UNITED STATES
CONSUMER PRODUCT SAFETY COMMISSION
Bethesda, MD 20814

Memorandum

Date: October 30, 2010

TO : Michael A. Babich, Ph.D., Project Manager, Phthalates, Section 108 of CPSIA

THROUGH: Mary Ann Danello, Ph.D., Associate Executive Director, Directorate for Health Sciences
Lori E. Saltzman, M.S., Director, Division of Health Sciences

FROM : Kent R. Carlson, Ph.D., Toxicologist, Directorate for Health Sciences
Leslie E. Patton, Ph.D., Toxicologist, Directorate for Health Sciences

SUBJECT : Toxicity Review of **Diisoheptyl phthalate (DiHP)**

The following memo provides the Versar Inc. and SRC, Inc. contractor's and U.S. Consumer Product Safety Commission's (CPSC's) Health Sciences staff assessment of the potential toxicity associated with **DiHP**.

CPSC staff assesses a product's potential health effects to consumers under the Federal Hazardous Substances Act (FHSA). The FHSA is risk-based. To be considered a "hazardous substance" under the FHSA, a consumer product must satisfy a two-part definition. First, it must be "toxic" under the FHSA, or present one of the other hazards enumerated in the statute. Second, it must have the potential to cause "substantial illness or injury during or as a result of reasonably foreseeable handling or use." Therefore, exposure and risk must be considered in addition to toxicity when assessing potential hazards under the FHSA (CPSC, 1992; summarized at 16 CFR 1500.135)

The FHSA addresses both acute and chronic hazards. While the FHSA does not require manufacturers to perform any specific battery of toxicological tests to assess the potential risk of chronic health hazards, the manufacturer is required to label a product appropriately according to the requirements of the FHSA. The first step in the risk assessment process is hazard

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identification, that is, a review of the available toxicity data for the chemical under consideration and a determination of whether the chemical is considered “toxic”. Chronic toxicity data (including carcinogenicity, neurotoxicity, and reproductive and developmental toxicity) are assessed by the CPSC staff using guidelines issued by the Commission (CPSC, 1992). If it is concluded that a substance is “toxic” due to chronic toxicity, then a quantitative assessment of exposure and risk is performed to evaluate whether the chemical may be considered a “hazardous substance”. This memo represents the first step in the risk assessment process; that is, the hazard identification step.

FINAL

TOXICITY REVIEW FOR DIISOHEPTYL PHTHALATE (DiHP, CASRN 71888-89-6)

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Prepared by:

Versar Inc.
6850 Versar Center
Springfield, VA 22151

and

SRC, Inc.
7502 Round Pond Road
North Syracuse, NY 13212

Prepared for:

Kent R. Carlson, Ph.D.
U.S. Consumer Product Safety Commission
4330 East West Highway
Bethesda, MD 20814

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LIST OF ABBREVIATIONS AND ACRONYMS

AGD	Anogenital distance
DiHP	Diisoheptyl phthalate
DNA	Deoxyribonucleic acid
ER	Estrogen receptor
GD	Gestation day
HRIPT	Human Repeated Insult Patch Test
LOAEL	Lowest-observed-adverse-effect level
NOAEL	No-observed-adverse-effect level
NTR	Non-treatment recovery
PBOX	Peroxisomal beta-oxidation
PND	Postnatal day
SD	Standard deviation
SE	Standard error

EXECUTIVE SUMMARY

This report is an update of a report previously prepared by CPSC (Patton, 2010).

DiHP is a moderate use plasticizer found in a variety of consumer products.

Exposure to DiHP resulted in an oral LD_{50} > 10,000 mg/kg in a rat study and a dermal LD_{50} > 3160 mg/kg in a rabbit study. No dermal irritation or sensitization was noted in one well-described human patch test study. In contrast, two rabbit studies described slight dermal erythema, and one guinea pig study weak sensitization following exposure. Weak conjunctival irritation was also reported in two rabbit studies. Insufficient data were available to make the determination of whether DiHP was associated with acute inhalation toxicity.

Evidence also supported the conclusion that DiHP was a chronic toxicant. Exposure to DiHP increased liver weight and pathologies, kidney weight and pathologies, pituitary weight, changed reproductive parameters (resorptions, post implantation loss, mean number of live fetuses, fetal implantations, reproductive organ weights and pathologies, sperm production and content), and changed developmental parameters (fetal weights, external visceral, and skeletal malformations) in developmental toxicity or 2 generation reproductive studies.

Acceptable daily intakes values (ADI's) are calculated when a given chemical is considered "toxic" and sufficient toxicity information is available. The ADI is the amount of a chemical that one may be exposed to on a daily basis without posing a significant risk of health effects to consumers.

ADI's were not estimated for DiHP relevant exposure durations for the general population or for other sensitive subpopulations because confirmatory data (additional studies) on toxicological endpoints were not available.

TOXICITY REVIEW FOR DIISOHEPTYL PHTHALATE
(DiHP CASRN 71888-89-6)

1. INTRODUCTION

This report summarizes available data for the identity, physicochemical properties, manufacture, supply, use, toxicity, and exposure information on diisoheptyl phthalate (DiHP). This assessment was prepared from a variety of review articles (NICNAS, 2008; EPA, 2010; ECB, 2006) as well as supplemental independent studies retrieved from literature searching.

Historically, concerns regarding most phthalates have been primarily associated with their potential to induce adverse reproductive/developmental effects in humans (NICNAS, 2008). The structural and physicochemical properties of certain phthalates that allow migration and leaching out of products, especially in soft plastics, have also been a concern (NICNAS, 2008).

2. IDENTITY and PHYSICOCHEMICAL CHARACTERISTICS

This section highlights the identity and key physicochemical properties of DiHP. DiHP is predominately comprised of a pair of 6-carbon esters linked to a benzene-dicarboxylic acid ring. The branched ester side chains are in an *ortho* configuration, in contrast to those found in isophthalates (*meta*) or terephthalates (*para*). DiHP is currently considered to belong to the Transitional Phthalate Esters group.

The identity and physicochemical properties of DiHP can be seen in Tables 2.1 and 2.2 (NICNAS, 2008; U.S. EPA, 2010; ECB, 2006).

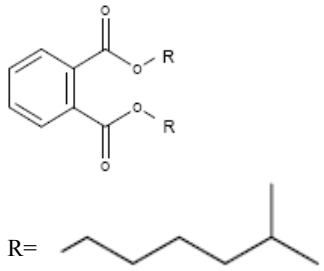
Table 2.1 Names, Structural Descriptors, and Molecular Formulas of DiHP (NICNAS, 2008)	
CAS Number:	71888-89-6
Chemical Name:	1,2-Benzenedicarboxylic acid, di-C6-8-branched alkyl esters, C7 rich
Common Name:	Diisoheptyl phthalate (DiHP)
Molecular Formula:	C22H34O4
Structural Formula:	
Molecular Weight:	363 (based on a di-C_7H_{15} alkyl ester)
Synonyms:	DiHP; Diisoheptyl phthalate ester; 1,2-Benzenedicarboxylic acid, diisoheptyl ester
Purity/Impurities/Additives:	Purity: >99.9% w/w; Impurities: ≤0.1% w/w, including isoheptyl alcohol (0.03%), diisoheptyl ether and isoheptyl benzoate (0.07%); Additives: none

Table 2.2 Physicochemical Properties of DiHP	
Property	**Value**
Physical state	Liquid (NICNAS, 2008; U.S. EPA, 2010)
Melting point	-45°C(NICNAS, 2008; U.S.EPA, 2010)
Boiling point	398°C (101.3 kPa; NICNAS, 2008); 393.5°C (101.3 kPa; ECB, 2006)
Density	994 kg/m3 (20°C; NICNAS, 2008)
Vapor pressure	9.33×10^{-8} kPa (25°C; NICNAS, 2008); 1.2×10^{-5} (25°C; U.S.EPA, 2010)
Water solubility	1.7×10^{-5} g/L (22°C; NICNAS, 2008); 0.01 mg/L (25°C; U.S. EPA, 2010)
Partition coefficient n-octanol/water (log Kow)	6.87 (NICNAS, 2008; U.S. EPA, 2010)
Henry's law constant	1.99 Pa-m^3/mole (25°C; U.S.EPA, 2010)
Flash point	>190ºC (Patton, 2010)

3. MANUFACTURE, SUPPLY, AND USE

Manufacture

In general, DiHP is manufactured commercially in a closed system by catalytically esterifying phthalic anhydride with various alcohols (isoheptyl or heptanol mixtures). As with other phthalates, the unreacted alcohols are recovered and reused, and the DiHP mixture is purified by vacuum distillation or activated charcoal. The purity of DiHP can achieve 99% or greater using current manufacturing processes (NICNAS 2008). The remaining fraction of the DiHP commercial mixture can also contain impurities such as isoheptyl alcohol (0.03 wt%), diisoheptyl ether and isoheptyl benzoate (0.07 wt%) (NICNAS, 2008), and a maximum of 0.1% water. The NTP has also suggested that DnHP may comprise as much as 25% of commercial mixtures of diisohexyl phthalate (CERHR, 2003). DiHP has previously been manufactured by ExxonMobil Chemical Company in the U.S. under the brand name Jayflex 77 and is currently listed as a product on Univar USA's website.

Recent information reported by the European Chemical Agency (ECHA) suggests that the manufacture of DiHP has ceased in the European Union (EU) and United States (U.S.) as of the end of 2010 (ECHA, 2011). The European Chemical Agency was unable to confirm whether DiHP is currently manufactured in China.

Supply

Production of diisoheptyl and diisohexyl phthalate peaked in the mid-/late-1990's at 48,000 metric tons and has subsequently declined. Recently, U.S. production of DiHP has decreased from 26,000 metric tons (2005) to 22,700 metric tons (2008) and is projected to decrease further to negligible levels (2013). DiHP's proportion of the total phthalate production market (3.9%) is also projected to decrease to negligible levels during the same period (Bizzari et al. 2007, 2009). The EPA IUR reports that the aggregated national production volume for DiHP was from 50 to < 100 million pounds during the reporting period (2006).

As with many plasticisers, U.S. consumption of DiHP has paralleled production estimates. Current consumption of DiHP has been reported as 22,000 metric tons (2008) and is projected to decrease to negligible amounts (2013). DiHP's proportion of the total phthalate consumption market (3.7%) is also projected to decrease to negligible amounts (Bizzari et al. 2009).

In the past 20 years, U.S. consumption (in metric tons) of DiHP has been within a metric ton or two less than production estimates. This suggests that most DiHP produced in the U.S. was utilized locally.

Production and consumption estimates for other countries are sparse. Bizzari et al. (2009) reported that the production of DiHP ceased in Japan in 2000, following the discontinuation of isoheptyl alcohol production (Kyowa Hakko). Reports of DiHP production and consumption in other countries were not located.

Use

Transitional phthalates are used as a primary or secondary plasticizer in industrial chemicals that are associated with polymers to impart flexibility in polyvinyl chloride (PVC) resins. DiHP is no longer manufactured in the EU and U.S., but it may still be used in limited quantities. NICNAS (2008) reported that in Australia, DiHP is used as a specialist PVC plasticizer and in screen printing inks. DiHP also is used in vinyl flooring, tile, carpet backing, molding and coating plastisols, and partial replacement for low molecular weight plasticizer, such as DEHP (ECHA, 2011). DiHP is a general purpose, strongly solvating, highly compatible phthalate plasticizer which processed faster than DOP (Exxon Mobil, 2003). DiHP may also be present in lubricating oils.

4. TOXICOKINETICS

No data examining the absorption, distribution, metabolism, or excretion of DiHP were located.

5. HAZARD INFORMATION

This section contains brief hazard summaries of the adverse effects of DiHP in a variety of animal and bacterial species. More detailed discussions of the studies can be viewed in the Appendices. When evaluating hazard study data, Consumer Product Safety Commission (CPSC) staff utilized the definitions for toxicity as presented in regulations (16 CFR §1500.3(c)(2)(ii)) and the chronic hazard guidelines (16 CFR §1500.135) in the Federal Hazardous Substances Act (FHSA; 15 U.S.C. 1261-1278). When considering the FHSA, substances that are "known" or "probable" toxicants are "toxic" and substances that are considered "possible" toxicants are "not toxic" (Table 5.1).

