2,2',4,4',5-Pentabromodiphenyl ether (BDE-99); CASRN 60348-60-9

Human health assessment information on a chemical substance is included in the IRIS database only after a comprehensive review of toxicity data, as outlined in the IRIS assessment development process. Sections I (Health Hazard Assessments for Noncarcinogenic Effects) and II (Carcinogenicity Assessment for Lifetime Exposure) present the conclusions that were reached during the assessment development process. Supporting information and explanations of the methods used to derive the values given in IRIS are provided in the guidance documents located on the IRIS website.

STATUS OF DATA FOR 2,2',4,4',5-PENTABROMODIPHENYL ETHER (BDE-99)

File First On-Line 06/30/2008

Category (section)	Assessment Available?	Last Revised
Oral RfD (I.A.)	yes	06/30/2008
Inhalation RfC (I.B.)	qualitative discussion	06/30/2008
Carcinogenicity Assessment (II.)	yes	06/30/2008

I. HEALTH HAZARD ASSESSMENTS FOR NONCARCINOGENIC EFFECTS

An IRIS assessment of commercial pentaBDE (CASRN 32534-81-9) was previously entered on IRIS on 08/01/1990. The commercial pentaBDE consisted mainly of pentaBDE (58%), tetraBDE (25%), and hexaBDE (13%) (Carlson, 1980a). An RfD of 0.002 mg/kg-day was derived, based on a no-observed-adverse-effect level (NOAEL) of 1.8 mg/kg-day for induction of hepatic enzymes in a 90-day oral study in rats (Carlson, 1980b) and using an uncertainty factor (UF) of 1,000 (10 each for interspecies and intraspecies variability and 10 for extrapolating from subchronic to chronic duration). Insufficient information was available to derive an RfC or to assess the carcinogenicity of this commercial grade pentaBDE.

I.A. REFERENCE DOSE (RfD) FOR ORAL EXPOSURE

Substance Name — 2,2',4,4',5-Pentabromodiphenyl ether (BDE-99)
CASRN — 60348-60-9
Section I.A. Last Revised — 06/30/2008

The RfD is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily oral exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. The RfD is intended for use in risk assessments for health effects known or assumed to be produced through a nonlinear (presumed threshold) mode of action. It is expressed in units of mg/kg-day. Please refer to the guidance documents at http://www.epa.gov/iris/backgrd.html for an elaboration of these concepts. Because RfDs can be derived for the noncarcinogenic health effects of substances that are also carcinogens, it is essential to refer to other sources of information concerning the carcinogenicity of this chemical substance. If the U.S. EPA has evaluated this substance for potential human carcinogenicity, a summary of that evaluation will be contained in Section II of this file.

I.A.1. ORAL RfD SUMMARY

Critical Effect	Point of Departure*	UF	RfD
Neurobehavioral effects	$BMDL_{1SD}$: 0.29 mg/kg	3,000	0.0001 mg/kg-day
Single dose gavage study in mice	BMD_{1SD}: 0.41 mg/kg		
Viberg et al. (2004a)			

* Conversion Factors and Assumptions -- $BMDL_{1SD}$ = 95% lower confidence limit on the maximum likelihood estimate of the dose corresponding to a change in the mean equal to one standard deviation (SD) of the control mean. BMD_{1SD} = maximum likelihood estimate of the dose corresponding to a change in the mean equal to one SD of the control mean.

I.A.2. PRINCIPAL AND SUPPORTING STUDIES

A study was carried out to determine whether exposure to BDE-99 during a period of rapid brain growth in neonatal mice could lead to disruption of the adult brain function (Viberg et al., 2004a). Single oral doses of 0, 0.4, 0.8, 4.0, 8.0, or 16 mg/kg BDE-99 (>99% purity) in a fat emulsion were given by gavage to male and female C57/B1 mice on postnatal day (PND)

10. Spontaneous motor behavior was tested at ages 2, 5, and 8 months in eight male and eight female mice randomly selected from three to five different litters in each treatment group at each testing occasion. Spontaneous motor behavior tests measured locomotion, rearing, and total activity for a 60-minute period, divided into three 20-minute periods, at each dose. In order to study time-dependent changes in habituation (2-month-old versus 8-month-old mice), an habituation ratio was calculated between the performance periods 40-60 minutes and 0-20 minutes for two of the spontaneous motor behavior variables, locomotion and rearing. There were no clinical signs of toxicity or effects on body-weight gain or body weight at any of the dose groups. Control mice showed habituation (i.e., a decrease in locomotion, rearing, and total activity in response to the diminishing novelty of the test chamber) over the three 20-minute test periods. There were significant dose-related changes in spontaneous motor behavior (locomotion, rearing, and total activity) at 0.8 mg/kg and above in male and female mice at ages 2, 5, and 8 months. These disturbances were also worse with increasing age. Male and female mice receiving doses of 0.8 mg/kg and higher showed significantly decreased activity (hypoactive) during the first 20 minute period and significantly increased activity (hyperactive) during the last 20-minute period, compared with control animals. Male and female mice exposed to the lowest dose of BDE-99 (0.4 mg/kg) did not significantly differ in activity in any of the three behavioral variables during any of the three 20-minute periods. The habituation capability for the locomotion and rearing variables was signicantly decreased in the 2- and 8- month-old male and female mice at 0.8 mg/kg and above, as evidenced by dose-related increases in the habituation ratio. The decline in habituation capability (i.e., the increase in the habituation ratio) was more pronounced in the 8-month-old mice than in the 2-month-old mice.

