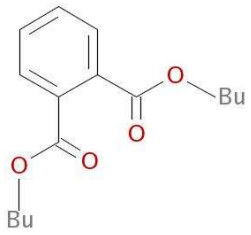
Dibutyl phthalate (DBP)
(CAS# 84-74-2)

(Synonyms: Di-n-butylphthalate, 1,2-Benzenedicarboxylic acid, dibutyl ester (9CI), Phthalic acid, dibutyl ester (6CI, 8CI), Bis-n-butyl phthalate, Butyl phthalate, DBP, DBP (ester), Dibutyl o-phthalate, Di(n-butyl) 1,2-benzenedicarboxylate, n-Butyl phthalate, Palatinol C, Phthalic acid di-n-butyl ester



Dibutyl phthalate (DBP) 8-hour REL

Reference Exposure Level	**0.23 μg/m^3**
Critical effects	Histopathological changes
Hazard Index target	Testis and mammary glands

1 Physical and Chemical Properties

Physical form	oily liquid
Molecular Formula	$C_{16}H_{22}O_4$
Density	1.045 g/cm^3 @ 20 ºC
Boiling point	340ºC @1,013 hPa
Melting point	-69 ºC
Vapor pressure	$9.7 + 3.3 . 10^{-5}$ hPa at 25ºC
Log K_{OW}	4.57
Solubility	*water soluble at 10 mg/l at 20*ºC

2 Production, Use, and Exposure

Dibutyl phthalate (DBP) is produced by the reaction of phthalic anhydride with n-butanol in the presence of concentrated sulphuric acid as a catalyst. Excess alcohol is recovered and recycled and the di-n-butyl phthalate is purified by vacuum distillation and/or activated charcoal. In 1998 the production volume of DBP in the EU was estimated at 26,000 tons, of which 8,000 tons was thought to be exported outside the EU (European Chemicals Bureau, 2000).

The largest usage of DBP in general is as a plasticizer in resins and polymers such as polyvinyl chloride (PVC). Plasticizers are incorporated into a plastic to increase its workability and distendability. DBP is also used in adhesives, printing inks, sealants/grouting agents, nitrocellulose paints, film coatings, and glass fibers. DBP is used in cosmetics as follows: a perfume solvent and fixative, a suspension agent for solids in aerosols, a lubricant for aerosol

valves, an anti-foamer, a skin emollient, and a plasticizer in nail polish and fingernail elongators (IPCS/WHO, 1995). The potential for high exposures to DBP in nail salons is being assessed by ARB.

DBP is or may be produced in the following chemical industries: basic chemicals (production of dibutyl phthalate); polymer industry (plasticizer in PVA, PVC) or rubber industry; solvents for nitrocellulose esters, colors, oils, and natural resins; softener; lacquer and varnish industry; the printing industry (ink); as an additive in the textile and pigment industry; and in insecticides. DBP is in many consumer products including: home furnishing, paints, clothing and cosmetic products (European Chemicals Bureau, 2000).

The human population may be exposed to dibutyl phthalate (DBP) by inhalation, ingestion or dermal contact at the workplace, from use of consumer products, and indirectly via the environment. Human exposure via the environment may occur through contact with contaminated air, water, soil or food. Exposure is sufficiently widespread that DBP in humans is monitored in the NHANES program. Workers in the polymer industry are potentially exposed, especially those workers that may have more direct contact with the substance (European Chemicals Bureau, 2000).

3 Pharmacokinetics and Metabolism

After oral administration in rats and hamsters, dibutyl phthalate (DBP) is rapidly absorbed and excreted. In rats and hamsters, more than 90% of the oral dose was excreted in urine within 24 to 48 hours and fecal excretion was low (1.0-8.2%) (European Chemicals Bureau, 2000). Humans can be exposed to DBP orally, through contaminated drinking water and food, as well as dermally via personal care products and contact with DBP-containing plastics. Following dermal exposure, rats absorbed DBP, 60% of the dose was excreted in urine within 7 days and 12% was found in the feces (European Chemicals Bureau, 2000). According to an *in vitro* study, human skin absorbed DBP slower (2.40 µg/cm^2/hour) than rat skin (93.35 µg/cm^2/hour) (European Chemicals Bureau, 2000). Data on absorption after exposure by inhalation were not available.