Table 5.1. Classification of Chronic Hazards (as per the FHSA)

Evidence	Human Studies	Animal Studies
Sufficient evidence	**Known**	**Probable**
Limited evidence	**Probable**	Possible
Inadequate evidence	Possible	—

Exposure to DiHP resulted in an oral LD_{50} > 10,000 mg/kg in a rat study and a dermal LD_{50} > 3160 mg/kg in a rabbit study. No dermal irritation or sensitization was noted in one well-described human patch test study. In contrast, two rabbit studies described slight dermal erythema, and one guinea pig study weak sensitization following exposure. Weak conjunctival irritation was also reported in two rabbit studies. Insufficient data were available to make the determination of whether DiHP was associated with acute inhalation toxicity.

Evidence supported the conclusion that DiHP was a chronic toxicant. Exposure to DiHP increased liver weight and pathologies, kidney weight and pathologies, pituitary weight, changed reproductive parameters (resorptions, post implantation loss, mean number of live fetuses, fetal implantations, reproductive organ weights and pathologies, sperm production and content), and changed developmental parameters (fetal weights, external visceral, and skeletal malformations) in developmental toxicity or two generation reproductive toxicity studies.

Acceptable daily intakes values (ADI's) are calculated when a given chemical is considered "toxic" and sufficient toxicity information is available. The ADI is the amount of a chemical that one may be exposed to on a daily basis without posing a significant risk of health effects to consumers. ADI's were not estimated for DiHP relevant exposure durations for the

general population or for other sensitive subpopulations because confirmatory data (additional studies) on toxicological endpoints were not available.

In the following discussions, hazard information was divided into sections thought to be of interest for regulatory matters (i.e., for labeling and other mitigation measures) as well as for biological and pathological consistency. More specifically, hazards were divided into whether the exposure was singular or repeated. Hazards associated with repeated exposures were further divided into groupings based on the affected organ system (i.e., hepatic, neurological, hematologic, etc.) and discussed in terms of the exposure duration if sufficient information existed to do so (***acute***, ≤14 days; ***intermediate-term*** or ***subchronic***, 15–364 days; ***long-term*** or ***chronic***, ≥365 days; and ***multigenerational***; ATSDR, 2007) where appropriate. Discrete study information can be reviewed in the Appendices.

ACUTE DOSE TOXICITY

5.1. Acute Oral Toxicity

The acute oral LD_{50} for DiHP in male Wistar rats is >10,000 mg/kg (Exxon Chemical Americas, 1979a). Rats from all treatment groups (1,000, 1,470, 2,150, 3,160, 4,640, 6,810, or 10,000 mg/kg-day; 5/group) showed clinical signs of toxicity, including lethargy, diarrhea, piloerection, chromorhinorrhea, chromodacryorrhea, and ptosis soon after dosing, but were generally healthy after day 5 of the 14-day observation period. No animals died from dosing in this study. At necropsy, there were no abnormal findings. Bio/Dynamics (1980; as cited in European Commission, 2000) also identified an acute oral LD_{50} value of >10,000 mg/kg in rats. No further information is available on this study.

The estimated LD_{50}s are higher than the oral LD_{50} range (50 to 5,000 mg/kg) required by the FHSA to conclude that a chemical is acutely toxic. The weight of evidence including probable animal data are sufficient, therefore, to support the conclusion that DiHP does not fit the definition of "acutely toxic" via oral exposure under the FHSA (16 CFR §1500.3(c)(2)(i)(A)).

5.2. Acute Dermal Toxicity

No mortality occurred in New Zealand White rabbits (2/sex) exposed to 3,160 mg/kg DiHP (apparently undiluted) on clipped, abraded skin under occluded conditions for 24 hours

and observed for 14 days after dosing (Exxon Chemical Americas, 1979b). No control group was used. Clinical signs of toxicity, including lethargy, diarrhea, and ptosis, were observed sporadically and may not have been related to treatment. All rabbits showed dilated hearts at necropsy.

Exxon Chemical Americas (1980a) reported no mortality among New Zealand White rabbits (2 sex/dose) exposed to undiluted DiHP on clipped, intact skin at 50, 200, 794, or 3,160 mg/kg for 24 hours under occluded conditions and observed for 14 days. Treated rats exhibited fecal staining (at ≥200 mg/kg) and nasal discharge (all doses). However, no treatment-related findings were reported at necropsy.

The estimated LD_{50}s from the studies above are higher than the dermal LD_{50} range (200 to 2,000 mg/kg) required by the FHSA to conclude that a chemical is acutely toxic. The weight of evidence including probable animal data are sufficient, therefore, to support the conclusion that DiHP does not fit the definition of "acutely toxic" via dermal exposure under the FHSA (16 CFR §1500.3(c)(2)(i)(C)).

5.3. Acute Inhalation Toxicity

No information was located regarding the acute inhalation toxicity of DiHP.

The lack of acute inhalation toxicity data for DiHP can be considered a data gap and supports the conclusion that there is "inadequate evidence" for the designation of DiHP as "acutely toxic" (< 200 mg/L) via inhalation under the FHSA (16 CFR §1500.3(c)(2)(i)(B)).

5.4. Primary Skin Irritation

In human subjects (14 females and 1 male) exposed to an unspecified amount of undiluted DiHP (>99% pure) for 24 hours under occluded conditions, no significant skin irritation was observed 30 minutes or 24 hours after patch removal (Medeiros et al., 1999). In a subsequent Human Repeated Insult Patch Test (HRIPT), application of 0.2 mL of undiluted DiHP (>99% pure) under occlusion for 24 hours 3 times/week for 3 weeks did not induce skin irritation (scored 24 and 48 hours after each patch removal) in 104 human subjects (Medeiros et al., 1999).

Slight erythema without edema was observed at 24 hours, but not subsequently, in three of four New Zealand White rabbits (two per sex; no control group) exposed to 3,160 mg/kg DiHP (undiluted) on clipped, abraded skin for 24 hours under occluded conditions and observed for 14 days (Exxon Chemical Americas, 1979b).

The intensity of skin irritation responses increased in a dose-related manner in New Zealand White rabbits (2 sex/group) exposed to undiluted DiHP on clipped, intact skin at 50, 200, 794, or 3,160 mg/kg for 24 hours under occluded conditions and observed for 14 days (Exxon Chemical Americas, 1980a). Slight erythema was noted in two of four animals exposed to 200 mg/kg and in all animals exposed to 794 mg/kg. Erythema was well-defined and accompanied by edema in all four animals exposed to 3,160 mg/kg.

No skin irritation was reported in humans following dermal exposure to DiHP in two studies. Slight to well-defined erythema accompanied by edema was noted in rabbits following exposure to DiHP in other studies. "Scores" for the rabbit studies were not able to be estimated, so could not be compared to the threshold for defining a skin irritant in the FHSA (16 CFR §1500.3(c)(4)).

The weight of evidence including sufficient human and animal data, therefore, supported the conclusion that DiHP did not fit the definition of a "primary irritant" or "corrosive" as outlined in the FHSA (16 CFR §1500.3(c)(4); 16 CFR §1500.3(c)(3)).

5.5. Primary Eye Irritation

One drop of undiluted DiHP applied to the conjunctival sac of six rabbits (four males and two females) elicited a positive conjunctival reaction in two rabbits 1 hour after treatment (Exxon Chemical Americas, 1979c). No irritation of the cornea, iris, or conjunctiva was observed at subsequent time points. Exxon Chemical Americas (1980b) reported that four of six rabbits administered undiluted DiHP showed conjunctival redness 1 and 4 hours following treatment; two of these animals also scored positive for iridial irritation at the same time points. No signs of eye irritation were evident after 72 hours.

The weight of evidence including sufficient animal data supported the conclusion that DiHP did not fit the definition of an ocular "corrosive" or "primary irritant" as outlined in the FHSA (16 CFR §1500.3(c)(4) and 16 CFR §1500.3(c)(3), respectively).

5.6. Sensitization

In the HRIPT described in Section 5.4 above, human subjects challenged with DiHP at a naïve application site 10–17 days following the last previous application of DiHP showed no evidence of a skin sensitization response (Medeiros et al., 1999).

A weak skin sensitization response was reported in guinea pigs during re-challenge (but not initial challenge) with DiHP (Exxon Biomedical Sciences, 1991; as cited in NICNAS, 2008 and European Commission, 2000). In another study, DiHP did not elicit a skin sensitization response in guinea pigs (Huntingdon Research Centre, 1994; as cited in NICNAS, 2008). No further information was available.

Sufficient human and animal data support the conclusion that DiHP did not fit the definition of a dermal "sensitizer" as defined in the FHSA (16 CFR §1500.3(c)(5)).

REPEAT DOSE TOXICITY

5.7. General Effects (Clinical Signs, Food/Water Consumption, Body Weight)

In a dose range-finding developmental toxicity study where pregnant Crl:CDBR rats were administered DiHP via gavage at up to 1,000 mg/kg-day on gestation days (GDs) 6–20 (Exxon Chemical Americas, 1997, 1996), no mortality or treatment-related clinical signs of toxicity were observed in treated dams. Relative to controls, body weights were generally lower in rats treated at ≥750 mg/kg-day; by GD 21, body weights were reduced by 8 and 14% at 750 and 1,000 mg/kg-day, respectively (Table 5.2). With respect to the entire gestation period (days 0–21), body weight gain was also decreased significantly at 750 and 1,000 mg/kg-day (17 and 32% lower than controls). Body weights adjusted for gravid uterus weight were similar to controls for all treatment groups, suggesting that observed reductions in body weight, particularly toward the end of gestation, were likely associated with decreased uterine weights. Food consumption was similar among treatment and control groups, and no treatment-related findings were reported at necropsy.

Table 5.2. Body Weights of Crl:CDBR Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)				
	0	250	500	750	1,000
Maternal data					
Number pregnant animals	7	7	6	6	7
Body weight gain (g) GDs 0–21	204.0 ± 14.7[a]	213.7 ± 12.2	221.3 ± 17.1	169.3 ± 24.2[b]	138.9 ± 46.8[b]
Body weight, GD 21 (g)	476.6 ± 26.4	487.1 ± 19.0	494.3 ± 23.8	440.3 ± 27.8[b]	411.1 ± 53.5[b]
Body weight adjusted for gravid uterus weight (g)	365.4 ± 20.3	371.1 ± 15.6	378.5 ± 19.7	364.2 ± 28.0	357.4 ± 23.3

[a]Mean ± standard deviation (SD).
[b]Significantly different from controls at $p < 0.05$ based on t-test performed for this review.

Sources: Exxon Chemical Americas (1997, 1996).

In the subsequent full developmental toxicity study, (McKee et al., 2006; Exxon Chemical Company, 1997), pregnant rats (25/group) were administered DiHP on GDs 6–20 via gavage at up to 750 mg/kg-day. With the exception of two dams that gave birth before scheduled sacrifice and were euthanized (one from the control group and one from the 300 mg/kg-day group), all animals survived to study termination. No treatment-related clinical signs of toxicity were reported, and no changes in food consumption were detected in treated

rats. Although the decrease (7%) in mean maternal body weight of females treated at 750 mg/kg-day was statistically significant, adjustment for gravid uterine weight resulted in a mean maternal body weight that was similar among all treatment groups (Table 5.3). The difference in terminal body weights observed for control and high-dose rats was due to a mean reduction in uterine weight of 30% in the high-dose group relative to controls.

Table 5.3. Body Weights of Crl:CDBR VAF/Plus Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)			
	0	100	300	750
Maternal data				
Number pregnant animals	23	21	22	23
Mean maternal body weight, day 21 (g)	441.6 ± 27.4[a]	440.5 ± 32.0	441.0 ± 30.9	412.2 ± 31.6[b]
Body weight adjusted for gravid uterus weight (g)	332.5 ± 26.6	334.9 ± 26.6	337.5 ± 21.4	335.7 ± 24.8

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$.

Sources: McKee et al. (2006); Exxon Chemical Company (1997).