No major gender differences in spontaneous motor behavior responses or habituation capability were seen in this study. The NOAEL was 0.4 mg/kg and the lowest-observed-adverse-effect level (LOAEL) was 0.8 mg/kg for significant changes in spontaneous motor behavior and decreases in the rearing and locomotion habituation capability in both male and female mice, worsening with increasing age.

A study conducted by Viberg et al. (2004b) used the same protocol as Viberg et al. (2004a). Single oral doses of 0, 0.2, 0.4, or 12 mg/kg of BDE-99 were given by gavage to male NMRI mice on PND 10, and spontaneous motor behavior (locomotion, rearing, and total activity) was measured at the age of 4 months. The NOAEL in this study was 0.4 mg/kg and the LOAEL was 12 mg/kg for decreased and increased spontaneous motor behavior variables during the first and third 20-minute period, respectively.

The study of Eriksson et al. (2001) of the same research group as Viberg et al. (2004a, b) was also conducted in NMRI male mice given single doses of 0, 0.8, or 12 mg/kg BDE-99 on PND 10. In addition to the spontaneous motor behavior tests (locomotion, rearing, and total

activity), habituation capability (the ratio between performance in spontaneous motor behavior periods 40-60 minutes and 0-20 minutes for the three different variables) was used to analyze alteration in habituation to a novel environment in 2 and 4-month-old mice. Swim-maze performance, a measure of learning and memory ability, was tested in 5-month-old mice given the high dose of BDE-99 (12 mg/kg). Mice receiving 0.8 and 12 mg/kg of BDE-99 displayed significantly less activity during the first 20-minute period and were significantly more active during the third 20-minute period. The decrease in habituation capability was more pronounced in 4-month-old mice than in 2-month-old mice, indicating worsening with increasing age. Performance of 5-month-old mice in the swim maze learning/memory test was significantly worse in mice exposed to 12 mg/kg BDE-99 than in control mice. The LOAEL in this study was 0.8 mg/kg for effects on spontaneous motor behavior, decreased habituation capability with age, and poor performance in the swim-maze test.

A similar study in rats was conducted by Viberg et al. (2005). Single oral doses of 0, 0.8, 8.0, or 16 mg/kg BDE-99 (>98% purity) in a 20% fat emulsion were given by gavage to male Sprague-Dawley rats on PND 10, and the spontaneous motor behavior tests described above were carried out in 2-month-old rats. Rats exposed on PND 10 to 8.0 and 16 mg/kg BDE-99 displayed significantly less activity for locomotion, rearing, and total activity during the first 20-minute period, while during the third 20-minute period they were significantly more active than the control animals for all three behavioral variables. Rats receiving BDE-99 at 0.8 mg/kg did not show any difference from controls in locomotion or rearing activities over the three 20 minute test periods. A slight decrease in the total activity variable was seen only during the first 20-minute period but returned to control levels during the second and third 20-minute periods. The NOAEL in this study was 0.8 mg/kg and the LOAEL was 8.0 mg/kg for significant changes in spontaneous motor behavior in two-month-old rats exposed to BDE-99 on PND 10. The NOAEL/LOAEL values in this study indicate that rats are equally or perhaps less sensitive than mice to the spontaneous motor behavior effects of BDE-99.

A study was undertaken by Eriksson et al. (2002) to investigate whether behavioral disturbances observed in adult mice following neonatal exposure to BDE-99 are induced during a defined neonatal brain developmental window of unique biological susceptibility. On PND 3, 10, or 19, male NMRI mice were given by gavage a single oral dose of 0 or 8 mg/kg BDE-99 in a 20% fat emulsion. Spontaneous motor behavior tests (locomotion, rearing, and total activity) were measured over three 20-minute periods in 4-month-old male mice. Mice neonatally exposed to BDE-99 on PND 3 or 10 showed decreased activity during the first 20 minute period and increased activity during the last 20-minute period for all three behavioral variables compared with the control groups, with the effects being more pronounced in mice exposed to BDE-99 on PND 10. In mice neonatally exposed to BDE-99 on PND 19, there were no changes in the three behavioral variables compared with controls. The behavioral disturbances observed in adult mice, following neonatal exposure to BDE-99, are therefore

induced during a defined critical period of neonatal brain development, and mice exposed on PND 10 are most susceptible to the neurotoxic effects of BDE-99.

A study was conducted by Viberg et al. (2002) to determine whether changes in spontaneous behavior in adult mice neonatally exposed to BDE-99 would include effects on the cholinergic system and thereby would alter the response in the adult animal to the cholinergic agent nicotine. On PND 10, male NMRI mice received by gavage BDE-99 (>98% purity) at 0 or 8 mg/kg. Two-month-old mice were subjected to spontaneous motor behavior testing (locomotion, rearing, and total activity) for a 60-minute period, divided into three 20-minute periods. Directly after the spontaneous behavior test, the mice were given a single subcutaneous injection of saline solution (control) or 0.08 mg/kg nicotine base and were immediately tested again for spontaneous motor behavior during another 60-minute period. This amount of nicotine is known to cause an increased activity level in normal adult NMRI mice. A decrease in locomotion, rearing, and total activity over the 60-minute test period was observed in control mice in response to the diminished novelty of the test chamber, but BDE-99-treated animals displayed significantly less activity (hypoactivity) for all three variables during the first 20-minute period and significantly increased activity (hyperactivity) during the last 20 minute period. Pair-wise testing between the nicotine-injected and saline-injected mice showed, as expected, a significant increase in response to nicotine in the neonatally vehicle-treated mice during the first 20-minute period (60-80 minutes) for all three variables (locomotion, rearing, and total activity). In contrast, animals treated with BDE-99 on PND 10 and injected with nicotine at the age of 2 months showed significantly decreased activity during the first 20-minute period (60-80 minutes) compared with BDE-99-treated animals injected with saline. The authors concluded that neonatal exposure to BDE-99 on PND 10 can affect the cholinergic system, seen as changes in the adult mouse response to the cholinergic agent nicotine.