The major part of DBP is hydrolysed to mono-n-butyl phthalate (MBP) and the corresponding alcohol prior to absorption by the small intestines, but hydrolysis can also occur in liver and kidneys. The metabolites that occur in urine are MBP, MBP-glucuronide, various ω- and ω-1-oxidation products of MBP (more polar ketones, carboxylates) and a small amount of free phthalic acid. Species differences in the excretion of MBP and its glucuronide were observed; rats excreted a larger proportion of unconjugated MBP in urine than hamsters (European Chemicals Bureau, 2000). There are no data on biotransformation after dermal exposure or exposure by inhalation.

A substantial fraction of DBP is initially excreted in the bile and subsequently enters the enterohepatic circulation. No significant accumulation in tissues was observed in laboratory animals after oral as well as dermal exposure; limited inhalation data revealed an indication for some accumulation in tissues.

DBP levels were measured after inhalation of 50 mg/m^3 by rats for 6 hours/day, for 3 or 6 months and DBP was found in brain, lungs, liver, kidneys, and testes. After exposure to 0.5 mg/m^3, DBP was detected in brain after 3 and 6 months, in lungs (2 of 3 animals after 6 months), liver (1 of 3 animals after 3 and 6 months), kidneys (1 of 3 animals after 3 and 6 months), and testes (in 3 animals after 3 months and 1 after 6 months) (Kawano et al., 1980 as cited in European Chemicals Bureau, 2000).

After oral administration of DBP to rats, mono-n-butyl phthalate (MBP) was detected in urine as well as MBP glucuronide, various oxidation products of MBP (more polar ketones and carboxylates), and a small amount of free phthalic acid. Species differences have been observed in the excretion of unconjugated and conjugated MBP (ratio of MBP-glucuronide:unconjugated MBP is 1 in rat, 1.5 in guinea-pig and 2.3 in hamster) in vivo. In vitro studies with liver homogenates from rat, baboon, and ferret have demonstrated hydrolysis of DBP to MBP. Hydrolysis of DBP to MBP is very rapid in rat liver microsomal fractions in vitro. There are also species differences in phthalate diester hydrolase activity: baboon>rat>ferret (European Chemicals Bureau, 2000).

An oral study in Sprague-Dawley rats using 500 or 1,500 mg ^{14}C-labelled DBP/kg bw on day 14 of gestation, investigated transplacental transfer of DBP and its metabolites. The level of radioactivity in the maternal plasma was three times greater than in the placenta and embryo. The radioactivity in embryonic tissues was only 0.12-0.15% of the administered dose. Most of the radioactivity in maternal plasma, placenta, and the embryo was in the form of MBP and MBP-glucuronide. Only small amounts of unchanged DBP were found and there was no evidence of accumulation in maternal or embryonic tissues.

4 Acute Toxicity

4.1 Human Toxicity

One study concerning accidental ingestion of DBP (10 g) by a 23-yr old man has been reported. Symptoms included nausea, vomiting and dizziness followed by lacrimation, photophobia and pain in the eyes several hours later. Finally the cornea was severely damaged (keratitis erosiva). Urinalysis showed microhaematuria, oxalate crystals and pathological leucocyte counts. Recovery occurred within 14 days after treatment with mydriatics and antibiotics (Cagianut, 1954).

4.2.1 Animal Toxicity

Several acute studies have been carried out with different species and by different routes. They are summarized in Table 4.2.1. For dibutyl phthalate (DBP), the inhalation 4h LC_{50} value for the rat is 15.68 mg/L, the rat oral LD_{50} value is 6,300 mg/kg bw, and the dermal LD_{50} is >20,000 mg/kg bw for the rabbit (European Chemicals Bureau, 2000).

Acute studies by inhalation revealed a 2-h LC_{50} value for the mouse of 25 mg/L (first LC_{50} in Table 4.2.1), exposure resulted in pronounced irritation of mucous membranes of the eyes and

upper respiratory tract, slowed respiration, ataxia, and paralysis of hind legs (Voronin, 1975, results summarized in European Chemicals Bureau, 2000).