In a two-generation reproductive toxicity study (McKee et al., 2006; ExxonMobil Chemical Company, 2003), where groups of male and female Crl:CD(SD) IGS BR rats (30 sex/group; 6 weeks old at study initiation) were exposed to DiHP at 0, 1,000, 4,500, or 8,000 ppm in the diet, mortality occurred in F0 parental animals and in F1 animals post-weaning (one control male, one low-dose female, one mid-dose male, four high-dose males, and two males in the non-treatment recovery [NTR] group), but was not definitively associated with treatment. No clinical signs of toxicity were observed in F1 animals; signs in F2 animals were limited to effects on external genitalia in males of the 8,000 ppm group, and included observations of hypospadias (7/30 males) and absent (2/30 males) or undescended testes (2/30 males). With the exception of 1 male (of 30) in the 1,000 ppm group, which also had an undescended testis, clinical signs of toxicity were not observed in any other treatment group. Body weights and body weight gains were similar among treatment and control groups during pre-mating (F0 and F1 animals), and gestation and lactation (F0 generation); however, the body weights of high-dose F1 females were reportedly decreased significantly ($p < 0.05$) throughout gestation and lactation. Based on visual inspection of the data (presented graphically), body weights for females treated at 8,000 ppm appeared to stay within ~10% of controls during gestation; data for body weights during lactation were not shown.

5.8. Hepatotoxicity

In the developmental toxicity study conducted by Exxon Chemical Company (1997) and published in McKee et al. (2006) where dams were treated with DiHP via gavage on GDs 6–20, significant increases in absolute and relative liver weights were reported at ≥300 mg/kg-day (Table 5.4).

Table 5.4. Liver Weights in Crl:CDBR VAF/Plus Rats Administered DiHP Via Gavage on GDs 6–20.

Endpoint	Dose (mg/kg-day)			
	0	**100**	**300**	**750**
Number pregnant animals	23	21	22	23
Absolute liver weight (g)	16.0 ± 2.2[a]	17.2 ± 2.0	17.8 ± 1.9[b]	19.2 ± 1.9[b]
Relative liver weight	0.049 ± 0.005	0.051 ± 0.004	0.053 ± 0.004[b]	0.057 ± 0.004[b]

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$.

Sources: McKee et al. (2006); Exxon Chemical Company (1997).

In the two-generation reproductive toxicity study (McKee et al., 2006; ExxonMobil Chemical Company, 2003), relative liver weight was significantly increased (by 10–23%) at necropsy in F0 rats exposed to DiHP at ≥4,500 ppm compared with controls (Table 5.5). Similarly, relative liver weight was significantly increased in adult F1 males at 8,000 ppm (9%), and in adult F1 females at ≥4,500 ppm (15–19%). Histopathological examinations of the liver revealed minimal centrilobular hypertrophy in F1 males (4,500 and 8,000 ppm groups) and females (8,000 ppm group; incidence data not reported). Hepatocellular vacuolization (both generations) and necrosis (F1 generation) were noted in the livers of males (only) exposed to DiHP at the high dose (data not shown).

Table 5.5. Liver Weights in Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet Over Two Generations				
	Dietary Level (ppm)			
Endpoint	0	1,000	4,500	8,000
Relative liver weight; F0 adults				
Males	3.55 ± 0.28 (29)[a]	3.60 ± 0.32 (30)	3.92 ± 0.32 (30)[b]	4.15 ± 0.30 (28)[b]
Females	4.02 ± 0.34 (28)	4.13 ± 0.21 (27)	4.50 ± 0.33 (25)[b]	4.96 ± 0.29 (28)[b]
Relative liver weight; F1 adults				
Males	3.87 ± 0.40 (30)	3.76 ± 0.39 (30)	4.05 ± 0.32 (29)	4.23 ± 0.29 (28)[b]
Females	3.70 ± 0.37 (30)	3.92 ± 0.33 (30)	4.27 ± 0.36 (29)[b]	4.42 ± 0.52 (30)[b]

[a]Mean ± SD. Number of animals is indicated in parentheses.
[b]Significantly different from controls at $p < 0.05$.

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

Relative liver weight was significantly increased (by about 18–33% based on data presented graphically) in male F344 rats (5/group) treated with DiHP in the diet at 12,000 ppm (~1,200 mg/kg-day) for 2 or 4 weeks (Smith et al., 2000). A small (but statistically significant) increase in liver weight was also observed in rats treated at 1,000 ppm (~100 mg/kg-day) for 2 (but not 4) weeks. Relative liver weights were similar among treated mice (B6C3F1 males exposed at up to 6,000 ppm, or approximately 1,100 mg/kg-day) and controls. Peroxisomal beta-oxidation (PBOX) activity (a marker of peroxisome proliferation) was increased significantly (approximately 3–5-fold) in the livers of rats and mice treated with DiHP at the high dose (12,000 and 6,000 ppm for rats and mice, respectively) for up to 4 weeks. Although gap junctional intercellular communication was not affected by treatment, rates of hepatocellular replicative deoxyribonucleic acid (DNA) synthesis increased significantly (by approximately 30–40%) in rats exposed to DiHP at ≥1,000 ppm (~100 mg/kg-day) and in mice exposed to DiHP at ≥500 ppm (~90 mg/kg-day) for 2 or 4 weeks.

The weight of evidence from the above studies supported the conclusion that there was "sufficient animal evidence" for the designation of DiHP as a "hepatotoxicant."

5.9. Renal Toxicity

At necropsy in the two-generation reproductive toxicity feeding study (McKee et al., 2006; ExxonMobil Chemical Company, 2003), relative kidney weight was significantly increased (7–15%) in F0 rats exposed to DiHP at ≥4,500 ppm compared with controls (Table 5.6). In F1 adults, relative kidney weight was increased 9% in males exposed to DiHP at

≥4,500 ppm. Histopathological examinations of the kidney showed a significantly increased incidence of chronic progressive nephropathy in F0 and F1 males treated at 8,000 ppm; the study authors suggested that the onset of this disorder was magnified by DiHP treatment. Hydronephrosis (associated with observations of dilated renal pelvis) was also noted in F1 males exposed to DiHP at 4,500 or 8,000 ppm (incidence data not reported).

The weight of evidence from the above studies supported the conclusion that there was "limited animal evidence" for the designation of DiHP as a "renal toxicant."

Table 5.6. Kidney Effects in F1 Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet

Endpoint	Dietary Level (ppm)			
	0	1,000	4,500	8,000
Relative kidney weight; F0 adults				
Males	0.71 ± 0.05 (29)[a]	0.73 ± 0.076 (30)	0.76 ± 0.06 (30)[b]	0.82 ± 0.06 (28)[b]
Females	0.78 ± 0.69 (28)	0.78 ± 0.05 (27)	0.84 ± 0.07 (25)[b]	0.84 ± 0.06 (28)[b]
Relative kidney weight; F1 adults				
Males	0.68 ± 0.06 (30)	0.68 ± 0.05 (30)	0.74 ± 0.06 (29)[b]	0.74 ± 0.05 (28)[b]
Females	0.74 ± 0.070 (30)	0.75 ± 0.05 (30)	0.79 ± 0.06 (29)[b]	0.76 ± 0.06 (30)

[a]Mean ± SD, number of animals is indicated in parentheses.
[b]Significantly different from controls at $p < 0.05$.

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

5.10. Immunotoxicity

After topical application 6 hours/day, 5 times/week for 2 weeks (under semi-occlusive conditions) and re-challenge 1 week later, B6C3F1 mice (10/group) treated with DiHP (undiluted) did not show increased induction of immunoregulatory cytokines (namely IgE, IL-4, or IL-13) in the serum or IL-4 or IL-13 mRNA levels compared with controls (Butala et al., 2004).

5.11. Endocrine Toxicity

In the two-generation reproductive toxicity study (ExxonMobil Chemical Company, 2003), where groups of rats were exposed to DiHP at up to 8,000 ppm in the diet, the absolute mean weight of the pituitary was reportedly increased in high-dose F1 males, and correlated with

microscopic findings of *pars distalis* hypertrophy and/or hyperplasia. Cystic degeneration of the adrenal cortex was also noted in the same treatment group (incidence data were not shown).

In pregnant mice (5/group) with an estrogen receptor (ER)-mediated luciferase reporter gene system, significant induction of estrogenic activity was not detected in the tissues (liver, tibia, or femurs) of dams or in the fetuses exposed to DiHP (via the oral route on GDs 8–15 or via the intraperitoneal route on GD 14) at 100 mg/kg-day (ter Veld et al., 2009). However, a significant reduction in luciferase activity (of 20–40%) was observed in the placentas of DiHP-treated females. In males, estrogenic activity was not induced after treatment with DiHP via the intraperitoneal or oral routes of exposure at up to 100 mg/kg-day in the pituitary, brain, tibia, femur, testis, adrenal, or kidney (ter Veld et al., 2008). Other studies showed that DiHP exhibited little or no estrogenic activity in vitro using ER competitive binding and/or gene reporter assays (Takeuchi et al., 2005; Zacharewski et al., 1998).

Endpoints associated with the estrous cycle and levels of sex hormones in the serum are discussed in the reproductive toxicity section.

5.12. Reproductive Toxicity

In the dose range-finding developmental toxicity study (Exxon Chemical Americas, 1997, 1996), increased resorptions and post-implantation loss (3–6-fold), and decreased mean number of live fetuses/litter (27–50%) and fetuses/implantation sites (1.3–2-fold) were observed at ≥750 mg/kg-day (Table 5.7). The incidence of resorbed, dead, or malformed fetuses was increased 3-fold at 750 mg/kg-day and 6-fold at 1,000 mg/kg-day compared with controls. Although an increase in the ratio of male to female fetuses was also noted, the study authors indicated that difficulties in determining the sex of fetuses using an external sexing procedure (and results from the full developmental toxicity study) contribute to significant uncertainty with respect to these data.

Table 5.7. Significant Reproductive Effects in Crl:CDBR Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)				
	0	**250**	**500**	**750**	**1,000**
Number pregnant animals	7	7	6	6	7
Resorptions/litter	1.71 ± 2.06[a]	0.86 ± 0.90	1.33 ± 1.37	5.00 ± 3.74[b]	9.86 ± 5.43[b]
Resorptions/implantation sites	0.10 ± 0.13	0.05 ± 0.05	0.07 ± 0.07	0.32 ± 0.23[b]	0.57 ± 0.32[b]
Post-implantation loss (%)	10.3 ± 13.3	5.0 ± 5.3	7.0 ± 7.0	31.6 ± 23.3[b]	57.0 ± 32.4[b]
Fetuses/implantation sites	0.90 ± 0.13	0.95 ± 0.05	0.93 ± 0.07	0.69 ± 0.23[b]	0.43 ± 0.32[b]
Live fetuses/litter	14.86 ± 2.67	16.00 ± 1.00	15.83 ± 1.17	10.83 ± 3.87[b]	7.43 ± 5.44[b]
Males	6.57 ± 3.21	7.43 ± 2.88	6.50 ± 2.88	3.33 ± 2.34[b]	2.43 ± 2.23[b]
Females	8.29 ± 1.80	8.57 ± 3.10	9.33 ± 3.50	7.50 ± 3.62[b]	4.86 ± 4.14[b]
Affected fetuses[c]	1.7 ± 2.1	0.9 ± 0.9	1.3 ± 1.4	5.7 ± 3.7[b]	10.3 ± 5.4[b]

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$.
[c]Affected fetuses=(resorptions + dead + malformed) fetuses.

Sources: Exxon Chemical Americas (1997, 1996).

In the full developmental toxicity study (McKee et al., 2006; Exxon Chemical Company, 1997), significant increases in the mean number of resorptions/dam and resorptions/implantation sites (6-fold), increased post-implantation loss (7-fold) and significant decreases in the mean numbers of fetuses/implantation sites (27%) and viable fetuses/dam (30%) were observed at the high dose (750 mg/kg-day; Table 5.8). Uterine weight was also significantly decreased with respect to controls in this dose group (30% lower than controls). The incidence of resorbed, dead, or malformed fetuses was about 10-fold higher at 750 mg/kg-day than in controls. Endpoints of reproductive function were similar to controls for all other treatment groups.