The objective of the Ankarberg (2003) study was to determine whether neonatal exposure to nicotine could affect the susceptibility of adult mice to BDE-99. On PND 10, NMRI mice received subcutaneous injections of saline or nicotine base at 0.033 mg/kg twice daily for 5 consecutive days (total daily dose, 0.066 mg/kg-day). At the age of 5 months, the animals received by gavage 0 or 8 mg/kg BDE-99 in a 20% fat emulsion. Mice were tested at the age of 5 months for spontaneous motor activity (locomotion, rearing, and total activity) for three 20-minute periods, 24 hours after exposure to BDE-99. Control animals, animals that received 0.066 mg/kg-day nicotine base neonatally but were not given the BDE-99, and animals that received only 8 mg/kg BDE-99 as adults showed a normal decrease in activity over the 60-minute test period, indicating a normal habituation pattern in response to the diminished novelty of the test chamber. However, animals that received nicotine on PND 10 and BDE-99 as adults showed a lack of habituation. These animals displayed hypoactive behavior in the first 20 minutes of the spontaneous behavior test but became hyperactive in the last 20 minutes

of the test period, indicating that the neonatal nicotine exposure affected the susceptibility to BDE-99 in the adult animals. At the age of 7 months, the animals were again tested for spontaneous motor behavior. The lack of habituation in the nicotine BDE-99-treated mice was even more pronounced, indicating a disturbance that worsens with age. Overall, this study indicates that neonatal nicotine exposure affects the susceptibility to BDE-99 in the adult animal.

The neurobehavioral effect of perinatal exposure to BDE-99 was investigated in mice (Branchi et al., 2002). BDE-99 dissolved in corn oil was administered by gavage at 0, 0.6, 6, or 30 mg/kg-day to groups of CD-1 Swiss female mice from gestational day (GD) 6 through PND 21, at which time the pups were weaned. Body-weight gain of pregnant females, pregnancy duration, proportion of successful deliveries, pup-sex ratio, and body-weight gain of pups from birth to weaning were not affected by treatment with BDE-99. Male and female pups were used in a series of tests to assess somatic and neurobehavioral development from PNDs 2-20. These tests were carried out every 2 days and included hair growth; day of eyelid and ear opening and of incisor eruption; righting, forelimb stick grasp, and forelimb placing reflexes; level and vertical screen tests; screen climbing test; and pole grasping test. Ultrasonic vocalization on PNDs 4, 8, and 12 and homing tests on PND 11 were also carried out on male and female pups. In addition, an open-field apparatus was used to test locomotion (horizontal movement), rearing (vertical movement), and thigmotaxis (time and distance traveled close to the walls) in males and one female for 30-minute sessions on PNDs 22 and 34 and for 60-minute sessions on PNDs 60 and 120. In the battery of tests carried out from PNDs 2-20, BDE-99 treatment did not affect somatic development (hair growth and day of eyelid and ear opening and incisor eruption). There was a statistically significant 2-day delay in appearance of screen-climbing response in the high-dose group (30 mg/kg-day); all other responses based on neuromotor coordination from PNDs 2-20 were not affected by BDE-99 treatment. No effects were seen in pups from any of the treatment groups on ultrasonic vocalization or homing performance assessed on PND 11 both for distance traveled and latency to reach the scent area. In the open-field test, there was no statistically significant difference in activity between controls and treatment groups on PND 22. However, BDE-99 exposure affected several behavioral/activity parameters in the open-field arena on PNDs 34, 60, and 120, indicating that behavioral alterations due to perinatal BDE-99 exposure seem to worsen with increasing age, becoming clearly evident around one month of age. On PND 34, mice were hyperactive in the 0.6 and 6 mg/kg-day dose groups but not at 30 mg/kg-day. Mice exhibited a statistically significant increase in rearing frequency, with mice in the 0.6 mg/kg-day dose group being more hyperactive than mice in the 6 mg/kg-day dose group. The mice in the 6 mg/kg-day dose group also exhibited signs of increased locomotion as evidenced by increases in distance traveled, although this was not apparent at 0.6 and 30 mg/kg-day. Thigmotactic response, considered an index of anxiety, was not affected at any dose. On PND 60, mice in the 0.6 mg/kg-day group, but not in the 6 and 30 mg/kg-day groups, displayed significantly

more locomotion compared with controls. Thigmotactic behavior on PND 60, measured as percent of time spent near the walls, was significantly lower at the medium dose only (6 mg/kg-day) in comparison with control mice, indicating a less marked fearful response in this treated group. At adulthood (PND 120), the 0.6 and 6.0 mg/kg-day groups displayed significantly lower levels of locomotion (in contrast to hyperactivity measured prior to PND 120) than controls during the last part of the 60-minute test session. At this age, rearing and thigmotaxis were not affected at any BDE-99 dose. The authors concluded that prenatal and postnatal exposure of mice to BDE-99 produced a transient hyperactivity, characterized by an inverted dose-response relationship, which ended around 4 months of age. The lack of a clear dose relationship in the behavior/activity changes in this study does not permit clear identification of the NOAEL/LOAEL for alteration in behavioral or activity parameters.