Table 4.2.1 Studies of Acute Toxicity of DBP in Animals

Acute Toxicity	Species	Results	Reference*
Inhalation	**mouse**	LC_{50} (2 h) 25 mg/L	Voronin, 1975
	rat	LC_{50} (4 h) .15.68 mg/L	Greenough et al 1981
	rat	LC_{50} (n/a) 4.25 mg/L	RTECS, 1993
Oral	**mouse**	LD_{50} 5,289 mg/kg bw	RTECS, 1993
	mouse	LD_{50} 4,840 mg/kg bw	BIBRA, 1987
	rat	LD_{50} 8,000 mg/kg bw	Smith, 1953
	rat	LD_{50} 6,300 mg/kg bw	BASF, 1961
	guinea-pig	LD_{50} 10,000 mg/kg bw	RTECS, 1993b
Dermal	**rabbit**	LD_{50} >20,000 mg/kg bw	Clayton and Clayton, 1994; RTECS, 1993
Other routes			
i.v.	mouse	LD_{50} 720 mg/kg bw	RTECS, 1993
i.m.	rat	LD_{50} >8,000 mg/kg bw	Smith, 1953
i.p.	mouse	LD_{50} 3,400 – 4,000 mg/kg bw	BASF, 1961; Calley et al., 1966; Lawrence et al., 1975.
i.p.	rat	LD_{50} 3,178 mg/kg bw	Singh et al., 1972
i.p.	rat	LD_{50} ca.4,200 mg/kg bw	BASF, 1958
s.c.	mouse	LD_{50} 20,800 mg/kg bw	RTECS, 1993

***** Table with citations taken from European Chemicals Bureau Risk Assessment Report (European Chemicals Bureau, 2000)

Sprague-Dawley rats (n = 5 males, 5 females) were exposed for 4 hours to an aerosol of 15.68 mg DBP/L of air and observed for 14 days. A second delivery of DBP was tested at 12.45 and 16.27 mg/L one month later. Respirable fraction (diameter <4.7 µm) was 56.9, 64.4 and 59.9% at 15.68, 12.45 and 16.27 mg/L, respectively. In the 15.68 mg/L group 2/5 male and 3/5 female animals died, whereas no mortality was observed in the 12.45 and 16.27 mg/L groups. The LC_{50} value was estimated to be 15.68 mg/L in this study which was performed under GLP conditions (second LC_{50} in Table 4.2.1). At 15.68 mg/L a reduction in respiratory rate was also observed. Lung/body weight ratios were elevated at 15.68 mg/L but lower than controls in males at 12.45 and 16.27 mg/L. Macroscopy of the lungs revealed red/dark foci in several animals scattered amongst the treatment groups. One male and one female rat exposed to 15.68 mg/L had white foci in all lung lobes. Dark red areas were seen in the lungs of 2 females at 12.45 mg/L and in 1 male and 1 female rat at 16.27 mg/L (Greenough et al., 1981, results summarized in European Chemical Bureau Risk Assessment).

A third LC_{50} study in the rat revealed a value of 4.25 mg/L (third LC_{50} in Table 4.2.1). However the original Russian report of this study is not available, only a summary is provided and the exposure time was not given (RTECS, 1993c, results summarized in European Chemical Bureau Risk Assessment).

Cats exposed for 5.5 h to 1 mg/L showed irritation of nasal mucous membranes as did mice exposed for 2 h to 0.25 mg/L (no further data available) (BIBRA, 1987; BUA, 1987, results summarized in European Chemical Bureau Risk Assessment). At a concentration of 11 mg/L, salivation, restlessness and languor were induced in cats. Rapid recovery was seen after

cessation of exposure (no further data are available) (BUA, 1987, results summarized in European Chemical Bureau Risk Assessment).

4.3 Repeated Dose Toxicity

Concerns for adverse local effects in the respiratory tract arise as a consequence of repeated inhalation exposure in all occupational exposure scenarios.

In a 28-day inhalation study in Wistar rats to 509 mg DBP/m^3 (~0.5 mg DBP/L) as aerosol (head-nose exposed for 6 hours/day, 5 days/week exposure for 4 weeks) caused red crust formation of snouts. At all concentrations (1.18, 5.57 or 509 mg/m3) including the lowest 1.18 mg/m3 (~0.001 mg/L) local histopathological effects in nasal cavity and larynx were seen, but no signs of inflammation. A dose-dependent increase of hyperplasia of mucous cells at some sites of levels II, III and IV of the nasal cavity and a dose-dependent increased incidence of squamoid metaplasia at level I of the larynx was observed (Gamer et al., 2000, results summarized in European Chemical Bureau Risk Assessment).

Table 4.3.1. Repeated Dose Toxicity Test of DHP

Inhalation Studies	NOAEL	LOAEL	Effects	References
Subchronic,Wister rats (4 wk, 6 hr/d, 5 d/wk; 0, 1.18, 5.57 or 509 mg/m3)	n/a (LOAEL was lowest dose tested)	1.18 mg/m^3	local histopathological effects in nasal cavity and larynx	Gamer et al., 2000

For repeated inhalation exposure a NOAEL of 509 mg DBP/m^3 (the highest concentration tested) for systemic effects including neurotoxic effects can be established based on the 28-day inhalation study in rats performed according to current standards. For local effects after repeated inhalation exposure, a LOAEL of 1.18 mg/m^3 can be derived from the same 28-day inhalation study.