Table 5.8. Significant Effects in Crl:CDBR VAF/Plus Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)			
	0	**100**	**300**	**750**
Number pregnant animals	23	21	22	23
Uterine weight (g)	109 ± 11[a]	106 ± 9	103 ± 23	76 ± 26[b]
Resorptions/dam	0.74 ± 1.01	0.62 ± 0.74	0.59 ± 0.85	4.70 ± 3.55[b]
Resorptions/implantation sites	0.05 ± 0.06	0.04 ± 0.06	0.04 ± 0.06	0.31 ± 0.23[b]
Post-implantation loss (%)	4.6 ± 6.0	4.0 ± 4.8	4.2 ± 6.1	31.1 ± 23.0[b]
Fetuses/implantation sites	0.95 ± 0.06	0.96 ± 0.05	0.96 ± 0.06	0.69 ± 0.23[b]
Live fetuses/dam	15.09 ± 1.56	14.57 ± 1.29	14.00 ± 3.46	10.65 ± 3.97[b]
Males	7.30 ± 2.38	6.71 ± 2.08	7.27 ± 1.78	5.61 ± 2.62[b]
Females	7.78 ± 2.21	7.86 ± 2.59	6.73 ± 2.45	5.04 ± 2.57[b]
Affected fetuses[c]	1.0 ± 1.1	0.9 ± 1.0	0.8 ± 1.0	9.8 ± 4.4[b]

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$.
[c]Affected fetuses=(resorptions + dead + malformed) fetuses.

Sources: McKee et al. (2006); Exxon Chemical Company (1997).

In the two-generation reproductive toxicity study (McKee et al., 2006; ExxonMobil Chemical Company, 2003), no significant differences in reproductive outcomes were reported in the F0 generation (≥87% of females reportedly became pregnant and numbers/sex of live offspring were similar in all treatment groups). However, impairments in both mating and fertility were apparent in high-dose F1 animals. In the F1 generation, indices of mating and fertility were reduced 33–40% in both sexes at 8,000 ppm compared with controls (Table 5.9). Similarly, F0 males showed no significant effects on sperm endpoints and females showed no significant effects on estrous cyclicity or gestational length. However, adult F1 males exhibited significant decreases in left testis sperm concentration and sperm production rate at ≥1,000 ppm (each decreased 39%); left cauda epididymal sperm concentration was also significantly reduced (by 44%) at 8,000 ppm (Table 5.9). The study authors noted that these effects could be related to clinical signs of toxicity associated with the external genitalia (hypospadias, absent or undescended testes) observed in F1 males. No effects on other sperm parameters (motility or morphology) were observed. Likewise, numbers of primordial follicles in the ovaries of treated females were comparable with controls.

Table 5.9. Significant Reproductive Effects in F1 Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet

Endpoint	Dietary Level (ppm)			
	0	1,000	4,500	8,000
Mating index; F1 adults[a]	29/30 (96.7%) 29/30 (96.7%)	28/30 (93.3%) 28/30 (93.3%)	27/29 (93.1%) 27/29 (93.1%)	17/30 (56.7%)[b] 19/30 (63.3%)[b]
Fertility index; F1 adults[c]	24/30 (80.0%) 24/30 (80.0%)	24/30 (80.0%) 24/30 (80.0%)	20/29 (69.0%) 20/29 (69.0%)	12/28 (42.9%)[b] 12/30 (40.0%)[b]
Sperm parameters; F1 adults				
Left cauda epididymis sperm concentration (10^6/g)	662.8 ± 149 (23)[d]	700 ± 225 (23)	717 ± 244 (24)	374 ± 319 (20)[b]
Left testis sperm concentration (10^6/g)	93.2 ± 14.8 (30)	56.7 ± 14.8 (21)[b]	57.6 ± 21.6 (20)[b]	49.5 ± 44.3 (28)[b]
Sperm production rate (10^6/day)	15.3 ± 2.4 (30)	9.3 ± 2.4 (21)[b]	9.4 ± 3.5 (20)[b]	8.1 ± 7.3 (28)[b]
Testis weight (g)				
Right	1.90 ± 0.18 (30)	1.96 ± 0.17 (30)	1.98 ± 0.25 (29)	1.30 ± 0.75 (27)[b]
Left	1.91 ± 0.19 (30)	1.93 ± 0.23 (30)	1.95 ± 0.28 (29)	1.35 ± 0.71 (28)[b]
Epididymis weight (g)				
Right	0.76 ± 0.07 (30)	0.76 ± 0.07 (30)	0.73 ± 0.10 (29)	0.47 ± 0.25 (23)[b]
Left	0.77 ± 0.10 (30)	0.76 ± 0.10 (30)	0.73 ± 0.11 (29)	0.51 ± 0.24 (24)[b]
Cauda epididymis weight (g)				
Right	0.34 ± 0.04 (30)	0.35 ± 0.03 (30)	0.31 ± 0.06 (29)	0.20 ± 0.10 (23)[b]
Left	0.35 ± 0.05 (30)	0.35 ± 0.05 (30)	0.33 ± 0.06 (29)	0.23 ± 0.10 (24)[b]
Ovary weight (g)	0.13 ± 0.02 (30)	0.12 ± 0.03 (30)	0.12 ± 0.02 (29)	0.11 ± 0.02 (30)[b]
Histopathology; F1 adults				
Seminiferous tubule degeneration	2/?[e]	1/?	7/29	22/27
Hypospermia (epididymis)	0/30	0/30	1/29	10/23[f]

[a]Male (female) mating index (%)=[number males (females) with evidence of mating]/[total number males (females) used for mating].
[b]Significantly different from controls at $p < 0.05$ (as reported by the study authors).
[c]Male fertility index (%)=[number males siring a litter]/[number males used for mating]. Female fertility index (%)=[number females confirmed pregnant]/[number females used for mating].
[d]Mean ± SD, number of animals is indicated in parentheses.
[e]Number affected/number examined.
[f]Significantly different from controls at $p < 0.05$ based on Fisher's test performed for this analysis.

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

Also in the adult F1 generation, the weights of several reproductive organs were significantly affected by treatment with DiHP at 8,000 ppm, including the testes (decreased 29–32%), epididymides (decreased 34–38%), cauda epididymides (decreased 34–41%), seminal vesicle with coagulating gland (data not shown), and prostate (data not shown) in males, and the ovary (decreased 15%) in females. With respect to the histology of the reproductive organs, degeneration of the seminiferous tubules was noted in adult F1 males, primarily in the 4,500 and 8,000 ppm treatment groups (Table 5.9; data not reported in their entirety). The severity of this

lesion reportedly increased in a dose-related manner, with 1 of 7 mid-dose and 10 of 22 affected high-dose F1 males having lesions classified as severe. In the epididymis, significant hypospermia, affecting 43% of examined adult F1 males, was observed at 8,000 ppm. Microscopic degeneration, decreased secretion, and/or absence of other parts of the reproductive tract (including the testes, prostate, seminal vesicles, seminiferous tubules, vas deferens, coagulating gland, and epididymis) were noted in adult F1 males exposed to DiHP at 8,000 ppm, but the incidence of these lesions were not quantified.

In immature ovarectomized Sprague-Dawley rats (24–25 days of age; 10/group), wet uterine weight was not significantly increased after treatment with DiHP (combined with other phthalate diesters) for 4 days via gavage at up to 2,000 mg/kg-day; mature ovarectomized rats (51–56 days of age) administered DiHP alone using the same treatment conditions did not induce a significant degree of vaginal epithelial cell cornification (Zacharewski et al., 1998). In agreement with these in vivo data, DiHP did not exhibit estrogenic activity in vitro using ER competitive binding and gene expression assays.

The weight of evidence from the above studies supported the conclusion that there was "sufficient animal evidence" for the designation of DiHP as a "reproductive toxicant."

5.13. Prenatal, Perinatal, and Post-natal Toxicity

In the dose range-finding developmental toxicity study (Exxon Chemical Americas, 1997, 1996), the number of live fetuses/litter was significantly decreased (27–50%) and the incidence of dams with affected (resorbed, dead, or malformed) fetuses was significantly increased (3–6-fold) in rats treated at ≥750 mg/kg-day (Table 5.10). Mean weights of male and female fetuses were 14–18% lower in the 750 and 1,000 mg/kg-day groups relative to controls. External malformations observed at 750 mg/kg-day (four fetuses from three litters) and 1,000 mg/kg-day (three fetuses from two litters) included encephalocele, cleft palate, filamentous tail, atresia ani, acaudia, exencephaly, anasarca, cranial hematomas, and/or protruding tongue. Stunted growth (defined as body weight ≤4.0 g) was noted in eight and nine fetuses (from four litters) at 750 and 1,000 mg/kg-day, respectively. A single case of stunted growth at 250 mg/kg-day was observed, but was not considered to be treatment-related by the researchers.

Table 5.10. Significant Developmental Effects in Crl:CDBR Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)				
	0	**250**	**500**	**750**	**1,000**
Number pregnant animals	7	7	6	6	7
Live fetuses/litter	14.86 ± 2.67[a]	16.00 ± 1.00	15.83 ± 1.17	10.83 ± 3.87[b]	7.43 ± 5.44[b]
Affected dams[c]	1.7 ± 2.1	0.9 ± 0.9	1.3 ± 1.4	5.7 ± 3.7[b]	10.3 ± 5.4[b]
Mean body weight (g)					
Males	5.52 ± 0.47	5.37 ± 0.35	5.47 ± 0.49	4.54 ± 0.45[b]	4.73 ± 0.50[b]
Females	5.43 ± 0.50	5.13 ± 0.34	5.25 ± 0.60	4.58 ± 0.55[b]	4.58 ± 0.68[b]

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$.
[c]Affected dams=(resorptions + dead + malformed) fetuses.

Sources: Exxon Chemical Americas (1997, 1996).

In the full developmental toxicity study (McKee et al., 2006; Exxon Chemical Company, 1997), the number of viable fetuses per dam was reduced by 30% and mean fetal weights (both males and females) were reduced by 10–11% at 750 mg/kg-day relative to controls (Table 5.11). Also at this dose, increased incidences of external, visceral, and skeletal malformations were observed; numbers of litters with the aforementioned malformations were increased by 53, 61, and 100% compared with controls, respectively. External examinations of fetuses revealed increased incidences of stunted growth (defined as body weight <4 g) and anophthalmia at 750 mg/kg-day (12/23 and 6/23 litters, respectively); no significant external malformations were observed in lower dose groups. Visceral malformations were also increased only at the high-dose, namely incidences of ectopic testes or ovaries (8 or 6/23 litters, respectively versus 0/23 in controls), and/or malformations of the subclavian or innominate arteries (9 or 10/23 litters versus 1/23 or 0/23 in controls, respectively). Incidence data for fetuses with visceral malformations of the reproductive organs were not adequately reported, since only the total number of fetuses examined at each dose, but not the numbers of male and females examined, were provided. Litters exposed to 750 mg/kg-day DiHP showed numerous significant skeletal variations and malformations, including fused or malformed sternabrae, thoracic centra/arch genesis, and malformed rib cartilage, occurring in as few as 6 to as many as 20 of 23 litters. The incidence of rudimentary lumbar ribs (classified as a skeletal variation) was also increased at 300 mg/kg-day (11/23 litters versus 4/23 controls); no skeletal malformations were observed at this dose.

