UNITED STATES
CONSUMER PRODUCT SAFETY COMMISSION
Bethesda, MD 20814

Memorandum

Date: October 25, 2010

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SUBJECT : Toxicity Review of **Ditridecyl phthalate (DTDP)**

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 **DTDP**.

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 DITRIDECYL PHTHALATE (DTDP)

Contract No. CPSC-D-06-0006
Task Order 012

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LIST OF ABBREVIATIONS AND ACRONYMS

ALP	Alkaline phosphatase
CIPC	Chemical Investigation Promoting Council
CPSC	Consumer Product Safety Commission
DMSO	Dimethyl sulfoxide
DTDP	Ditridecyl phthalate
FHSA	Federal Hazardous Substances Act
HMWPE	High Molecular Weight Phthalate Esters
HSDB	Hazardous Substance Data Bank
LOAEL	Lowest-observed-adverse-effect level
LD_{50}	Median lethal dose
MCH	Mean cell hemoglobin
MCHC	Mean cell hemoglobin concentration
NOAEL	No-observed-adverse-effect level

EXECUTIVE SUMMARY

DTDP is a minor use plasticizer found in a variety of consumer products.

Oral exposure to DTDP resulted in LD_{50}s > 2000 mg/kg in Sprague-Dawley rats and > 60,800 mg/kg in male Carworth-Wistar rats. Dermal exposure to DTDP resulted in an LD_{50} > 19,000 mg/kg in rabbits. Skin irritation was not reported in a human dermal exposure study. Slight dermal irritation was reported following dermal exposure to rabbits. Slight corneal necrosis was reported in a poorly described rabbit eye study. Sensitization was not reported following human or guinea pig exposure to DTDP. Insufficient data were available to make the determination of whether DTDP was associated with acute inhalation toxicity.

Sufficient animal data in one study existed to support the conclusions that DTDP had subchronic toxicity in a variety of organ systems. DTDP induced effects in the liver (hepatocellular hypertrophy and increased liver weight in both sexes) and kidney (eosinophilic bodies in renal tubular cells and increased kidney weight in males) following 6-week gavage administration. Additional findings of uncertain toxicological significance include a few observations of mild hyperplasia in the renal pelvis epithelium and urinary bladder transitional epithelium in female rats.

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.

Overall, a lack of comprehensive studies pertaining to particular organ systems or exposure durations (i.e. acute, subchronic, or chronic) prohibited the calculation of an ADI for systemic toxicity. Even though NOAELs and LOAELs could be described for a particular study, the lack of supporting studies suggests that there was "inadequate evidence" for the designation of DTDP as a "chronic hazard" when considering FHSA criteria (16 CFR §1500.135).

TOXICITY REVIEW FOR DITRIDECYL PHTHALATE (DTDP, CASRN 119-06-2)

1. INTRODUCTION

This report summarizes available data on the identity, physicochemical properties, manufacture, supply, use, toxicity, and exposure associated with ditridecyl phthalate (DTDP). This assessment was prepared from a variety of review articles (NICNAS, 2008; U.S. EPA, 2010; HSDB, 2009; ECB, 2000) 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 soft plastics, have also been a concern (NICNAS, 2008).

2. IDENTITY and PHYSICOCHEMICAL CHARACTERISTICS

This section highlights the identity and key physicochemical properties of DTDP.

DTDP is comprised of a pair of 13-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*).

DTDP is a synonym for diisotridecyl phthalate (27253-26-5) or undecyl dodecyl phthalate (CAS 68515-47-9; bis(11-methyldodecyl) phthalate), the latter of which are described as C13-rich, di-C11-14 branched alkyl esters.

DTDP is currently considered to belong to the High Molecular Weight Phthalate Esters (HMWPE) group.

The identity and physicochemical properties of DTDP can be seen in Tables 2.1 and 2.2 (NICNAS, 2008; HSDB, 2009).