5 Other Toxicity

5.1 Human Studies

5.1.1 Dermal

Repeated dermal exposure arising from aerosol forming activities presents a concern for general systemic toxicity.

A 44-year-old man noticed eczema under a plastic watch strip on the left wrist. After transferring the watch to the right wrist, eczema also occurred there. Patch tests with the plastic strip, 20% colophony, 1% p-t-butylphenol, butylphenol formaldehyde resin and 5% DBP were all positive (solvent not given) (Husain, 1975)(Husain, 1975).

A 71-year-old woman suffered from recurrent “ear infections” since she wore a hearing aid. She developed dermatitis behind the ears and on the temples, where there was contact with the spectacle frames. Patch tests with 5% dibutyl phthalate in petrolatum, 5% dimethyl phthalate in petrolatum or 5% diethyl phthalate in petrolatum gave positive results. Patch tests with scrapings

from the spectacle frame or the hearing aid gave less positive reactions (Oliwiecki et al., 1991)(Oliwiecki et al., 1991).

Workers in a factory producing shoes from PVC granulate were patch tested with dibutylphthlate. Two groups of 30 workers, with and without dermatitis, respectively, were used. A control group of 30 persons was included in the study. Three of 30 Workers with dermatitis and 5/30 without dermatitis reacted positive at patch testing, while none of the controls reacted. Concentration of DBP and solvent used at patch testing was not given (Vidovic and Kansky, 1985).

Routine patch testing with a mixture of phthalate esters (2% dimethyl phthalate, 2% diethyl phthalate and 2% dibutyl phthalate in petrolatum) revealed one positive reaction in 1,532 persons tested (Schulsinger and Mollgaard, 1980)(Schulsinger and Mollgard, 1980).

Cosmetic products (nail polish with 6 or 9% DBP or deodorant with 4.5% DBP) or 5% DBP in petrolatum were patch tested on 13-159 persons in 11 different studies. The studies included 48-hour closed patch tests, modified maximization tests, modified repeated insult patch tests, 21-day cumulative irritancy tests, prophetic patch tests, controlled use studies (lasting 2 days or 4 weeks). In the majority of the studies (9/11) no irritation, contact sensitisation or photosensitisation was observed. Slight irritation was observed in 2/11 studies exposed to a 9% DBP nail polish and a 4.5% DBP deodorant, respectively. The persons received twenty-one 23 to 24-hour lasting patches on the same site of the back (summary only available) (European Chemicals Bureau, 2000).

5.2 Animal Studies

Animal studies on eye and skin irritation were inconclusive.

5.2.1 Oral study

An oral NOAEL of 152 mg/kg bw was derived from a 3-month dietary study in rats performed according to current standards. A NOAEL of 19.9 mg/kg bw for peroxisomal proliferation in rats was found in a special study examining this effect. However, it has to be noted that humans have a relative low sensitivity for this effect.

5.2.2 Developmental and Reproductive studies

Based on the available developmental studies in mice, an oral NOAEL of 100 mg/kg bw was derived for teratogenicity, embryotoxicity and maternal toxicity. At the next higher dose-level of 400 mg/kg bw, embryotoxic and teratogenic effects were seen in the presence of maternal toxicity.

In some special in vitro assays, DBP showed weak estrogenic activity. However, the estrogenic effects were not confirmed with in vivo studies. Therefore, the relevance of the estrogenic effects observed in vitro for the in vivo estrogenic toxicity of DBP is questionable. Moreover results of developmental studies described above were indicative of an antiandrogenic effect of

DBP rather than an estrogenic effect. The epidemiological study on possible reproductive effects in occupationally exposed women is inadequate for assessment of possible reproductive effects caused by DBP in humans in the working environment.