The effect of in utero exposure to BDE-99 on locomotor activity and male reproductive health was investigated in rat offspring (Kuriyama et al., 2005). Groups of Wistar rats were given by gavage single doses of 0, 0.06, or 0.3 mg/kg of BDE-99 (98% purity) in peanut oil on GD 6. Developmental landmarks (eruption of incisors, fur development, eye opening, and testes descent) and postnatal reflexes (development of spontaneous cliff-drop aversion reflex starting on PND 3 and ability to stay on a rotating rod for 3 minutes starting on PND 18) were evaluated in male and female pups. The eruption of incisors and the development of spontaneous cliff-drop aversion reflex were significantly delayed in the male offspring exposed to 0.3 mg/kg BDE-99. On PNDs 36 and 71, the circadian locomotor activity of male and female rats was evaluated over 24-hour periods. Locomotor activity was measured in individual offspring and included total activity measured as light beam interruption (LBI) counts per day, duration (hours) of activity per day, LBI count per active phase (an active phase is defined as when the animal begins to move until a pause), and duration of activity (minutes) per active phase. There was no difference between the sexes for all groups, and therefore the data for males and females were pooled. On PND 36, the total activity (LBI count), number of active hours per day, LBI counts per active phase, and duration of activity per active phase were significantly increased in the offspring of dams treated with 0.3 mg/kg BDE-99. On PND 71, both total activity and duration of activity per day were significantly increased at 0.06 and 0.3 mg/kg BDE-99 (Kuriyama et al., 2005).

The effects of in utero exposure to BDE-99 on body and organ weights and the male reproductive system of adult male offspring (PND 140) were also investigated in this study (Kuriyama et al., 2005). No effects were seen on body weight or absolute and relative liver, thymus, seminal vesicle, or prostate weights, sperm morphology, luteinizing hormone (LH), or testosterone in BDE-99-treated animals. Absolute and relative spleen weights were increased but not in a dose-dependent manner. Relative epididymis weight was significantly decreased at both doses, and relative testis weight was significantly decreased at the high dose only. Sperm numbers were significantly decreased compared with those in controls at both BDE-99

doses but not in a dose-dependent manner. Daily sperm production and spermatid count were significantly decreased at both doses in a dose-dependent manner.

Reproductive effects were also examined in this study (Kuriyama et al., 2005). Adult male offspring approximately 150 days old from the BDE-99-treated groups were mated with untreated females daily for 14 days to determine whether the males were fertile and could produce normal offspring. The dams were sacrificed on GD 21. Uterine and fetal weights, number of implantations, implantations per litter, viable fetuses per litter, percent total resorptions, and male/female sex ratio were all within the normal range of control in all treatment groups. Sexual behavior (ejaculatory, mounting and intromission latencies, intromission frequency, and number of penetrations before the first ejaculation) in 160-day-old male offspring (20/group) were also normal in all treatment groups compared with controls. The only effect seen was a significant decrease at both dose groups in the number of animals that had two or more ejaculations during 20 minutes of mating. No effects were seen on male fertility or sperm morphology at these doses. The LOAEL in this study was 0.06 mg/kg, the lowest dose tested, for increases in certain locomotor activity parameters on PND 71 and for decreased sperm production, spermatid count, and relative epididymis weight on PND 140.

The effect of BDE-99 on the female reproductive system was evaluated by Talsness et al. (2005), of the same research group as Kuriyama et al. (2005). A single dose of 0.06 or 0.3 mg/kg BDE-99 (98% purity) in peanut oil was administered by gavage to Wistar rats on GD 6. At approximately 5 months of age, 20 virgin female F_1 offspring from each group were mated with untreated males to evaluate fertility. Pregnancy rate, total implantation sites, mean implantation sites per gravid dam, total live fetuses per dam, resorption rate, and percentage of dams with resorptions in the F_1 females were not statistically different from controls at both doses of BDE-99. The only statistically significant effect noted was an increase in mean fetal weights at 0.06 mg/kg BDE-99 but not at 0.3 mg/kg BDE-99. Histological evaluation by electron or light microscopy of the ovary, uterus, and vagina was performed in the F_1 female offspring on PND 90. Electron and photomicrographs revealed qualitative ultrastructural changes in the ovaries and hyperplastic vacuolar degeneration of the vaginal epithelium in the F_1 offspring from the 0.06 and 0.3 mg/kg BDE-99. No significant changes were observed in the different ovarian follicle types following exposure to BDE-99, indicating that follicle numbers and maturation of follicles were unaffected. The LOAEL in this study was 0.06 mg/kg for qualitative ultrastructural changes in the ovaries and hyperplastic vacuolar degeneration of the vaginal epithelium in the F_1 offspring.

