaction (qPCR) was performed to quantitate gene

expression to validate the gene expression data obtained from microarray analysis. No clinical signs of

toxicity or body weight effects were observed. The relative liver weight was significantly decreased at

1 and 20 mg/kg/day by 9% (absolute liver weights were not reported). No changes were observed in

relative weights of heart, spleen, lung, or kidney. No exposure-related changes were reported in serum

cholesterol or triglyceride levels. Serum glucose levels were significantly increased by 12, 18, and 21%

in the 0.05, 1, and 20 mg/kg/day groups, compared with controls. Serum insulin was significantly

decreased by 50–60% at 1 and 20 mg/kg/day. Subsequent to this finding, the pancreas was evaluated

histologically. Consistent with the insulin findings, morphological changes were seen at 1 and

20 mg/kg/day, including blurred boundaries among pancreatic islet cells (quantitative data not reported).

Plasma SOD activity was significantly decreased in all exposed groups and plasma GSH was significantly

decreased at 1 and 20 mg/kg/day. Serum TNF-α was significantly increased at 1 and 20 mg/kg/day.

Additionally, decaBDE induced 1,257 liver gene transcript changes, and 18 canonical pathways were

significantly enriched. Four of them were involved in immune diseases, including autoimmune thyroid

disease, graft-versus-host disease, allograft rejection, and T1DM. Subsequently, gene act network and

gene coexpression network found that some major histocompatibility complex molecules and TNF-α

were involved in the T1DM pathway. A minimal LOAEL of 0.05 mg/kg/day was determined for this study based on the 12% increase in serum glucose levels. The change in glucose is considered a minimal LOAEL because it is unclear if the magnitude of change represents a biologically adverse effect; however, the increase in serum glucose is considered to be part of a spectrum of effects indicative of altered insulin homeostasis and toxicity to the pancreas, including decreased serum insulin and morphological changes in pancreatic islet cells observed at ≥1 mg/kg/day following decaBDE exposure.

The relevance of these findings to human health is uncertain. An analysis of cross-sectional NHANES data showed a significant increase in risk of diabetes associated with serum concentrations of BDE 153 (but not other congeners), although the risk was higher with exposure to 50–75th percentile BDE 153 concentrations than >75th percentile BDE 153 concentrations (Lim et al. 2008). Serum BDE 153 concentrations (but not BDE 28, BDE 47, BDE 85, BDE 99, BDE 100, or BDE 154) were also shown to be significantly associated with increased odds of developing gestational diabetes in a cohort of 258 pregnant women (Smarr et al. 2016). However, other cross-sectional and prospective studies found no relationship between serum PBDE concentrations and diabetes in an adult cohort from Wisconsin (Turyk et al. 2015), an elderly cohort in Finland (Airaksinen et al. 2011), or an elderly cohort in Sweden (Lee et al. 2011).

Only one other animal study evaluated the pancreas following decaBDE exposure. In rats exposed to BDE 209 via gavage for 28 days at doses of 0, 0.27, 0.82, 2.47, 7.4, 22.2, 66.7, or 200 mg/kg/day, slight or moderate insulitis was observed in the Langerhan's islets of the "majority of samples," but findings were not exposure-related (Van der ven et al. 2008a). Similarly, no exposure-related effects were observed for serum glucose levels (Van der ven et al. 2008a). The only other study evaluating serum glucose levels after decaBDE exposure instead reported reduced serum glucose levels in male rats exposed to 20 mg/kg/day of a dietary PBDE mixture containing 52.1% pentaBDE (DE-71), 44.2% decaBDE (BDE 209), and 0.4% octaBDE (DE-79) for 70 days (Ernest et al. 2012). The observed decreased glucose levels could be due to the pentaBDE component, as male rats exposed to pentaBDE at doses of 0.27–200 mg/kg/day for 28 days also showed decreased glucose levels; the study authors did not report the lowest dose at which glucose levels were significantly lower in male rats, but they reported a BMD_{10RD} of 179.55 mg/kg/day and a $BMDL_{10RD}$ of 66.7 mg/kg/day (Van der ven et al. 2008b). Other effects occurred at doses 4–40-fold higher than the observed pancreatic and related effects:

- A LOAEL of 2 mg/kg/day was identified for transient histopathological effects in the liver of male offspring and kidney of female offspring of rat dams exposed to BDE 209 from GD 10 to PND 21 (no NOAEL identified) (Fujimoto et al. 2011).

- A LOAEL of 10 mg/kg/day was identified for hepatocytic swelling in the liver, vacuolization in the interstitial cells of testes, and sperm damage in PND 71 male offspring of mouse dams exposed to BDE 209 from GD 0 to 17 (no NOAEL identified) (Tseng et al. 2008, 2013).

- A LOAEL of 20 mg/kg-day was identified for decreased anxiety in mice treated with BDE 209 by daily gavage for 15 days (no NOAEL identified) (Heredia et al. 2012).

- A LOAEL of 20.1 mg/kg/day was identified for altered hippocampal electrophysiology in rats exposed to BDE 209 on GD 1 to PND 41, PNDs 1–21, or PNDs 22–41 (no NOAEL identified) (Xing et al. 2009).

Additional information on the derivation of the intermediate-duration oral MRL for decaBDE is provided in Appendix A.

No MRL was derived for chronic-duration oral exposure to decaBDE. Only one chronic study of high-purity decaBDE has been conducted. In this study, F344 rats and B6C3F1 mice (50/sex/group per species) were administered a commercial decaBDE product (94–97% pure) in the diet for 103 weeks (NTP 1986). Calculated dietary doses based on body weight and food intake were 0, 1,120, or 2,240 mg/kg/day for male rats; 0, 1,200, or 2,550 mg/kg/day for female rats; 0, 3,200, or 6,650 mg/kg/day for male mice; and 0, 3,760, or 7,780 mg/kg/day for female mice. Animals were examined daily for clinical signs. Body weights and food consumption were measured throughout the study, and comprehensive gross and histological examinations were performed on all animals in all dose groups, including those that were moribund or died during the study. No hematology, clinical chemistry, or urine indices or thyroid hormone levels were evaluated. Liver degeneration and thrombosis were significantly ($p<0.05$) increased in male rats at 2,240 mg/kg/day; respective incidences in the control, low, and high dose groups were 13/50, 19/50, and 22/50 for degeneration and 1/50, 0/50, and 9/50 for thrombosis. The thrombosis was characterized by a near total occlusion of a major hepatic blood vessel by a dense fibrin coagulum. Neoplastic nodules in the liver were significantly increased in a dose-related manner in males exposed to doses ≥1,120 mg/kg/day and in females exposed to 2,550 mg/kg/day. However, no treatment-related increases were observed in the incidence of hepatocellular carcinomas. Other effects in exposed rats included fibrosis of the spleen, lymphoid hyperplasia of the mandibular lymph nodes, and acanthosis of the forestomach at 2,240 mg/kg/day. In mice, histopathological changes occurred in males exposed to 3,200 mg/kg/day, including centrilobular hypertrophy and granulomas in the liver and follicular cell hyperplasia in the thyroid. An MRL was not derived because the lowest tested dose, 1,120 mg/kg/day in male rats, is a LOAEL for a liver lesion (neoplastic nodules) that is precancerous and associated with thrombosis in the same tissue.