The fertility of male mice did not appear to be affected up to the highest dose-level of 1.0% in the diet (equivalent to 1410 mg/kg bw) in a one-generation study while female fertility was clearly affected at this dose-level. At 1.0% in the diet also embryotoxic effects were observed. The NOAEL in this study in mice is 0.3% in the diet equivalent to 420 mg/kg bw based on effects on maternal fertility and embryotoxicity. Concerning the available reproduction studies in rats a NOAEL of 50 mg/kg bw was established based on embryotoxicity in a one-generation reproduction study with exposure of females only. The same study protocol with exposure of male animals only, gave a NOAEL of 500 mg/kg bw. However in a two-generation reproduction study in rats with a continuous breeding protocol and with exposure of both male and female animals the lowest dose-level of 0.1 % in the diet (52 mg/kg bw for males and 80 mg/kg bw for females) appeared to be a LOAEL based on embryotoxic effects (Wine et al., 1997).
In conclusion, effects on pup weight and number of live pups per litter were seen in the absence of maternal toxicity at the lowest dose-level of 52 mg/kg bw in a 2-generation reproduction study in rats with a continuous breeding protocol. Other available reproduction studies in rats showed effects on fertility and embryotoxic effects at oral doses of 250 mg/kg bw.

Reproduction or fertility studies with dermal exposure or exposure by inhalation to DBP are not available.

Developmental studies in rats and mice have been performed. Embryotoxic as well as teratogenic effects were observed. In a study in mice the dose-level of 0.05% in the diet, equivalent to 100 mg/kg bw, was a NOAEL for maternal toxicity, embryotoxicity and teratogenicity. In a second study in mice 0.2% in the diet (ca. 350 mg/kg bw) was a NOAEL for embryotoxicity; in this last study the NOAEL for maternal toxicity and teratogenicity is 0.4% in the diet (ca. 660 mg/kg bw). In this study there is a limited evidence for teratogenicity at 1.0% in the diet (ca. 2100 mg/kg bw) in the presence of maternal toxicity. However this second study showed limitations regarding the number of animals and reporting.

In several recent developmental studies in rats delayed preputial separation and a markedly disturbed development of the male reproductive tract (internal and external) of rat offspring exposed via their mothers during gestation or during gestation and lactation, was observed at oral doses . 250 mg/kg bw. Maternal toxicity was seen at oral doses of 500 mg/kg bw. In female offspring sporadic cases of reproductive tract malformations were observed at doses of 250 mg/kg bw. Age at vaginal opening and estrus cyclicity were not affected. At the lowest oral dose level of 100 mg DBP/kg bw, studied in developmental studies in rats, still delayed preputial separation in male progeny was seen. The results of these studies indicate that DBP does not possess estrogenic activity but rather shows antiandrogenic activity. A NOAEL was not derived from the available developmental studies in rats.

Developmental studies with dermal exposure or exposure by inhalation to DBP are not available.

Table 5.2.1. Major findings from key studies for the reproductive and developmental effects of DBP in animal studies[1]

Study/ Reference	Animals	Treatment	General Toxicity	Major DART & LOAELs	NOAEL
Lee et al., 2004	Sprague-Dawley rats, 6-8 dams per group. Soy-free diet for dams and regular rodent diet for offspring after weaning.	Feed, 0, 20, 200, 2000, and 10,000 ppm of DBP in diet from gestational day (GD) 15 until postnatal day (PND) 21.	Slight decrease in body weight gain in dams from GD15 to GD 20 at 20, and 10,000 ppm only. No effect on feed consumption. Increased liver and kidney weights at 10,000ppm.	Developmental effects excluding postnatal exposure: reduced male-tofemale ratio and reduced AGD in male offspring on PND 2 at 10,000ppm. Female Reproductive Effects: hypoplasia of alveolar buds in the mammary glands in female offspring on PND 21in all treated groups, but not in adulthood (postnatal week 11 or 20). Male Reproductive Effects: Histopathological changes in the testis and mammary glands at >= 20ppm. **LOEL: 20 ppm (1.5-3.0 mg/kg-day)**	Not observed, based on endpoints for male and female reproductive effects.
Wine et al., 1997	Sprague-Dawley rats, NIH-07 diet NTP-RACB protocol	Feed, 0, 0.1, 0.5, and 1.0% (w/w) in diet.	Reduced body weights, increased liver and kidney weights at 1.0%.	Male and female reproductive effects: 1.0%: decreased live pups per litter and live pup weights. Abnormalities in development of the male reproductive system. 0.5%: Similar to 1.0% group. 0.1%: Reduction in live pups per litter in F1 pups and in live pup weight in F2 pups. LOEL: 0.1% (80 mg/kg-day in females)	Not observed based on endpoints for developmental effects.
Lehmann et al., 2004	Sprague-Dawley rats, 7 rats for the control group and 5 for each Treated group.	Gavage, 0. 0.1, 1.0, 10, 30, 50, 100, and 500 mg/kg-day from GD 12-19. For measurement of fetal testicular testosterone levels, 3-4 male fetuses from 1-4 litters per dose group were used.	Not reported.	Developmental and male reproductive effects: Dose-dependent decline in expression of genes and corresponding proteins involved in cholesterol transport, steriodogenesis, or cell survival. Significant reduction was observed at 0.1 mg/kg-day in genes for 3beta-HSD and c-Kit. Dose-dependent reduction in fetal testicular testosterone concentrations; obvious decline at 30 mg/kg-day, but statistically significant at >= 50 mg/kgday. LOEL: 30 mg/kg-day	10 mg/kg-day based on functional changes in the testis.
Mylchreest et al., 2000	Sprague-Dawley rats, 11-20 pregnant dams per	Gavage, 0, 0.5, 5, 50, 100, or 500 mg/kgday, GD 12-21.	No apparent maternal or general toxicity at all dose levels.	Developmental and male reproductive effects: 500 mg/kg-day: retained nipples, decreased AGD, reproductive tract malformations, and	50 mg/kg-day, based on lack of statistically significant increase in