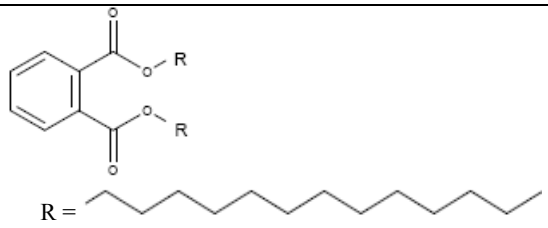
Table 2.1 Names, Structural Descriptors, and Molecular Formulas of DTDP (NICNAS, 2008)	
CAS Number:	119-06-2
Chemical Name:	1,2-Benzenedicarboxylic acid, 1,2-ditridecyl ester
Common Name	Ditridecyl phthalate (DTDP)
Molecular Formula:	C34H58O4
Structural Formula:	
Molecular Weight:	530.8 (based on a di-C13 phthalate ester)
Synonyms:	Ditridecyl phthalate; Bis(tridecyl) phthalate; Phthalic acid, ditridecyl ester.
Purity/Impurities/Additives:	Purity: >99.5% w/w Impurity: 0.1-0.3% w/w antioxidant Impurity: 0.5% ortho isomer of bisphenol A (Harris et al., 1997)

Table 2.2 Physicochemical Properties of DTDP (NCINAS, 2008)	
Property	**Value**
Physical state	Clear, viscous liquid (HSDB, 2008)
Melting point	-37°C (NICNAS,2008)
Boiling point	285ºC (3.5 mm Hg; Toxnet 2011); 501°C (101.3 kPa; HSDB, 2008; NICNAS, 2008)
Density	0.9525 g/cm^3 @ 25ºC (Toxnet, 2011); 950 kg/m^3 (HSDB, 2008; NICNAS, 2008)
Vapor pressure	2.5- 3.63 x 10^{-11} kPa (25°C; Toxnet, 2011; NICNAS, 2008)
Water solubility	1.48*10^{-9} mg/L @ 25ºC (Toxnet, 2011); 7 x 10^{-11} g/L (NICNAS, 2008)
Partition coefficient n-octanol/water (log Kow)	12.1 (NICNAS, 2008)
Henry's law constant	2.2e x 10^{-4} atm-cu m/mol (25°C; estimated; HSDB, 2008)
Flash point	470ºF (open cup; HSDB, 2008)

3. MANUFACTURE, SUPPLY, AND USE

Manufacture

In general, DTDP is manufactured commercially in a closed system by catalytically esterifying phthalic anhydride with C_{13} tridecyl-alcohols (tridecanol). As with other phthalates, the unreacted alcohols are recovered and reused, and the DTDP mixture is purified by vacuum distillation or activated charcoal. The purity of DTDP can achieve 99% or greater using current manufacturing processes (Toxnet, 2011). The remaining fraction of the DTDP commercial mixture can also contain 0.1-0.3 wt% of anti-oxidants such as 1,1,3-Tris (2-methyl-4-hydroxy-5-t-butylphenyl) butane (NICNAS, 2008; ExxonMobil, 2010) and impurities such as ortho isomers of bisphenol A (0.5%; Harris et al., 1997).

Supply

U.S. production of DTDP has been slowly increasing since the implementation of chemical tracking in 1975 (7,000 metric tons to 13-14,000 metric tons in early 2000's). Recently, production has slightly declined from 13,000 metric tons (2005) to 12,200 metric tons (2008). DTDP's proportion of the total phthalate production market has remained static at 2.1% over the past 5 years, although consultants estimate an average annual growth rate of 1.4-1.6% (Bizzari, 2007, 2009).

In the past 20 years, U.S. consumption (in metric tons) of DTDP has been within a metric ton or two less than production estimates, and currently, percentages of total phthalate consumption market are similar to production. This suggests that most DTDP produced in the U.S. is utilized locally and a small amount may be exported. Canadian imports of DTDP have historically been minimal when compared to the U.S. (0 to 1,600 metric tons; Bizzari, 2007, 2009).

Currently, ExxonMobil Chemical is the major U.S. producer of DTDP. In Mexico, two producers of DTDP (Egon Meyer, S.A. de C.V. and Especialidades Industriales y Quimicas, S.A. de C.V., 2006) have recently been reduced to just one (Egon Meyer, S.A. de C.V., 2009). Two suppliers of undecyl dodecyl phthalate exist in the U.S. (3B Scientific Corp, Scientific Polymer Products, Inc), one in Belgium (Brenntag, N.V.), and one in Norway (Exxon Mobil Chemical Corp.).