Among the 10 studies described above, the study of Viberg et al. (2004a) was selected as the principal study, and neurobehavioral developmental effects were identified as the critical effect. The primary reasons for selecting this study were (1) several different dose levels of BDE-99 were employed, (2) quantitative dose-response data were available with which to

conduct benchmark dose (BMD) modeling, (3) good model fits were obtained in subsequent BMD modeling, (4) a clear NOAEL was identified from this study, and (5) the results of this study are supported by several other studies in mice and rats. The RfD for BDE-99 was derived by using the BMD approach. The best-fit model was the power model, fit to data on rearing habituation in 8-month-old female mice, following exposure to BDE-99 on PND 10. The resulting BMD_{1SD} was 0.41 mg/kg, and the corresponding lower bound on the BMD ($BMDL_{1SD}$) was 0.29 mg/kg.

I.A.3. UNCERTAINTY FACTORS

UF = 3,000

The total UF of 3,000 was comprised of a factor of 10 to account for extrapolating animal data to humans (UF_A or interspecies variability), a factor of 10 to account for susceptible human subpopulations (UF_H or intrahuman variability), a factor of 3 to account for extrapolating from a single-dose exposure to a lifetime exposure (UF_S), and a factor of 10 to account for database deficiencies (UF_D). The rationale for application of each of these UFs is described below.

A 10-fold UF_A was used to account for laboratory animal to human interspecies differences. Although the toxicokinetics of BDE-99 in animals have been evaluated, no adequate description of toxicokinetics of BDE-99 in humans exists. The critical effect for deriving the RfD, altered behavior due to exposure during development, is expected to be relevant to humans. No quantitative data were identified to compare relative human and rodent sensitivity to these changes. However, given the longer period of brain development in humans as compared to rodents and the higher importance of cognitive function, it is appropriate to consider that humans may be more sensitive than rodents in the absence of specific data. Based on these considerations the default UF_A value of 10 was applied.

A default intraspecies UF_H of 10 was applied to account for variations in susceptibility within the human population (intrahuman variability). This factor accounts for the segment of the human population that may be more sensitive than the general population to exposure to BDE 99. A default value is warranted because insufficient information is currently available to assess human-to-human variability in BDE-99 toxicokinetics or toxicodynamics.

A UF_S of 3 was used to account for extrapolating from effects seen in a single-exposure neurodevelopmental study to a lifetime exposure. Exposure on PND 10 occurred during a period of rapid brain development in mice. Brain development does not continue at an equivalent rate over a mouse's lifetime and is more quiescent during adult life stages. Many brain structures have a very critical window during development in early life. Following BDE-99 exposure, toxicokinetic data suggest that a mouse urinary protein becomes functional some

time between PNDs 28 and 40, which leads to a dramatic increase in BDE-99 urinary excretion, especially in males. This increased excretion reduces the total body burden of BDE-99, including the levels of radiolabel reaching the brain 24-hours after dosing, in older mice compared with younger mice. These data suggest that the risk of neurodevelopmental effects in neonatal mice may be greater than in older mice because of rapid postnatal brain growth and coincident increased retention of BDE-99 and/or its metabolites. Therefore, chronic exposure is not expected to result in more serious effects. However, because the mice received only a single dose rather than repeated doses over multiple days within the hypothesized critical window, a threefold UF was applied.

A UF_L for LOAEL-to-NOAEL extrapolation was not used because the Agency's current approach is to address this factor as one of the considerations in selecting a benchmark response for benchmark dose modeling. In this case, a change in the mean equal to 1 SD of the control mean was assumed to represent a minimal biologically significant change.

An UF_D of 10 was used to account for database uncertainty. The available oral database for BDE 99 lacks prenatal developmental neurotoxicity studies and multigeneration reproductive toxicity studies, as well as conventional studies of subchronic and chronic toxicity. In addition, uncertainties regarding the effects of exposures during the prenatal period, extended postnatal exposures, and latent expression of early postnatal changes in the brain are addressed as a component of this database UF.

Application of a total UF of 3,000 to the $BMDL_{1SD}$ of 0.29 mg/kg results in an RfD for BDE-99 of 0.0001 mg/kg-day.

I.A.4. ADDITIONAL STUDIES/COMMENTS

The aim of the study by Skarman et al. (2005) was to determine the effects on plasma thyroxine (T_4) levels in juvenile mice following maternal gestational and lactational exposure to BDE-99. Groups of dams received 0 or 45 mg/kg of BDE-99 (>99% purity) in corn oil every third day, from GD 4 through PND 17, on a total of 10 occasions. The total dose of BDE-99 administered was therefore 450 mg/kg. Parallel groups of dams were similarly treated with a total dose of 450 mg/kg of a commercial pentaBDE (Bromkal 70-5DE with main constituents 37% BDE-99 and 35% tetraBDE-47). On GD 17, four dams from each treatment group were sacrificed and liver and blood samples collected. Pups were sacrificed on PND 11, 18, or 37 and liver and plasma samples collected. Dam and offspring body weights were not affected by BDE-99 or Bromkal treatment. Significantly increased liver-to-body-weight ratio was seen on PND 20 in dams treated with BDE-99 but not in their offspring on PND 11, 18, or 37. Pregnancy rate, gestation length, and litter size were not statistically different from controls. Plasma total and free T_4 in the pregnant dams on GD 17, in the postweaning dams on

PND 20, and in the offspring on PNDs 11, 18, and 37 were unaffected by BDE-99 treatment. On the other hand, plasma total and free T_4 were significantly reduced in the offspring of the Bromkal groups on PND 11 but returned to control levels by PND 18. Based on the above, the study of Skarman et al. (2005) shows that BDE-99 had no effect on plasma T_4 levels in dams and their offspring relative to controls at any sampling occasion, suggesting that other components in Bromkal are responsible for the reduction of T_4 levels in offspring on PND 11. One of the components of Bromkal is tetraBDE-47, which has been shown to cause a decrease in T_4 levels in mice and rats. These results indicate that interference with thyroid hormone homeostasis can vary significantly between PBDE homologs.