	group	Offspring examined on PND 1, 14, 21, or 38.		histopathological changes in the testis. 100 mg/kg-day: significant increase in the incidence of retained areolas or nipples. 50 and 5 mg/kg-day: obvious, but not statistically significant increase in the incidence of retained areolas or nipples. LOEL: 100 mg/kg-day	retained areolas or nipples. More discussions on this NOEL in the text.
Zhang et al., 2004	Sprague-Dawley rats, 20 pregnant dams per group	Gavage, 0, 50, 250, or 500 mg/kg-day, GD 1 – PND 21. Male offspring examined on PND 14, 21, and 70.	No apparent maternal or general toxicity at all dose levels.	Developmental, male, and female reproductive effects: 250 and 500 mg/kg-day: apparent adverse effect on the development of the male reproductive system. LOEL: 250 mg/kg-day	50 mg/kg-day

[1]Taken from OEHHA 2007 "Proposition 65 Maximum Allowable Dose Level (MADL) for Reproductive Toxicity for Di(n-butyl)phthalate (DBP)."

The study by Mychreest et al. (2000) characterized the dose-dependent adverse effects of DBP on the male reproductive system following gestational and lactational exposure. Based on statistically significant increase on PND 14 in the incidence of retained areolas or nipples in the male offspring from dams treated with 100 or 500 mg/kg-day DBP, the authors concluded that 50 mg/kg-day was the no observable adverse effect level (NOAEL) for DBP in their study. The increase in the incidence of retained nipples is statistically significant at doses ≥ 100 mg/kg-day, but the increase at doses of 5 and 50 mg/kg-day (42.1% and 50% based on litters, respectively, compared to 26.3% in the control) is apparent (Mylchreest et al., 2000)(Mychreest et al., 2000).

Abnormal retention of areolas or nipples in males indicates alteration in the mammary gland. Numerous studies have shown in animals and humans that sex hormones (androgens and estrogens) during pregnancy and early postnatal life determines the development and growth of the mammary gland (OEHHA 2007). Inhibition of testosterone production and/or action by treatment with anti-androgenic compounds during the late gestational period in rats not only causes development of breast tissues and nipples in male fetuses similar to that in females, but also results in abnormal expression of estrogen receptors and C-19 steroid aromatase. Therefore, the developmental status of the nipple and mammary gland is an indicator for the function of androgens during the critical developmental period. Moreover, nipples, once developed, remain morphologically permanent. Ductal and lobular-alveolar development of the mammary gland during the postnatal period is characterized by hormonally controlled morphological changes involving apoptosis. Therefore, histological evaluation of mammary gland tissues may be more sensitive and accurate in assessing the developmental status of mammary glands, as compared to gross examination for retention of nipples during the postnatal period. (OEHHA, 2007).

In the study by Lehmann et al. (2004), testicular testosterone concentrations were significantly lower in male fetuses from dams exposed to DBP at 50 mg/kg-day and higher doses during gestation, compared to controls. Testicular testosterone concentration is an endpoint indicative of

a decrease in testosterone production (a functional change) in the testis (Lehmann et al., 2004)(Lehmann et al., 2004).