Table 5.11. Significant Developmental Effects in Crl:CDBR VAF/Plus Offspring Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day) 0	100	300	750
Number litters (fetuses)	23 (347)	21 (306)	22 (308)	23 (245)
Viable fetuses/dam	15.09 ± 1.56	14.57 ± 1.29	14.00 ± 3.46	10.65 ± 3.97[a]
Body weight[b] (g)				
Males	5.30 ± 0.44[c]	5.28 ± 0.35	5.40 ± 0.41	4.73 ± 0.52[a]
Females	5.02 ± 0.41	5.02 ± 0.38	5.09 ± 0.39	4.53 ± 0.57[a]
Fetuses/litter with malformations	0.26 ± 0.54	0.24 ± 0.44	0.18 ± 0.39	5.13 ± 2.56[a]
Fetuses/litter with variations	1.5 ± 0.9	1.3 ± 1.8	2.0 ± 2.0	5.1 ± 2.1[a]
External observations				
Litters: external malformations	1/23	2/21	0/22	13/23[a]
Stunted growth[d]	2/23 (2/347)[e]	1/21 (1/306)	1/22 (1/308)	12/23 (25/245)[a]
Anophthalmia	0/23 (0/347)	1/21 (1/306)	0/22 (0/308)	6/23 (7/245)[a]
Microphthalmia	0/23 (0/347)	0/21 (0/306)	0/22 (0/308)	3/23 (4/245)
Visceral observations				
Discolored liver	0/23 (0/172)	0/21 (0/151)	1/22 (1/155)	6/23 (7/122)[a]
Litters: visceral malformations	4/23	3/21	4/22	18/23[a]
Ectopic testes[f]	0/23 (0/?)	0/21 (0/?)	0/22 (0/?)	8/23 (11/?)[a]
Ectopic ovaries[f]	0/23 (0/?)	0/21 (0/?)	0/22 (0/?)	6/23 (10/?)[a]
Abnormal origin subclavian artery	1/23 (1/172)	1/21 (1/151)	0/22 (0/155)	9/23 (11/122)[a]
Agenesis innominate artery	0/23 (0/172)	1/21 (1/151)	0/22 (0/155)	10/23 (13/122)[a]
Skeletal observations				
Litters: skeletal variations	20/23	10/21	15/22	23/23
Sternabrae Asymmetric	0/23 (0/175)	0/21 (0/155)	2/22 (2/153)	13/23 (20/123)[a]
Bifid	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	8/23 (11/123)[a]
Hypoplastic	1/23 (1/175)	0/21 (0/155)	1/22 (1/153)	13/23 (31/123)[a]
Ribs				
Rudimentary lumbar	4/23 (6/175)	6/21 (10/155)	11/22 (27/153)[a]	23/23 (94/123)[a]
Well-formed lumbar	1/23 (1/175)	0/21 (0/155)	0/22 (0/153)	9/23 (14/123)[a]
Vertebrae				
Thoracic centra bifid	2/23 (4/174)	3/21 (3/155)	1/22 (3/153)	14/123 (9/23)[a]
Thoracic centra hypoplastic	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	5/23 (5/123)[a]
Lumbar centra dumbbell-shaped	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	5/23 (6/123)[a]
Litters: skeletal malformations	0/23	1/21	0/22	23/23[a]
Sternabrae Fused	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	20/23 (58/123)[a]
Malformed	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	11/23 (21/123)[a]
Fused ribs	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	7/23 (8/123)[a]
Vertebrae				
Thoracic centra/arch genesis	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	6/23 (7/123)[a]
Rib Anlage Fused (thoracic)	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	15/23 (26/123)[a]
Short	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	12/23 (17/123)[a]

[a]Significantly different from controls at $p < 0.05$.
[b]Calculated on a per litter basis.
[c]Mean ± SD.
[d]Stunted growth was not included in tallies of external malformations.
[e]Number of individuals (n) is given in parentheses.
[f]Incidence data could not be reported because distribution of males and females was not reported.

Sources: McKee et al. (2006); Exxon Chemical Company (1997).

In the two-generation reproductive toxicity assessment (McKee et al., 2006; ExxonMobil Chemical Company, 2003), treatment with DiHP did not affect numbers of F1 or F2 pups born, numbers of live pups/litter, or offspring survival up to postnatal day (PND) 21. In general, pup weights were lower in DiHP-exposed rats than controls during lactation; significantly decreased pup weights were reported in F1 females at 8,000 ppm group (PND 21), in F2 males at 4,500 and 8,000 ppm, and F2 females at 8,000 ppm group (PNDs 14 and 21) (data not shown). Anogenital distance (AGD) was significantly decreased in F1 males, but not females, exposed to DiHP at 8,000 ppm; AGD was decreased by 15% relative to controls at birth (on PND 1) and at study termination (on PND 156) in this group (AGD was not assessed at all time points in all dose groups; see Table 5.12). AGD adjusted to the cube root of body weight was also reportedly significantly decreased in high-dose F1 males (data not shown). A significant reduction in AGD (of 10%) persisted in F1 males in the NTR group (treated only through lactation) on PND 156 (date of scheduled necropsy). In the F2 generation, AGD (absolute values [and values adjusted for pup weight; not shown]) were significantly decreased (by 13–22%) in males at ≥4,500 ppm. In F1 males, the incidence of retained nipples was significantly increased at 8,000 ppm (5–7% incidence, versus 0% in controls). F1 males, but not F1 females, also showed a delay in the time required to reach puberty; balanopreputial separation occurred in males exposed to DiHP at 8,000 ppm (8,000 ppm and NTR groups) 2–4 days later than non-exposed controls. Five F1 males in the 8,000 ppm group were excluded from the analyses because balanopreputial separation did not occur before study termination. External malformations of the genitalia were observed in F1 adult males of the 8,000 ppm group, including hypospadias (7/30 males) and absent (2/30 males) or undescended (2/30 males) testes. One male (of 30) in the 1,000 ppm group also had an undescended testis.

Table 5.12. Significant Effects in the Offspring of Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet					
	Dietary Level (ppm)				
Endpoint	**0**	**1,000**	**4,500**	**8,000**	**NT Group[a]**
AGD (mm); F1 males					
PND 1	4.34 ± 0.34 (28)[b]	4.32 ± 0.36 (27)	4.25 ± 0.47 (25)	3.71 ± 0.41 (28)[c]	Not assessed
PND 21	18.0 ± 2.4 (30)	Not assessed	Not assessed	15.0 ± 1.7 (30)[c]	15.5 ± 1.5 (29)[c]
PND 156	48.7 ± 2.9 (29)	Not assessed	Not assessed	41.6 ± 3.6 (28)[c]	43.8 ± 3.7 (28)[c]
AGD (mm); F2 males					
PND 1	4.32 ± 0.39 (24)	4.26 ± 0.40 (23)	3.74 ± 0.36 (19)[c]	3.38 ± 0.31 (12)[c]	Not assessed
Retention thoracic nipples (%); F1 males					
Day 11	0	0	0	7.1[c]	Not assessed
Day 12	0	0	0	5.4[c]	Not assessed
Day 13	0	0	0	6.3[c]	Not assessed
Time to balanopreputial separation (days); F1 males	46.1[d]	45.4	47.4	50.3[c]	48.2[c]

[a]Animals in the "non-treated" NT group were exposed to DiHP at 8,000 ppm until the end of the lactation period and then held without treatment until terminal sacrifice (PND 156).
[b]Mean ± SD, number of animals is indicated in parentheses.
[c]Significantly different from controls at $p < 0.05$ (as reported by the study authors).
[d]Mean value as provided in text; standard error (SE) or SD were not shown.

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

The weight of evidence from the above studies supported the conclusion that there was "sufficient animal evidence" for the designation of DiHP as a "probable developmental toxicant."

5.14. Carcinogenicity

Genotoxicity

In a test in *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537, and TA1538 using DiHP (at 250, 500, 1,000, 2,500, or 5,000 mg/mL), DiHP was not shown to be mutagenic in the presence or absence of metabolic activation (Exxon Biomedical Sciences, 1995; as cited in NICNAS, 2008 and European Commission, 2000). Chromosomal aberrations were not induced by DiHP (at concentrations of 499, 1,250, 2,500, 3,750, or 4,990 mg/mL) in Chinese hamster ovary cells in the presence of absence of metabolic activation (Hazelton Laboratories America, 1991; as cited in NICNAS, 2008 and European Commission, 2000).

Initiation and Promotion

No initiation and promotion studies were located for DiHP.

Carcinogenicity Studies

No carcinogenicity or chronic toxicity studies were located for DiHP.

The weight of evidence from the above studies supported the conclusion that there was "insufficient animal evidence" for the designation of DiHP as a "carcinogen."

6. EXPOSURE

ECHA (2011) has reported that occupational exposure to DiHP may occur through inhalation of aerosols and dermal contact with this compound at workplaces where it is produced or used. The available monitoring data indicate that the general population may be exposed to DiHP via inhalation of ambient air and dermal contact with products containing DiHP, such as PVC flooring and coatings (ECHA, 2011). Exposure data specific to DiHP were not found.

7. DISCUSSION

Appendix A provides a summary of the no-observed-adverse-effect level (NOAEL) and lowest-observed-adverse-effect level (LOAEL) values for organ-specific endpoints for DiHP, most of which are derived from the gestational studies reported in Exxon Chemical Americas (1997, 1996), Exxon Chemical Company (1997), and McKee et al. (2006) and the two-generation reproductive toxicity study reported in ExxonMobil Chemical Company (2003) and McKee et al. (2006).

The most sensitive measures of effect were obtained in the two-generation reproductive study. This study found reproductive effects in F1 males at 1,000 ppm, the lowest dietary concentration tested, and further effects on sexual development of male offspring at higher doses. Effects progressed from decreased testis sperm concentration and sperm production rate in F1 adult males at ≥1,000 ppm (50–91 mg/kg-day in this group) to decreased AGD in F2 male pups at ≥4,500 ppm (222–750 mg/kg-day) to decreased AGD in F1 male pups, increased thoracic nipple retention in F1 male pups, increased time to balanopreputial separation in F1 male juveniles, decreased testis and epididymis weights and increased testicular and epididymal

lesions in F1 male adults, increased incidence of malformations of the external genitalia (hypospadias and absent or undescended testes) in F1 adult males, and reduced mating and fertility of F1 adults at 8,000 ppm in the diet (404–1,360 mg/kg-day).

These findings are supported by the gavage gestational exposure studies, which identified 750 mg/kg-day as a LOAEL for embryotoxicity and fetotoxicity, as shown by increased resorptions and post-implantation loss, decreased viable fetuses, decreased fetal weights, and increased incidences of external, visceral, and skeletal malformations, including ectopic testes or ovaries. In these studies, overt maternal effects occurred at the same doses as the developmental effects, but dam liver weights were increased at lower doses (LOAEL=300 mg/kg-day, NOAEL=100 mg/kg-day). Increased liver and kidney weights were also seen at relatively low doses in the multigeneration study (≥4,500 ppm [222–750 mg/kg-day]) and accompanied by histopathological findings (increases incidences of centrilobular hypertrophy and vacuolation in the liver and hydrophenosis/dilated pelvis in the kidney) in this study.

The short-term repeated-dose study of Smith et al. (2000) found liver effects at even lower doses. In this study, liver weight and the rate of hepatocellular replicative DNA synthesis were increased at ≥1,000 ppm (~100 mg/kg-day) in rats. Replicative DNA synthesis was also increased in mice exposed to DiHP at ≥500 ppm (~90 mg/kg-day). PBOX activity was increased at higher doses in both species.

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Appendix A. Summary of Endpoints by Organ Systems

Table A.1. Summary of NOAELs/LOAELs Identified for DiHP by Organ System

Species (Sex)	Exposure Route	Dose (Number Animals Per Dose Group)	Dose Duration	Effect Category	Toxicological Endpoint	Toxicological Basis	Citation
F344 rat (M)	Oral (diet)	0, 1,000, or 12,000 ppm 0, 100, or 1,200 mg/kg-day (estimated for this review) (5/group)	2 or 4 weeks	Liver	NOAEL=None LOAEL=100 mg/kg-day	Increased relative liver weight and hepatocellular replicative DNA synthesis; increased PBOX at 1,200 mg/kg-day	Smith et al., 2000
B6C3F1 mouse (M)	Oral (diet)	0, 500, or 6,000 ppm 0, 90, or 1,100 mg/kg-day (estimated for this review) (5/group)	2 or 4 weeks	Liver	NOAEL=None LOAEL=90 mg/kg-day	Increased hepatocellular replicative DNA synthesis; increased PBOX at 1,100 mg/kg-day	Smith et al., 2000
Crl:CDBR rat (F)	Gavage in corn oil	0, 250, 500, 750, or 1,000 mg/kg-day (7/group)	GDs 6–20	General	NOAEL=500 mg/kg-day LOAEL=750 mg/kg-day	Decreased body weight	Exxon Chemical Americas, 1997, 1996
				Reproduction	NOAEL=500 mg/kg-day LOAEL=750 mg/kg-day	Decreased uterine weight; increased resorptions and postimplantation loss at 1,000 mg/kg-day	
				Development/fetus	NOAEL=500 mg/kg-day LOAEL=750 mg/kg-day	Decreased number of live fetuses; decreased fetal weights; increased incidence of external malformations	