Hakk et al. (2002) examined the effect of BDE-99 on total T_4 (TT_4) plasma levels in young adult male Sprague-Dawley rats. A single oral dose of 8 mg/kg of ^{14}C-BDE-99 in corn oil was given to groups of conventional and bile-duct-cannulated rats. Average total plasma T_4 concentration (bound and free) was 1.7 µg/dL in the control rats. In the treated conventional rats, the average TT_4 levels increased approximately twofold to 3.2 µg/dL at 3 days after exposure and remained elevated at 3.0 µg/dL at 6 days after exposure, but by day 12 the levels returned to control levels of 1.9 µg/dL. TT_4 levels in plasma of the bile-duct-cannulated rats were not significantly different from control levels.

For more detail on Susceptible Populations, exit to the toxicological review, Section 4.7 (PDF).

I.A.5. CONFIDENCE IN THE ORAL RfD

Study -- Low
Data Base -- Low
RfD -- Low

Confidence in the principal study is low because the study protocol was unique and did not conform to health effects test guidelines for neurotoxicity, the dosing regimen did not include gestation and lactation exposure, several pups per litter were used for the behavioral testing, and only single doses were given. Confidence in the database is low because it lacks developmental and reproductive toxicity and neurotoxicity studies, and standard chronic or subchronic studies of systemic toxicity. As a result, the overall confidence in the RfD assessment is low.

For more detail on Characterization of Hazard and Dose Response, exit to the toxicological review, Section 6 (PDF).

I.A.6. EPA DOCUMENTATION AND REVIEW OF THE ORAL RfD

Source Document -- U.S. EPA (2008)

TThis document has been reviewed by EPA scientists, interagency reviewers from other federal agencies, and the public, and peer reviewed by independent scientists external to EPA. A summary and EPA's disposition of the comments received from the independent external peer reviewers and from the public is included in Appendix A of the Toxicological Review of 2,2',4,4',5-Pentabromodiphenyl Ether (BDE-99) (U.S. EPA, 2008). ***To review this appendix, exit to the toxicological review, Appendix A, Summary Of External Peer Review And Public Comments And Disposition (PDF)***.

Agency Completion Date -- 06/30/2008

I.A.7. EPA CONTACTS

Please contact the IRIS Hotline for all questions concerning this assessment or IRIS, in general, at (202) 566-1676 (phone), (202) 566-1749 (fax), or hotline.iris@epa.gov (email address).

I.B. REFERENCE CONCENTRATION (RfC) FOR INHALATION EXPOSURE

Substance Name — 2,2',4,4',5-Pentabromodiphenyl ether (BDE-99)
CASRN — 60348-60-9
Last Revised — 06/30/2008

The RfC is an estimate (with uncertainty spanning perhaps an order of magnitude) of a continuous inhalation exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. The RfC considers toxic effects for both the respiratory system (portal of entry) and for effects peripheral to the respiratory system (extrarespiratory effects). The inhalation RfC (generally expressed in units of mg/m^3) is analogous to the oral RfD and is similarly intended for use in risk assessments for health effects known or assumed to be produced through a nonlinear (presumed threshold) mode of action.

Inhalation RfCs are derived according to *Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry* (U.S. EPA, 1994). Because RfCs can also be derived for the noncarcinogenic health effects of substances that are carcinogens, it is essential to refer to other sources of information concerning the carcinogenicity of this

chemical substance. If the U.S. EPA has evaluated this substance for potential human carcinogenicity, a summary of that evaluation will be contained in Section II of this file.

I.B.1. INHALATION RfC SUMMARY

No data are available for deriving a reference concentration for BDE-99.

I.B.2. PRINCIPAL AND SUPPORTING STUDIES

Not applicable.

I.B.3. UNCERTAINTY FACTORS

Not applicable.

I.B.4. ADDITIONAL STUDIES/COMMENTS

Not applicable.

For more detail on Susceptible Populations, exit to the toxicological review, Section 4.7 (PDF).

I.B.5. CONFIDENCE IN THE INHALATION RfC

Not applicable.

For more detail on Characterization of Hazard and Dose Response, exit to the toxicological review, Section 6 (PDF).

I.B.6. EPA DOCUMENTATION AND REVIEW OF THE INHALATION RfC

Source Document -- U.S. EPA (2008)

This document has been reviewed by EPA scientists, interagency reviewers from other federal agencies, and the public, and peer reviewed by independent scientists external to EPA. A summary and EPA's disposition of the comments received from the independent external peer reviewers and from the public is included in Appendix A of the *Toxicological Review of 2,2',4,4',5-Pentabromodiphenyl Ether (BDE-99)* (U.S. EPA, 2008). ***To review this appendix, exit to the toxicological review, Appendix A, Summary Of External Peer Review And Public Comments And Disposition (PDF)***.

Agency Completion Date -- 06/30/2008

I.B.7. EPA CONTACTS

Please contact the IRIS Hotline for all questions concerning this assessment or IRIS, in general, at (202) 566-1676 (phone), (202) 566-1749 (fax), or hotline.iris@epa.gov (email address).