Under Proposition 65, OEHHA developed a Maximum Allowable Dose Level (MADL) of 8.7 µg/d for DBP as a reproductive toxicant based on a study by Lee et al. (2004). This study and the derived MADL will be used as the basis for the 8-hr REL after adjustment for inhalation exposure. Lee et al. (2004) examined five groups of pregnant IGS (Sprague-Dawley) rats, 6-8 dams per group, were treated with 0, 20, 200, 2,000, or 10,000 ppm DBP in soy-free diet from GD 15 to postnatal (PND) 21. The lowest dose of 20 ppm of DBP in a soy-free diet was estimated by the study authors to be equivalent to 1.5-3.0 mg/kg-day. At this dose, there was no difference in reproductive organ weights or ano-genital distance (AGD) in male pups between treated and control groups. However, 4% of pups retained areolas or nipples on PND 14, as compared to 0% in the control, although the increase was not statistically significant. When the offspring were examined on PND 21 (the last day of treatment), a lower number of germ cells in the seminiferous epithelium and histopathological changes in the mammary gland were observed in 4 of 8 male pups examined (as compared to none of 8 pups in the control group, $p<0.05$). In female pups, 4 of 8 examined at this time point had hypoplasia of the alveolar bud in the mammary gland ($p<0.05$). These findings suggest that DBP at 20 ppm in diet altered testicular and mammary gland development in male pups, and affected mammary gland development in female pups. These results are consistent with the findings of the Mychereest et al. (2000)) that DBP causes alterations in nipple development and Lehmann et al. (2004) that showed decreased testosterone production in males at very low doses. Therefore 20 ppm, was estimated by the Lee et al. (2004) study to be the LOAEL for the male and female reproductive toxicity.

Based on food consumption and average body weights of dams during the treatment period, the authors estimated that the dams in the five groups were exposed to 0, 1.5-3.0, 14.4-28.5, 148.2-290.9, and 712.3-1371.8 mg/kg-day of DBP for the control, 20, 200, 2,000, and 10,000 ppm groups, respectively. The average (mean) intake of DBP between PND 10 and 21 in each group was the highest and was approximately two-fold higher than that between GD 15-20, due to increased food consumption in the presence of reduced body weight gains for the period of PND 10-21 in each group.

In summary, DBP at 10,000 ppm in the diet from GD 15 to PND 21 caused: reduced number of male pups per litter, decreased AGD, reduced testicular weights, reduced germ cells and delayed germ cell development in the testis, increased number of male pups with retained nipples, and abnormal development of the mammary gland. Some of these effects were also observed in animals in the 20-, 200- or 2,000-ppm groups. At 20 ppm, a significantly increased number of animals had a reduced number of spermatocytes, clearly indicating the effect of DBP on the testicular development at this dose level (Lee et al., 2004). In addition to nipple retention, Lee et al. (2004) also reported dramatic histopathological changes in the mammary gland in male offspring in all DBP-treated groups at PND 21 and PNW 11. At PND 21, the day of treatment termination, the authors observed histopathological changes indicative of growth or development of mammary gland in males. Inhibition of testosterone production or action by treatment with anti-androgenic compounds during the late gestational period in rats causes development of breast tissues (OEHHA, 2007).

In addition to the male reproductive effects, Lee et al. (2004) found increased incidence of hypoplasia of alveolar bud in the developing mammary gland in female pups on PND 21. Compared to the zero incidence in control group, 4, 3, 4, and 4 out of 8 female pups examined for each group (20-, 200-, 2,000-, and 10,000-ppm, respectively) had this abnormal histopathological change in the mammary gland. Even with the small group sizes, the increases were statistically significant at 20-, 2,000-, and 10,000-ppm group, but not in the 200-ppm group. Thus, histopathological changes in the mammary gland in female offspring exposed perinatally to DBP at concentrations of $\geq$ 20 ppm in diets indicate female reproductive effects of DBP at these doses.

To summarize, both male and female reproductive effects were observed in animals treated with 20 ppm of DBP in diet, the lowest dose used in the study by Lee et al. (2004). The male reproductive effects at this dose are manifested as increased number of male pups with reduced germ cell in the testis on PND 21, increased percentage of male pups with retained nipples on PND 14, increased number of male pups with dilatation of alveolar bud in the mammary gland on PND 21, increased numbers of male pups with degeneration of alveolar cells and alveolar atrophy in the mammary gland on PNW 11 and 20. All these DBP and thus are biologically significant. For the female reproductive effects, perinatal exposure to DBP at 20 ppm in diet caused apparent histopathological changes in the mammary gland of the female pups that are consistent with other reported female reproductive toxicity of DBP in female rats. Therefore, 20 ppm in diet, the lowest dose used in the study by Lee et al. (2004), is considered to be a LOEL. The reproductive effects of DBP as observed in the study by Lee et al. (2004) were the consequence of maternal exposures. This dose, equivalent to 1.5, 2.4, or 3.0 mg/kg-day of DBP to the dam for the periods of GD 15-20, PND 2-10, or PND 10-21, respectively, is considered as the LOAEL for the reproductive effects observed in this study. The estimated dose for the gestational period, 1.5 mg/kg-day, is the lowest LOEL for the developmental, male, and female reproductive toxicity of DBP among all the relevant studies that OEHHA has reviewed for establishing an acute REL for DBP. The study by Lee et al. (2004) is thus identified as "the most sensitive study deemed to be of sufficient quality" and will be used as the basis for acute REL calculation.