Table A.1. Summary of NOAELs/LOAELs Identified for DiHP by Organ System							
Species (Sex)	**Exposure Route**	**Dose (Number Animals Per Dose Group)**	**Dose Duration**	**Effect Category**	**Toxicological Endpoint**	**Toxicological Basis**	**Citation**
Crl:CDBR VAF/Plus rat (F)	Gavage in corn oil	0, 100, 300, or 750 mg/kg-day (25/group)	GDs 6–20	General	NOAEL=300 mg/kg-day LOAEL=750 mg/kg-day	Decreased body weight	McKee et al., 2006; Exxon Chemical Company, 1997
				Liver	NOAEL=100 mg/kg-day LOAEL=300 mg/kg-day	Increased absolute and relative liver weights	
				Reproduction	NOAEL=300 mg/kg-day LOAEL=750 mg/kg-day	Decreased uterine weight; increased resorptions and postimplantation loss	
				Development/fetus	NOAEL=300 mg/kg-day LOAEL=750 mg/kg-day	Decreased viable fetuses; decreased fetal weights; increased incidences of external, visceral, and skeletal malformations	
Crl:CD(SD)IGS BR rat (M&F)	Oral (diet)	0, 1,000, 4,500, or 8,000 ppm 0, 50–168, 222–750, or 404–1,360 mg/kg-day (calculated by the researchers) (30/sex/dose)	Starting at 6 weeks of age (F0 animals) and continuing for two generations	General	NOAEL=309–750 mg/kg-day LOAEL=543–1,360 mg/kg-day	Decreased body weight in F1 females during gestation and lactation	McKee et al., 2006; ExxonMobil Chemical Company, 2003
				Liver	NOAEL=50–168 mg/kg-day LOAEL=222–750 mg/kg-day	Increased relative liver weight in F0 and F1 adult males and females; minimal centrilobular hypertrophy in males (females at higher dose); vacuolation in males at higher dose	
				Kidney	NOAEL=50–168 mg/kg-day LOAEL=222–750 mg/kg-day	Increased relative kidney weight in F0 and F1 adult males and females; hydronephrosis/dilated renal pelvis in F1 adult males	
				Endocrine	NOAEL=227–416 mg/kg-day LOAEL=419–764 mg/kg-day	Increased absolute pituitary weight in F1 adult males; *pars distalis* hypertrophy/ hyperplasia in F1 adult males; cystic degeneration of the adrenal cortex in F1 adult males	

Table A.1. Summary of NOAELs/LOAELs Identified for DiHP by Organ System							
Species (Sex)	**Exposure Route**	**Dose (Number Animals Per Dose Group)**	**Dose Duration**	**Effect Category**	**Toxicological Endpoint**	**Toxicological Basis**	**Citation**
				Reproduction	Parental males: NOAEL=None LOAEL=50–91 mg/kg-day Parental females: NOAEL=309–750 mg/kg-day LOAEL=543–1,360 mg/kg-day	Decreased testis sperm concentration and sperm production rate in F1 adult males; decreased testis and epididymis weights, increased seminiferous tubule degeneration and hypospermia, and decreased mating and fertility in F1 adult males at higher doses; decreased ovary weight, mating, and fertility in F1 adult females	
				Development/pup	NOAEL=50–168 mg/kg-day LOAEL=222–750 mg/kg-day	Decreased AGD and pup weight in F2 male pups; decreased AGD in F1 male pups, increased retention of thoracic nipples (assessed F1 male pups only), increased time to balanopreputial separation (assessed F1 male pups only), increased incidence of malformations of the external genitalia [hypospadias and absent or undescended testes] in F1 adult males, and decreased female pup weights at higher doses	

Appendix B. Critical Study Reviews

Exxon Chemical Americas (1997, 1996)

As part of a dose range-finding developmental toxicity study, timed-pregnant Crl:CDBR rats (7/group) were administered DiHP in corn oil via gavage at 0, 250, 500, 750, or 1,000 mg/kg-day on GDs 6–20 (Exxon Chemical Americas, 1997, 1996). Mortality and clinical signs of toxicity were monitored daily; body weights and feed consumption were measured on GDs 0, 6, 9, 12, 15, 18, 20 (body weight only), and 21. Animals were sacrificed on GD 21, and Cesarean sections were performed. Animals were subjected to gross necropsy, and uterine weights (with attached ovaries) were noted. Numbers of corpora lutea, implantation sites, live and dead fetuses, and early and late resorptions were recorded. Fetuses were sexed, weighed, and examined externally for gross malformations.

Significant effects in DiHP-treated rats are summarized in Table B.1 (Exxon Chemical Americas, 1997, 1996). No mortality occurred, and no treatment-related clinical signs were evident in treated dams. Relative to controls, body weights were lower in rats treated at ≥750 mg/kg-day for most of the treatment period; by GD 21, body weights were reduced by 8 and 14% at 750 and 1,000 mg/kg-day, respectively. Body weights adjusted for gravid uterus weight were similar to controls for all treatment groups, suggesting that these observed reductions in body weight, particularly toward the end of gestation, were associated with decreased uterine weights. Compared to controls, reductions in mean uterine weight of 31% at 750 mg/kg-day and 52% at 1,000 mg/kg-day were observed. Food consumption was similar among treatment and control groups, and no treatment-related findings were reported at necropsy. In the 1,000 mg/kg-day group, increased numbers of resorptions (6-fold), increased post-implantation loss (5-fold), and an increased incidence of dams with affected (i.e., resorbed, dead, or malformed) fetuses (6-fold) were observed. Rats treated at 1,000 mg/kg-day also showed decreases in the total numbers of live fetuses (2-fold), male fetuses (3-fold), and fetuses/implantation sites (2-fold). The significance of the 3-fold decrease in the number of male fetuses at 1,000 mg/kg-day was unclear due to difficulties in determining fetal sex using an external sexing procedure. The total number of live fetuses and the incidence of affected fetuses were also significantly altered in rats treated at 750 mg/kg-day (decreased 27% and increased 3-fold, respectively). Mean weights of male and female fetuses were 14–18% lower in the 750 and 1,000 mg/kg-day groups relative to controls. External malformations observed in the 750 and 1,000 mg/kg-day dose groups included encephalocele, cleft palate, filamentous tail, atresia ani, acaudia, exencephaly, anasarca, cranial hematomas, and protruding tongue. Stunted

growth (body weight ≤4.0 g) was noted in eight and nine fetuses at 750 and 1,000 mg/kg-day, respectively. A single case of stunted growth was observed at 250 mg/kg-day, but was considered by the researchers to be unrelated to treatment.

Table B.1. Significant Effects in Crl:CDBR Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)				
	0	250	500	750	1,000
Maternal data					
Number pregnant animals	7	7	6	6	7
Body weight gain (g) GDs 0–21	204.0 ± 14.7[a]	213.7 ± 12.2	221.3 ± 17.1	169.3 ± 24.2[b]	138.9 ± 46.8[b]
Body weight, GD 21 (g)	476.6 ± 26.4	487.1 ± 19.0	494.3 ± 23.8	440.3 ± 27.8[b]	411.1 ± 53.5[b]
Body weight adjusted for gravid uterus weight (g)	365.4 ± 20.3	371.1 ± 15.6	378.5 ± 19.7	364.2 ± 28.0	357.4 ± 23.3
Uterine weight (g)	111.1 ± 14.7	116.0 ± 9.3	115.8 ± 11.9	76.2 ± 25.6[b]	53.7 ± 33.3[b]
Resorptions	1.71 ± 2.06	0.86 ± 0.90	1.33 ± 1.37	5.00 ± 3.74	9.86 ± 5.43[b]
Resorptions/implantation sites	0.10 ± 0.13	0.05 ± 0.05	0.07 ± 0.07	0.32 ± 0.23	0.57 ± 0.32[b]
Fetuses/implantation sites	0.90 ± 0.13	0.95 ± 0.05	0.93 ± 0.07	0.69 ± 0.23	0.43 ± 0.32[b]
Post-implantation loss (%)	10.3 ± 13.3	5.0 ± 5.3	7.0 ± 7.0	31.6 ± 23.3	57.0 ± 32.4[b]
Total live fetuses	14.86 ± 2.67	16.00 ± 1.00	15.83 ± 1.17	10.83 ± 3.87[b]	7.43 ± 5.44[b]
Total affected dams[c]	1.7 ± 2.1	0.9 ± 0.9	1.3 ± 1.4	5.7 ± 3.7[b]	10.3 ± 5.4[b]
Fetal data					
Mean body weight (g)					
Males	5.52 ± 0.47	5.37 ± 0.35	5.47 ± 0.49	4.54 ± 0.45[b]	4.73 ± 0.50[b]
Females	5.43 ± 0.50	5.13 ± 0.34	5.25 ± 0.60	4.58 ± 0.55[b]	4.58 ± 0.68[b]

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$ based on t-test (for continuous data) or Fisher's exact test (for incidence data) performed for this review.
[c]Affected dams=(resorptions + dead + malformed) fetuses.

Sources: Exxon Chemical Americas (1997, 1996).

A LOAEL of 750 mg/kg-day and a NOAEL of 500 mg/kg-day for maternal (decreased body weight due to decreased uterine weight) and developmental (decreased live fetuses, decreased fetal weights and increased external malformations) effects are identified from this study (Exxon Chemical Americas, 1997, 1996).

McKee et al. (2006); Exxon Chemical Company (1997)

In the subsequent full study, timed-pregnant Crl:CDBR VAF/Plus rats (25/group) were administered DiHP in corn oil via gavage at 0, 100, 300, or 750 mg/kg-day on GDs 6–20 (McKee et al., 2006; Exxon Chemical Company, 1997). Examinations for viability occurred twice daily and clinical signs were regularly monitored. Body weights were measured prior to the start of the experiment and on GDs 0, 6, 9, 12, 15, 18, 20, and 21; food consumption was measured on GDs 6, 9, 12, 15, 18, and 21. Animals were sacrificed on GD 21, and Cesarean sections were performed. Animals were subjected to gross necropsy and uterine weights (with attached ovaries) were noted. Numbers of corpora lutea, implantation sites, live and dead fetuses, and early and late resorptions were recorded. Fetuses were sexed, weighed, and examined for external malformations. Half of the fetuses were examined for visceral malformations; the remaining fetuses were examined for skeletal malformations. Fetal sex was confirmed internally for fetuses that were examined for visceral abnormalities. The fetus was considered the unit for statistically analyses.

Maternal data are summarized in Table B.2. With the exception of two dams that gave birth before scheduled sacrifice and were euthanized (one from the control group and one from the 300 mg/kg-day group), all animals survived to study termination (McKee et al., 2006; Exxon Chemical Company, 1997). No treatment-related clinical signs of toxicity were reported, and no changes in food consumption were detected in treated rats. Mean maternal body weight was significantly decreased (by 7%) in animals treated at 750 mg/kg-day on GD 21; however, adjustment of these body weights for gravid uterine weight resulted in maternal body weight values that were similar among all treatment groups (including controls; Table B.2). The difference in terminal body weights observed for control and high-dose rats was due to a mean reduction in uterine weight of 30% in the high-dose group relative to controls. Significant increases in absolute and relative liver weights were reported at ≥300 mg/kg-day. In the high-dose group, significant increases in the mean number of resorptions/dam (6-fold) and post-implantation loss (7-fold) and a significant decrease in mean number of viable fetuses/dam (30%) were observed.

Table B.2. Significant Effects in Crl:CDBR VAF/Plus Rats Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)			
	0	100	300	750
Maternal data				
Number pregnant animals	23	21	22	23
Mean maternal body weight, day 21 (g)	441.6 ± 27.4[a]	440.5 ± 32.0	441.0 ± 30.9	412.2 ± 31.6[b]
Body weight adjusted for gravid uterus weight (g)	332.5 ± 26.6	334.9 ± 26.6	337.5 ± 21.4	335.7 ± 24.8
Uterine weight (g)	109 ± 11	106 ± 9	103 ± 23	76 ± 26[b]
Absolute liver weight (g)	16.0 ± 2.2	17.2 ± 2.0	17.8 ± 1.9[b]	19.2 ± 1.9[b]
Relative liver weight	0.049 ± 0.005	0.051 ± 0.004	0.053 ± 0.004[b]	0.057 ± 0.004[b]
Corpora lutea/dam	16.3 ± 1.5	16.0 ± 1.2	16.9 ± 3.2	15.9 ± 1.6
Resorptions/dam	0.74 ± 1.01	0.62 ± 0.74	0.59 ± 0.85	4.70 ± 3.55[b]
Post-implantation loss (%)	4.6 ± 6.0	4.0 ± 4.8	4.2 ± 6.1	31.1 ± 23.0[b]
Viable fetuses/dam	15.1 ± 1.6	14.6 ± 1.3	14.0 ± 3.5	10.6 ± 4.0[b]

[a]Mean ± SD.
[b]Significantly different from controls at $p < 0.05$.