II. CARCINOGENICITY ASSESSMENT FOR LIFETIME EXPOSURE

Substance Name — 2,2',4,4',5-Pentabromodiphenyl ether (BDE-99)
CASRN — 60348-60-9
Section II. Last Revised — 06/30/2008

This section provides information on three aspects of the carcinogenic assessment for the substance in question: the weight-of-evidence judgment of the likelihood that the substance is a human carcinogen, and quantitative estimates of risk from oral and inhalation exposure. Users are referred to Section I of this file for information on long-term toxic effects other than carcinogenicity.

The rationale and methods used to develop the carcinogenicity information in IRIS are described in the *Guidelines for Carcinogen Risk Assessment* (U.S. EPA, 2005a) and the *Supplemental Guidance for Assessing Susceptibility from Early-Life Exposure to Carcinogens* (U.S. EPA, 2005b). The quantitative risk estimates are derived from the application of a low-dose extrapolation procedure, and are presented in two ways to better facilitate their use. First, route-specific risk values are presented. The "oral slope factor" is a plausible upper bound on the estimate of risk per mg/kg-day of oral exposure. Similarly, a "unit risk" is a plausible upper bound on the estimate of risk per unit of concentration, either per µg/L drinking water (see Section II.B.1.) or per $\mu g/m^3$ air breathed (see Section II.C.1.). Second, the estimated concentration of the chemical substance in drinking water or air when associated with cancer risks of 1 in 10,000, 1 in 100,000, or 1 in 1,000,000 is also provided.

II.A. EVIDENCE FOR HUMAN CARCINOGENICITY

II.A.1. WEIGHT-OF-EVIDENCE CHARACTERIZATION

Under the *Guidelines for Carcinogen Risk Assessment* (U.S. EPA, 2005a), there is "inadequate information to assess the carcinogenic potential" of 2,2',4,4',5-pentabromodiphenyl ether (BDE-99).

For more detail on Characterization of Hazard and Dose Response, exit to the toxicological review, Section 6 (PDF).

For more detail on Susceptible Populations, exit to the toxicological review, Section 4.7 (PDF).

II.A.2. HUMAN CARCINOGENICITY DATA

Not applicable.

II.A.3. ANIMAL CARCINOGENICITY DATA

Not applicable.

II.A.4. SUPPORTING DATA FOR CARCINOGENICITY

BDE-99 was not mutagenic in *Salmonella typhimurium* or *Escherichia coli* assays, with and without S9, or in the *Allium cepa* chromosome aberration test (Evandri et al., 2003).

II.B. QUANTITATIVE ESTIMATE OF CARCINOGENIC RISK FROM ORAL EXPOSURE

II.B.1. SUMMARY OF RISK ESTIMATES

Not applicable.

II.B.2. DOSE-RESPONSE DATA

Not applicable.

II.B.3. ADDITIONAL COMMENTS

Not applicable.

II.B.4. DISCUSSION OF CONFIDENCE

Not applicable.

II.C. QUANTITATIVE ESTIMATE OF CARCINOGENIC RISK FROM INHALATION EXPOSURE

II.C.1. SUMMARY OF RISK ESTIMATES

Not applicable.

II.C.2. DOSE-RESPONSE DATA

Not applicable.

II.C.3. ADDITIONAL COMMENTS

Not applicable.

II.C.4. DISCUSSION OF CONFIDENCE

Not applicable.

II.D. EPA DOCUMENTATION, REVIEW, AND CONTACTS (CARCINOGENICITY ASSESSMENT)

II.D.1. EPA DOCUMENTATION

Source Document -- U.S. EPA (2008))

This document has been reviewed by EPA scientists, interagency reviewers from other federal agencies, and the public, and peer reviewed by independent scientists external to EPA. A summary and EPA's disposition of the comments received from the independent external peer reviewers and from the public is included in Appendix A of the *Toxicological Review of 2,2',4,4',5-Pentabromodiphenyl Ether (BDE-99)* (U.S. EPA, 2008). ***To review this appendix, exit to the toxicological review, Appendix A, Summary Of External Peer Review And Public Comments And Disposition (PDF)***.

II.D.2. EPA REVIEW

Agency Completion Date -- 06/30/2008

II.D.3. EPA CONTACTS

Please contact the IRIS Hotline for all questions concerning this assessment or IRIS, in general, at (202) 566-1676 (phone), (202) 566-1749 (fax), or hotline.iris@epa.gov (email address).

III. [reserved]
IV. [reserved]
V. [reserved]

VI. BIBLIOGRAPHY

Substance Name — 2,2',4,4',5-Pentabromodiphenyl ether (BDE-99)
CASRN — 60348-60-9

VI.A. ORAL RfD REFERENCES

Ankarberg, E. (2003) Neurotoxic effects of nicotine during neonatal brain development. Comprehensive summaries of Uppsala Dissertations from the Faculty of Science and Technology 907. Acta Universitatis Upsaliensis, Uppsala, Sweden.

Branchi, I; Alleva, E; Costa, LG. (2002) Effects of perinatal exposure to a polybrominated diphenyl ether (PBDE 99) on mouse neurobehavioural development. Neurotoxicol 23(3):375-384.

Carlson, GP. (1980a) Induction of xenobiotic metabolism in rats by short-term administration of brominated diphenyl ethers. Toxicol Lett 5:19-25.

Carlson, GP. (1980b) Induction of xenobiotic metabolism in rats by brominated diphenyl ethers administered for 90 days. Toxicol Lett 6:207-212.