Data on the production and consumption (or import and export) of DTDP in other countries either is not available or has been combined into multi-phthalate groups, so is not useable for this report.

DTDP is listed as a high production volume chemical by the U.S. EPA (chemicals produced or imported in the U.S. in 1990 and/or 1994 (HSDB, 2009). Production volumes reported in the Hazardous Substance Data Bank (HSDB) indicate that the production range was >500 thousand - 1 million pounds in 2002. These production volumes are for non-confidential chemicals reported under the U.S. EPA Inventory Update Rule. NICNAS (2008) has reported that in Europe, the estimated production of HMWPE is approximately 60-100 ktonnes per year and represents about one third of the world production. Production volume data specific to DTDP were not reported.

Use

The high molecular weight phthalate esters are used primarily as industrial chemicals that are associated with polymers to impart flexibility in polyvinyl chloride (PVC) resins. They are also used as synthetic base stocks for industrial lubricating oils (at 10-30% in oils such as Torcula® Fluid DE 68 a synthetic oil mist lubricant with low temperature properties) and compressor fluids (NICNAS, 2008). DTDP is generally used in wiring and cable jacketing and insulation in the automotive and building industries because of its low volatility and high permanence (NICNAS, 2008). In fact, data show that DTDP has a low migration rate (low mobility) from finished products when compared to DOP, DINP, or DIDP at temperatures 30 - 80ºC (Exxon Mobil, 2010). According to Godwin (2010), DTDP has the desired plasticizer attributes such as low volatility and conductivity, and resistance to oil and grease. DTDP has also been proposed for use in fly fishing line (http://www.freepatentsonline.com/5207732.html) and liquid correction fluid (2.5-2.8 wt%; SureChem, 2011). DTDP can be blended with trimellitates when high-temperature ratings are required.

4. TOXICOKINETICS

No toxicokinetic data were located for DTDP.

5. HAZARD INFORMATION

This section contains brief hazard summaries of the adverse effects of DTDP 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	—

Oral exposure to DTDP resulted in LD_{50}s > 2000 mg/kg in Sprague-Dawley rats and > 60,800 mg/kg in male Carworth-Wistar rats. Dermal exposure to DTDP resulted in an LD_{50} > 19,000 mg/kg in rabbits. Skin irritation was not reported in a human dermal exposure study. Slight dermal irritation was reported following dermal exposure to rabbits. Slight corneal necrosis was reported in a poorly described rabbit eye study. Sensitization was not reported following human or guinea pig exposure to DTDP. Insufficient data were available to make the determination of whether DTDP was associated with acute inhalation toxicity.

Evidence supported the conclusion that DTDP was a subchronic toxicant. Exposure to DTDP induced decrements in body weight, changes in blood chemistry, increases in liver and kidney weight, and adverse changes in kidney and bladder histopathology following subchronic administration.

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 DTDP relevant exposure durations for the general population or for other sensitive subpopulations because confirmatory data on

toxicological endpoints and discrete methodological details of the reporting study 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

No treatment-related deaths, clinical signs of toxicity, body weight changes, or autopsy findings were observed in a single-dose toxicity study of male (n=5) and female (n=5) Sprague-Dawley rats administered DTDP (purity 93.7–100%) via gavage (in corn oil) at 2,000 mg/kg and observed for 14 days following dosing (CIPC, 2010a, b). An earlier study reported an acute oral LD_{50} value of >64 mL/kg (>60,800 mg/kg using the reported density of 950 kg/m^3 for DTDP [NICNAS, 2008]) in male Carworth-Wistar rats (n=5 per dose level, number of dose levels tested unspecified) given a single gavage dose of undiluted DTDP and observed for 14 days (Smyth et al., 1962).

Sufficient methodological details were provided in these studies to consider them acceptable for use. The estimated LD_{50} from the Smyth et al. (1962) study was substantially higher than the oral LD_{50} range (50–5,000 mg/kg) required by the FHSA to conclude that a chemical is acutely toxic. In addition, the Chemical Investigation Promoting Council (CIPC) study reported no mortalities at 2,000 mg/kg, the highest dose tested. The weight of evidence including probable animal data are sufficient, therefore, to support the conclusion that DTDP 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

Smyth et al. (1962) reported an acute dermal LD_{50} value of >20 mL/kg (>19,000 mg/kg using the reported density of 950 kg/m^3 for DTDP [NICNAS, 2008]) in male albino New Zealand rabbits (n=4 per dose level, number of dose levels tested unspecified) given a 24-hour occluded dermal exposure to undiluted DTDP on clipped skin and observed for 14 days.