6.0 Derivation of Interim Acute REL (1-hour exposure)

No studies of short-term exposure to DBP were located that were appropriate for the derivation of an acute REL. While an LC_{50} was reported, this value represents the upper limit for acute exposures that are compatible with survival without regard to protecting health. As such the LC_{50}values are not the preferred basis for the derivation of an acute REL, which requires consideration of effects much less severe than lethality.

In the course of an 8-hour exposure, intermittent spikes in exposure levels are included in the time-weighted average addressed with the 8-hr REL. The values associated with 8-hr RELs are typically lower than allowed for acute 1-hr exposures, due to the longer exposure duration and possibility of recurring exposures. Therefore application of the 8-hr REL to exposure scenarios involving short-term peaks in concentration should be health protective in most cases.

Derivation of Interim 8-hour REL

Study	Lee et al., 2004
Study population	Sprague-Dawley rats
Exposure method	Oral in feed
Exposure continuity	
Exposure duration	In diet from GD 15 to PND 21
Critical effects	Histopathological changes in the testis and mammary glands.
LOAEL	1.5 mg/kg-day
NOAEL	0.15 mg/kg-day
LOAEL uncertainty factor (UF_L)	(10)
Convert NOEL for 58 kg woman	0.15 mg/kg-day x 58 kg = 8.7 mg/day
Cumulative uncertainty factor	1,000
MADL adult-oral	8.7 ug/day (8.7 mg/day / 1000)
Route to route extrapolation	0.4 mg/m^3(8.7 mg/day/20 m^3/d)
Chronic to 8-h adjustment	(6/8 * 5/7)
Reference Exposure Level	**0.23 ug/m^3**

The 8-hour Reference Exposure Level is a concentration at or below which adverse noncancer health effects would not be anticipated for repeated 8-hour exposures. Inhalation studies were not available, so this draft 8-hour REL is derived from a feeding study by Lee et al (2004) for which OEHHA previously estimated a maximum allowable daily dose (MADL).of 8.7 µg/day In that study, Sprague-Dawley rats were exposed to 0, 20, 200, 2,000, or 10,000 ppm DBP in soy-free diet from GD 15 to postnatal (PND) 21. The lowest dose of 20 ppm of DBP in a soy-free diet was estimated by the study authors to be equivalent to 1.5-3.0 mg/kg-day. The animal study has a systemic endpoint of histopathological changes in the testis and mammary glands of male and female rats, respectively.

A LOAEL uncertainty factor of 10 was used because a NOAEL was not determined for the study. The time adjustment for the 8-hour REL used was 6/8 hours * 5/7 days/week. The NOAEL is converted to a milligram per day dose level by multiplying the assumed female human body weight (female = 58 g) and dividing by the route adjustment (oral to inhalation 20 m^3/d). A cumulative UF of 1000 was used in determining the oral MADL.

7 Environmental fate

DBP may be released into the environment during its production and subsequent life cycle stages, including disposal. Emissions to water and air are expected to be the most important entry routes of DBP. General characteristics of DBP which are relevant for the exposure assessment are discussed in the following subparagraphs.

The Henry's law constant of $1.81x10^{-6}$ m^3/mol indicates that DBP will only slowly volatilize from surface waters, i.e. virtually all of the DBP will remain in the water phase at equilibrium. The octanol/water partition coefficient (K_{ow}) of DBP is high and consequently the equilibrium between water and organic carbon in soil or sediment will be very much in favour of the soil or sediment. Soil and sediment thus appear to be important sinks for DBP. Resuspension of DBP from the sediment to the water column may occur. Although DBP is only poorly soluble in water, it may be transported in water following the adsorption of DBP to humic substances. Despite its low volatility, DBP has been reported as particulate and as a vapour in the atmosphere. In the air DBP is transported and removed by both wet and dry deposition. Photooxidation by OH radicals contributes to the elimination of DBP from the atmosphere with a half-life of 42 hr. The high K_{ow} of DBP indicates that the substance has a potential for bioaccumulation (ATSDR).

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