Sources: McKee et al. (2006); Exxon Chemical Company (1997).

Fetal data are summarized in Table B.3 (McKee et al., 2006; Exxon Chemical Company, 1997). Mean fetal weights (both males and females) were reduced by 10–11% at 750 mg/kg-day relative to controls. At the high dose, increased incidences of external, visceral, and skeletal malformations were observed; numbers of litters with the aforementioned malformations were increased by 53, 61, and 100% compared with controls, respectively. External examinations of fetuses revealed increased incidences of stunted growth (defined as body weight <4 g) and anophthalmia at 750 mg/kg-day (12/23 and 6/23 litters, respectively); no significant external malformations were observed in lower dose groups. Visceral malformations were also increased only at the high dose, namely incidences of ectopic testes or ovaries (8 or 6/23 litters, respectively versus 0/23 in controls), and/or malformations of the subclavian or innominate arteries (9 or 10/23 litters versus 1/23 or 0/23 in controls, respectively). Incidence data for fetuses with visceral malformations of the reproductive organs were not adequately reported, since only the total number of fetuses examined at each dose, but not the numbers of male and females examined, were reported. Litters exposed to 750 mg/kg-day DiHP also showed numerous significant skeletal variations and malformations, including fused or malformed sternabrae, thoracic centra/arch genesis, and malformed rib cartilage, each occurring in 6–20/23 litters. The incidence of rudimentary lumbar ribs (classified as a skeletal variation) was

also increased at 300 mg/kg-day (11/23 litters versus 4/23 controls); no skeletal malformations were observed in this dosage group.

Table B.3. Significant Effects in Crl:CDBR VAF/Plus Offspring Administered DiHP Via Gavage on GDs 6–20

Endpoint	Dose (mg/kg-day)			
	0	100	300	750
Fetal data				
Number litters (fetuses)	23 (347)	21 (306)	22 (308)	23 (245)
Body weight[a] (g)				
Males	5.30 ± 0.44[b]	5.28 ± 0.35	5.40 ± 0.41	4.73 ± 0.52[c]
Females	5.02 ± 0.41	5.02 ± 0.38	5.09 ± 0.39	4.53 ± 0.57[c]
Sex distribution[a]				
Males	7.30 ± 2.38 (168)[d]	6.71 ± 2.08(141)	7.27 ± 1.78(160)	5.65 ± 2.64(130)
Females	7.78 ± 2.21(179)	7.86 ± 2.59(165)	6.73 ± 2.45(148)	5.00 ± 2.52(115)[c]
Fetuses/litter with malformations	0.26 ± 0.54	0.24 ± 0.44	0.18 ± 0.39	5.13 ± 2.56[c]
Fetuses/litter with variations	1.5 ± 0.9	1.3 ± 1.8	2.0 ± 2.0	5.1 ± 2.1[c]
Total affected fetuses/litter	1.0 ± 1.1	0.9 ± 1.0	0.8 ± 1.0	9.8 ± 4.4[c]
External observations				
Litters: external malformations	1/23	2/21	0/22	13/23[c]
Stunted growth[e]	2/23 (2/347)	1/21 (1/306)	1/22 (1/308)	12/23 (25/245)[c]
Anophthalmia	0/23 (0/347)	1/21 (1/306)	0/22 (0/308)	6/23 (7/245)[c]
Microphthalmia	0/23 (0/347)	0/21 (0/306)	0/22 (0/308)	3/23 (4/245)
Visceral observations				
Discolored liver	0/23(0/172)	0/21(0/151)	1/22 (1/155)	6/23 (7/122)[c]
Litters: visceral malformations	4/23	3/21	4/22	18/23[c]
Ectopic testes[f]	0/23 (0/?)	0/21 (0/?)	0/22 (0/?)	8/23 (11/?)[c]
Ectopic ovaries[f]	0/23 (0/?)	0/21 (0/?)	0/22 (0/?)	6/23 (10/?)[c]
Abnormal origin subclavian artery	1/23 (1/172)	1/21 (1/151)	0/22 (0/155)	9/23 (11/122)[c]
Agenesis innominate artery	0/23 (0/172)	1/21 (1/151)	0/22 (0/155)	10/23 (13/122)[c]
Skeletal observations				
Litters: skeletal variations	20/23	10/21	15/22	23/23
Sternabrae Asymmetric	0/23 (0/175)	0/21 (0/155)	2/22 (2/153)	13/23 (20/123)[c]
Bifid	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	8/23 (11/123)[c]
Hypoplastic	1/23 (1/175)	0/21 (0/155)	1/22 (1/153)	13/23 (31/123)[c]
Ribs				
Rudimentary lumbar	4/23 (6/175)	6/21 (10/155)	11/22 (27/153)[c]	23/23 (94/123)[c]
Well-formed lumbar	1/23 (1/175)	0/21 (0/155)	0/22 (0/153)	9/23 (14/123)[c]
Vertebrae				
Thoracic centra bifid	2/23 (4/174)	3/21 (3/155)	1/22 (3/153)	14/123 (9/23)[c]
Thoracic centra hypoplastic	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	5/23 (5/123)[c]
Lumbar centra dumbbell-shaped	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	5/23 (6/123)[c]

Table B.3. Significant Effects in Crl:CDBR VAF/Plus Offspring Administered DiHP Via Gavage on GDs 6–20				
	Dose (mg/kg-day)			
Endpoint	0	100	300	750
Litters: skeletal malformations	0/23	1/21	0/22	23/23[c]
Sternabrae Fused	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	20/23 (58/123)[c]
Malformed	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	11/23 (21/123)[c]
Fused ribs	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	7/23 (8/123)[c]
Vertebrae				
Thoracic centra/arch genesis	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	6/23 (7/123)[c]
Rib Anlage Fused (thoracic)	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	15/23 (26/123)[c]
Short	0/23 (0/175)	0/21 (0/155)	0/22 (0/153)	12/23 (17/123)[c]

[a]Calculated on a per litter basis.
[b]Mean ± SD.
[c]Significantly different from controls at $p < 0.05$.
[d]Number of individuals (n) is given in parentheses.
[e]Stunted growth was not included in tallies of external malformations.
[f]Incidence data could not be reported because distribution of males and females was not reported.

Sources: McKee et al. (2006); Exxon Chemical Company (1997).

The most sensitive endpoint in this study (McKee et al., 2006; Exxon Chemical Company, 1997) was increased liver weight in the dams, which occurred with a LOAEL of 300 and a NOAEL of 100 mg/kg-day. More overt effects in the dams and developmental effects occurred with a LOAEL of 750 mg/kg-day and a NOAEL of 300 mg/kg-day, including decreased maternal body and uterine weights, increased resorptions and post-implantation loss, decreased viable fetuses, decreased fetal weights, and increased incidences of external, visceral, and skeletal malformations. The increased incidence of a single skeletal variation, rudimentary lumbar ribs, in the 300 mg/kg-day fetuses is not considered to be adverse.

McKee et al. (2006); ExxonMobil Chemical Company (2003)

Groups of male and female Crl:CD(SD)IGS BR rats (30/sex/group; 6 weeks old at study initiation) were fed DiHP in the diet at 0, 1,000, 4,500, or 8,000 ppm for two generations (McKee et al., 2006; ExxonMobil Chemical Company, 2003). Dosing of F0 parents was initiated at 6 weeks of age and continued through pre-mating and mating and until necropsy (males) or pre-mating, mating, gestation, and lactation and until necropsy (females). F1 animals were administered DiHP from the time of weaning (PND 22) and continuing through the same periods of pre-mating, mating (with non-littermate animals of the opposite sex from the same treatment group), gestation, and lactation. Pre-mating periods were at least 70 days. Daily doses

of DiHP (estimated by the study authors) showed significant variation depending upon life stage; estimated doses for F0 and F1 animals are shown in Table B.4. F0 and F1 females were permitted to deliver and rear their pups until weaning on PND 21. Mortality was monitored twice daily; detailed clinical examinations were performed weekly. Body weights were also recorded weekly with the exception of females during gestation and lactation, during which time body weights were recorded more frequently (on GDs 0, 4, 7, 11, 14, 17, and 20 and lactation days 1, 4, 7, 14, and 21). Food consumption was measured on the same days in which body weights were recorded. Estrous cycling (including average cycle length) was monitored in F0 females starting 3 weeks prior to mating and until mating was detected.

Table B.4. Daily Chemical Intake for Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet

	Dietary Concentration (ppm)			
Group	**0**	**1,000**	**4,500**	**8,000**
Dose (mg/kg-day)				
F0 males				
Prior to breeding	0	81	343	623
After breeding	0	50	222	404
F1 males				
Prior to breeding	0	91	416	764
After breeding	0	50	227	419
F0 females				
Prior to breeding	0	89	406	726
Gestation period	0	64	304	532
Lactation period	0	162	716	1,289
F1 females				
Prior to breeding	0	100	462	833
Gestation period	0	64	309	543
Lactation period	0	168	750	1,360

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

On the day of birth (PND 0), pups were weighed, sexed, and examined for gross malformations (McKee et al., 2006; ExxonMobil Chemical Company, 2003). Numbers of live and dead pups/litter were recorded. On PND 4, litters were reduced at random to eight pups each (4 pups/sex when possible); remaining pups were weighed and sacrificed. Body weights were recorded and detailed clinical examinations were performed on PNDs 1, 4, 7, 14, and 21 and weekly thereafter; sexing of individual pups was determined on PNDs 4 and 21. Additional endpoints assessed in pups included AGD (on PNDs 1 and 21 and at necropsy [F1 pups] or PND 1 [F2 pups]) and thoracic nipple retention (on PNDs 11–13 [F1 pups only]). Males and females (30/group) were chosen on PND 22 to continue in the study as F1 generation adults. An

additional group of high-dose males (n=30) were assigned to the NTR group; these animals were not dosed with DiHP after weaning (PND 21). Males selected for the F1 generation were examined for balanopreputial separation (daily on PNDs 35–65 and weekly thereafter); females were examined for vaginal patency (on PND 25). Balanopreputial separation and AGD were monitored in NTR males on a regimen similar to F1 males. Sperm parameters (including motility and morphology [all males] and concentrations in the epididymis and testis [control and high-dose F0 animals and all F1 males]) were evaluated. All adults (F0 and F1 generations) were sacrificed and subjected to complete necropsy after weaning of their offspring. Organ weights (adrenal glands, brain, epididymides, kidneys, liver, ovaries, pituitary, prostate, seminal vesicles with coagulating glands, spleen, testes, thymus, and uterus with oviducts and cervix) were recorded. Histological examinations of the same tissues and including the vagina, vas deferens, and all gross (internal) lesions were performed. Counts of primordial follicles were performed from ovaries collected from 10 control and high-dose F1 females (selected at random). Unselected F1 pups and all F2 pups were also necropsied (on PND 21); selected organ weights (of the brain, spleen, and thymus) were recorded for a subset of these animals.

Significant effects in parental animals associated with DCHP treatment are summarized in Table B.5 (McKee et al., 2006; ExxonMobil Chemical Company, 2003). Mortality occurred (one control male, one low-dose female, one mid-dose male, four high-dose males, and two males in the NTR group), but was not definitively associated with treatment. No clinical signs of toxicity were observed. Body weights and body weight gains were similar among treatment and control groups during pre-mating (F0 and F1 animals), and gestation and lactation (F0 generation); however, the body weights of high-dose F1 females were reportedly decreased significantly ($p < 0.05$) throughout gestation and lactation. Based on visual inspection of the data (presented graphically), body weights for females treated at 8,000 ppm appeared to stay within ~10% of controls during gestation; data for body weights during lactation were not shown. F0 males showed no effects on sperm endpoints, and F0 females showed no adverse effects on estrous cyclicity or length of gestation. F1 males exhibited significant decreases in left testis sperm concentration and sperm production rate at ≥1,000 ppm (each decreased 39%); left cauda epididymal sperm concentration was also significantly reduced (by 44%) at 8,000 ppm (Table B.5). No effects on sperm motility or sperm morphology were observed, and numbers of primordial follicles in the ovaries of treated females were comparable with controls. External malformations of the genitalia were observed in F1 adult males of the 8,000 ppm group, including hypospadias (7/30 males) and absent (2/30 males) or undescended (2/30 males) testes. One male (of 30) in the 1,000 ppm group also had an undescended testis.