Eriksson, P; Jakobsson, E; Fredriksson, A. (2001) Brominated flame retardants: a novel class of developmental neurotoxicants in our environment? Environ Health Perspect 109(9):903-908.

Eriksson, P; Viberg, H; Jakobsson, E; et al. (2002) A brominated flame retardant, 2,2',4,4',5-pentabromodiphenyl ether: uptake, retention, and induction of neurobehavioural alterations in mice during a critical phase of neonatal brain development. Toxicol Sci 67(1):98-103.

Hakk, H; Larsen, G; Klasson-Wehler, E. (2002) Tissue disposition, excretion and metabolism of 2,2',4,4',5-pentabromodiphenyl ether (BDE-99) in the male Sprague-Dawley rat. Xenobiotica 32(5):369-382.

Kuriyama, SN; Talsness, CE; Grote, K; et al. (2005) Developmental exposure to low dose PBDE 99: effects on male fertility and neurobehavior in rat offspring. Environ Health Perspect 113:149-154.

Skarman, E; Darnerud, PO; Ohrvik, H; et al. (2005) Reduced thyroxine levels in mice perinatally exposed to polybrominated diphenyl ethers. Environ Toxicol Pharmacol 19:273-281.

Talsness, CF; Shakibaei, M; Kuriyama, S; et al. (2005) Ultrastructural changes observed in rat ovaries following in utero and lactational exposure to low doses of a polybrominated flame retardant. Toxicol Lett 157:189-202.

U.S. EPA (Environmental Protection Agency). (2008) Toxicological review of 2,2',4,4',5-pentabromodiphenyl ether (BDE-99). Integrated Risk Information System (IRIS), National Center for Environmental Assessment, Washington, DC. Available online at http://www.epa.gov/iris.

Viberg, H; Fredriksson A; Eriksson, P. (2002) Neonatal exposure to the brominated flame retardant 2,2',4,4',5-pentabromodiphenyl ether causes altered susceptibility in the cholinergic transmitter system in the adult mouse. Toxicol Sci 67(1):104-107.

Viberg, H; Fredriksson, A; Eriksson, P. (2004a) Investigations of strain and/or gender differences in developmental neurotoxic effects of polybrominated diphenyl ethers in mice. Toxicol Sci 81:344-353.

Viberg, H; Fredriksson, A; Jakobsson, E; et al. (2004b) Neonatal exposure to the brominated flame-retardant, 2,2',4,4',5-pentabromodiphenyl ether, decreases cholinergic nicotinic receptors in hippocampus and affects spontaneous behaviour in the adult mouse. Environ Toxicol Pharmacol 17:61-65.

Viberg, H; Fredriksson, A; Eriksson, P. (2005) Deranged spontaneous behavior and decrease in cholinergic muscarinic receptors in hippocampus in the adult rat, after neonatal exposure to the brominated flame-retardant, 2,2',4,4',5-pentabromodiphenyl ether (PBDE 99). Environ Toxicol Pharmacol 20:283-288.

VI.B. INHALATION RfC REFERENCES

U.S. EPA. (1994) Methods for derivation of inhalation reference concentrations and application of inhalation dosimetry. Environmental Criteria and Assessment Office, Office of Health and Environmental Assessment, Cincinnati, OH; EPA/600/8-90/066F. Available from the National Technical Information Service, Springfield, VA, PB2000-500023, and online at http://cfpub.epa.gov/ncea/raf/recordisplay.cfm?deid=71993.

U.S. EPA. (2008) Toxicological review of 2,2',4,4',5-pentabromodiphenyl ether (BDE-99). Integrated Risk Information System (IRIS), National Center for Environmental Assessment, Washington, DC. Available online at at http://www.epa.gov/iris.

VI.C. CARCINOGENICITY ASSESSMENT REFERENCES

Evandri, MG; Mastrangelo, S; Costa, LG; et al. (2003) In vitro assessment of mutagenicity and clastogenicity of BDE-99, a pentabrominated diphenyl ether flame retardant. Environ Mol Mutagen 42:85-90.

U.S. EPA. (2005a) Guidelines for carcinogen risk assessment. Federal Register 70(66):17765-18717. Available online at http://www.epa.gov/cancerguidelines/.

U.S. EPA. (2005b) Supplemental guidance for assessing susceptibility from early-life exposure to carcinogens. Risk Assessment Forum, Washington, DC; EPA/630/R-03/003F. Available online at http://www.epa.gov/cancerguidelines.

U.S. EPA. (2008) Toxicological review of 2,2',4,4',5-pentabromodiphenyl ether (BDE-99). Integrated Risk Information System (IRIS), National Center for Environmental Assessment, Washington, DC. Available online at http://www.epa.gov/iris.

VII. REVISION HISTORY

Substance Name — 2,2',4,4',5-Pentabromodiphenyl ether (BDE-99)
CASRN — 60348-60-9
File First On-Line - 06/30/2008

Date	Section	Description
06/30/2008	I., II.	RfD, RfD, and cancer assessment first on line

VIII. SYNONYMS

Substance Name — 2,2',4,4',5-Pentabromodiphenyl ether (BDE-99)
CASRN — 60348-60-9
Section VIII. Last Revised — 06/30/2008

- Benzene, 1,2,4,-tribromo-5-(2,4-dibromophenoxy)-
- 2,2',4,4',5-pentabromodiphenyl ether
- BDE-99
- Diphenyl ether, pentabromo
- PBDE 99