Table B.5. Significant Effects in F1 Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet

Endpoint		Dietary Level (ppm)			
		0	1,000	4,500	8,000
Parental animals					
Mating index; F1 adults[a]	M	29/30 (96.7%)	28/30 (93.3%)	27/29 (93.1%)	17/30 (56.7%)[b]
	F	29/30 (96.7%)	28/30 (93.3%)	27/29 (93.1%)	19/30 (63.3%)[b]
Fertility index; F1 adults[c]	M	24/30 (80.0%)	24/30 (80.0%)	20/29 (69.0%)	12/28 (42.9%)[b]
	F	24/30 (80.0%)	24/30 (80.0%)	20/29 (69.0%)	12/30 (40.0%)[b]
Sperm parameters; F1 adults					
Left cauda epididymis sperm concentration (10^6/g)		662.8 ± 149 (23)[d]	700 ± 225 (23)	717 ± 244 (24)	374 ± 319 (20)[b]
Left testis sperm concentration (10^6/g)		93.2 ± 14.8 (30)	56.7 ± 14.8 (21)[b]	57.6 ± 21.6 (20)[b]	49.5 ± 44.3 (28)[b]
Sperm production rate (10^6/day)		15.3 ± 2.4 (30)	9.3 ± 2.4 (21)[b]	9.4 ± 3.5 (20)[b]	8.1 ± 7.3 (28)[b]
Organ weights; F0 adults					
Relative liver weight		3.55 ± 0.28 (29)[e]	3.60 ± 0.32 (30)	3.92 ± 0.32 (30)[b]	4.15 ± 0.30 (28)[b]
		4.02 ± 0.34 (28)[f]	4.13 ± 0.21 (27)	4.50 ± 0.33 (25)[b]	4.96 ± 0.29 (28)[b]
Relative kidney weight		0.71 ± 0.05 (29)[e]	0.73 ± 0.076 (30)	0.76 ± 0.06 (30)[b]	0.82 ± 0.06 (28)[b]
		0.78 ± 0.69 (28)[f]	0.78 ± 0.05 (27)	0.84 ± 0.07 (25)[b]	0.84 ± 0.06 (28)[b]
Organ weights; F1 adults					
Relative liver weight		3.87 ± 0.40 (30)[e]	3.76 ± 0.39 (30)	4.05 ± 0.32 (29)	4.23 ± 0.29 (28)[b]
		3.70 ± 0.37 (30)[f]	3.92 ± 0.33 (30)	4.27 ± 0.36 (29)[b]	4.42 ± 0.52 (30)[b]
Relative kidney weight		0.68 ± 0.06 (30)[e]	0.68 ± 0.05 (30)	0.74 ± 0.06 (29)[b]	0.74 ± 0.05 (28)[b]
		0.74 ± 0.070 (30)[f]	0.75 ± 0.05 (30)	0.79 ± 0.06 (29)[b]	0.76 ± 0.06 (30)
Testis weight (g)					
Right		1.90 ± 0.18 (30)	1.96 ± 0.17 (30)	1.98 ± 0.25 (29)	1.30 ± 0.75 (27)[b]
Left		1.91 ± 0.19 (30)	1.93 ± 0.23 (30)	1.95 ± 0.28 (29)	1.35 ± 0.71 (28)[b]
Epididymis weight (g)					
Right		0.76 ± 0.07 (30)	0.76 ± 0.07 (30)	0.73 ± 0.10 (29)	0.47 ± 0.25 (23)[b]
Left		0.77 ± 0.10 (30)	0.76 ± 0.10 (30)	0.73 ± 0.11 (29)	0.51 ± 0.24 (24)[b]
Cauda epididymis weight (g)					
Right		0.34 ± 0.04 (30)	0.35 ± 0.03 (30)	0.31 ± 0.06 (29)	0.20 ± 0.10 (23)[b]
Left		0.35 ± 0.05 (30)	0.35 ± 0.05 (30)	0.33 ± 0.06 (29)	0.23 ± 0.10 (24)[b]
Ovary weight (g)		0.13 ± 0.02 (30)	0.12 ± 0.03 (30)	0.12 ± 0.02 (29)	0.11 ± 0.02 (30)[b]

Table B.5. Significant Effects in F1 Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet

Endpoint	Dietary Level (ppm)			
	0	1,000	4,500	8,000
Histopathology; F1 adults				
Seminiferous tubule degeneration	2/?[g]	1/?	7/29[h]	22/27[h]
Hypospermia (epididymis)	0/30	0/30	1/29	10/23[i]

[a]Male (female) mating index (%)=[number males (females) with evidence of mating]/[total number males (females) used for mating].
[c]Male fertility index (%)=[number males siring a litter]/[number males used for mating]. Female fertility index (%)=[number females confirmed pregnant]/[number females used for mating].
[d]Mean ± SD, number of animals is indicated in parentheses.
[e]Males.
[f]Females.
[g]Number affected/number examined
[h]Lesion was classified as severe in 1 of 7 mid-dose animals and 10 of 22 high-dose animals.
[b]Significantly different from controls at $p < 0.05$ (as reported by the study authors).
[i]Significantly different from controls at $p < 0.05$ based on Fisher's test performed for this analysis.

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

The study authors reported no significant differences in reproductive outcomes (mating success [≥87% of females reportedly became pregnant in all treatment groups], numbers of live offspring, or sex distribution of pups) in the F0 generation; however, impairments in both mating and fertility were apparent in high-dose animals (males and females) of the F1 generation. In the F1 generation, indices of mating and fertility were reduced 33–40% in both sexes at 8,000 ppm compared with controls (Table B.5).

At necropsy, relative weights of the liver and kidney were significantly increased (by 10–23 and 7–15%, respectively) in F0 rats (males and females) exposed to DiHP at ≥4,500 ppm compared with controls (McKee et al., 2006; ExxonMobil Chemical Company, 2003) (Table B.5). In F1 adults, relative liver weight was significantly increased in males at 8,000 ppm (9%) and in females at ≥4,500 (15–19%), and relative kidney weight was increased 9% in males exposed to DiHP at ≥4,500 ppm. Also in the F1 adults, absolute pituitary weight was reportedly increased (males only) at 8,000 ppm, and correlated with microscopic findings of *pars distalis* hypertrophy and/or hyperplasia. Cystic degeneration of the adrenal cortex was also noted in the same treatment group (incidence data were not shown). The weights of several reproductive organs were significantly affected in F1 males by treatment with DiHP at 8,000 ppm, including the testes (decreased 29–32%), epididymides (decreased 34–38%), cauda epididymides (decreased 34–41%), seminal vesicle with coagulating gland (data not shown), and prostate (data

not shown) in males, and the ovary (decreased 15%) in females. Histopathological examinations of the liver revealed minimal centrilobular hypertrophy in F0 and F1 adult males at 4,500 and 8,000 ppm, and in F0 and F1 adult females at 8,000 ppm (incidence data not shown). Hepatocellular vacuolization was noted in males (but not females) exposed to DiHP at the high dose (both generations). In the kidney, F0 and F1 adult males reportedly showed significantly increased incidences of chronic progressive nephropathy at 8,000 ppm; the study authors suggested that the onset of this disorder was magnified by DiHP treatment. Hydronephrosis (associated with observations of dilated renal pelvis) was also noted in F1 adult males exposed to DiHP at 4,500 or 8,000 ppm (incidence not reported). With respect to the reproductive organs, degeneration of the seminiferous tubules was noted in F1 adult males, primarily in the 4,500 and 8,000 ppm treatment groups (Table B.5; all data were not reported). The severity of this lesion increased in a dose-related manner, with 1 of 7 mid-dose and 10 of 22 affected high-dose F1 males having lesions classified as severe. In the epididymis, significant hypospermia, affecting 43% of examined F1 males, was observed at 8,000 ppm. Microscopic degeneration, decreased secretion, and/or absence of other parts of the reproductive tract (including the testes, prostate, seminal vesicles, seminiferous tubules, vas deferens, coagulating gland, and epididymis) were noted in F1 adult males exposed to DiHP at 8,000 ppm, but the incidences of these lesions were not quantified.

Significant effects observed in the offspring are summarized in Table B.6 (McKee et al., 2006; ExxonMobil Chemical Company, 2003). Treatment with DiHP did not affect numbers of F1 or F2 pups born, numbers of live pups/litter, or offspring survival to PND 21. In general, pup weights were lower in DiHP-exposed rats than controls during lactation; significantly decreased pup weights were reported in F1 females at 8,000 ppm group (PND 21), and in F2 males at 4,500 and 8,000 ppm and F2 females at 8,000 ppm group (PNDs 14 and 21; data not shown). AGD was significantly decreased in F1 males, but not females, exposed to DiHP at 8,000 ppm; AGD was decreased by 15% relative to controls at birth (on PND 1) and at study termination (on PND 156) in this group (AGD was not assessed at all time points in all dose groups). AGD adjusted to the cube root of body weight was also reportedly significantly decreased in high-dose F1 males (data not shown). A significant reduction in AGD (of 10%) persisted in F1 males in the NTR group on PND 156. In the F2 generation, AGD (absolute values, and values adjusted for pup weight [not shown]) were significantly decreased in males at ≥4,500 ppm. In F1 males, the incidence of retained nipples was significantly increased at 8,000 ppm (5–7% incidence, versus 0% in controls). F1 males, but not F1 females, also showed a delay in the time required to reach puberty; balanopreputial separation occurred in males exposed to DiHP at 8,000 ppm (8,000 ppm and NTR groups) 2–4 days later than non-exposed controls. Five F1 males in the

8,000 ppm group were excluded from the calculation because balanopreputial separation did not occur before study termination.

Table B.6. Significant Effects in the Offspring of Crl:CD(SD)IGS BR Rats Administered DiHP in the Diet

Endpoint	Dietary Level (ppm)				
	0	1,000	4,500	8,000	NT Group[a]
AGD (mm); F1 males					
PND 1	4.34 ± 0.34 (28)[b]	4.32 ± 0.36 (27)	4.25 ± 0.47 (25)	3.71 ± 0.41 (28)[c]	Not assessed
PND 21	18.0 ± 2.4 (30)	Not assessed	Not assessed	15.0 ± 1.7 (30)[c]	15.5 ± 1.5 (29)[c]
PND 156	48.7 ± 2.9 (29)	Not assessed	Not assessed	41.6 ± 3.6 (28)[c]	43.8 ± 3.7 (28)[c]
AGD (mm); F2 males					
PND 1	4.32 ± 0.39 (24)	4.26 ± 0.40 (23)	3.74 ± 0.36 (19)[c]	3.38 ± 0.31 (12)[c]	Not assessed
Retention thoracic nipples (%); F1 males					
Day 11	0	0	0	7.1[c]	Not assessed
Day 12	0	0	0	5.4[c]	Not assessed
Day 13	0	0	0	6.3[c]	Not assessed
Time to balanopreputial separation (days); F1 males	46.1[d]	45.4	47.4	50.3[c]	48.2[c]

[a]Animals in the "non-treated" NT group were exposed to DiHP at 8,000 ppm until the end of the lactation period and then held without treatment until terminal sacrifice (PND 156).
[b]Mean ± SD, number of animals is indicated in parentheses.
[c]Significantly different from controls at $p < 0.05$ (as reported by the study authors).
[d]Mean value as provided in text; SE or SD were not shown.

Sources: McKee et al. (2006); ExxonMobil Chemical Company (2003).

Decreased testis sperm concentration and sperm production rate were the most sensitive endpoints in this study (McKee et al., 2006; ExxonMobil Chemical Company, 2003). These effects were seen in F1 male adults at all dose levels, so that the low dose level of 1,000 ppm (50–91 mg/kg-day in this group) was a LOAEL. A NOAEL was not identified. Other effects on sexual development of male offspring were seen at 4,500 ppm (decreased AGD in F2 male pups) or 8,000 ppm (decreased AGD in F1 male pups, increased thoracic nipple retention in F1 male pups, increased time to balanopreputial separation in F1 male juveniles, decreased testis and epididymis weights and increased testicular and epididymal lesions in F1 male adults, increased incidence of malformations of the external genitalia [hypospadias and absent or undescended testes] in F1 adult males, and reduced mating and fertility of F1 adults. Increases in liver and kidney weight in F0 and F1 adults of both sexes were also seen at ≥4,500 ppm.