Sufficient methodological details were provided in this study to support the conclusion that DTDP 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

CPSC and contract staff did not find any information regarding the acute inhalation toxicity of DTDP.

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

5.4. Primary Skin Irritation

In a repeated insult patch test, 104 human subjects were tested concurrently for dermal responses to DTDP and six other phthalate esters (Medeiros et al., 1999). The induction phase consisted of 24-hour occluded applications of each phthalate ester to its own unique site on the forearm; induction applications were repeated 3 times/week for a total of nine applications. Dermal reactions were scored 48 or 72 hours after each induction application. There was no evidence of a dermal irritation response to any of the phthalate esters during induction.

Smyth et al. (1962) reported a primary dermal irritation grade of 2/10 in albino rabbits (n=5) following 24 hours of uncovered exposure of clipped skin to 0.01 mL undiluted DTDP. This grade of symptoms was supported by minor visible capillary injection, essentially a minimum of dermal irritation.

Dermal irritation was not noted in a human study and minimal dermal irritation was noted in an animal study. The estimated "scores" from these studies are expected not to exceed five, 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 supported the conclusion that DTDP did not fit the definition of "corrosive" as outlined in the FHSA (16 CFR §1500.3(c)(3)) or a "primary irritant" when considering FHSA criteria (16 CFR §1500.3(c)(4)).

5.5. Primary Eye Irritation

Smyth et al. (1962) reported a primary eye irritation grade of 2/10 in rabbits following ocular instillation of 0.5 mL undiluted DTDP. This grade of symptoms suggests the occurrence of slight corneal necrosis.

Even though slight corneal necrosis was reported in the one study reviewed, the lack of additional information on the ocular properties of DTDP can be considered a data gap and supports the conclusion that there is "inadequate evidence" for the designation of DTDP as a "primary irritant" or "corrosive" under the FHSA (16 CFR §1500.3(c)(3) and 16 CFR §1500.3(c)(4)), respectively.

5.6. Sensitization

In the repeated insult patch test of Medeiros et al. (1999), the induction phase, described in Section 5.4 above, was followed by a rest period of 10–17 days and a challenge phase that consisted of 24-hour applications of each phthalate ester to its own naïve site; dermal reactions were scored at 48 and 96 hours post-application. There was no evidence of a sensitization response during the challenge phase of this study.

DTDP did not induce a sensitization response in a guinea pig maximization test using the Buehler method (as reported in Medeiros et al., 1999). No further details of this study were provided.

A sufficient weight of human and animal evidence suggests that DTDP does not fit the definition of a "strong sensitizer" as defined in the FHSA (16 CFR §1500.3(c)(5)).

REPEAT DOSE TOXICITY

DTDP exposure resulted in decrements in female body weight and body weight gain and induced effects on the liver (hepatocellular hypertrophy and increased liver weight in both sexes) and kidney (eosinophilic bodies in renal tubular cells and increased kidney weight in males) following 6-week gavage administration. Additional findings of uncertain toxicological significance include a few observations of mild hyperplasia in the renal pelvis epithelium and urinary bladder transitional epithelium in female rats. This information supports the conclusion that DTDP has adverse subchronic effects. Overall, a lack of comprehensive studies pertaining to particular organ systems or exposure durations (i.e. acute, subchronic, or chronic) prohibited the calculation of an ADI for systemic toxicity. Even though NOAELs and LOAELs could be described for a particular study, the lack of supporting studies suggests that there was "inadequate evidence" for the designation of DTDP as a "chronic hazard" when considering FHSA criteria (16 CFR §1500.135).

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

In the only available repeated-dose animal study (a combined repeated-dose oral gavage toxicity and reproductive/developmental toxicity screening test in rats) (CIPC, 2010b, c), no deaths were observed, even at the high dose of 250 mg/kg-day. Increased salivation was observed following dosing in 1/13 of the 250 mg/kg-day males during treatment weeks 3 and 5 and in 7/13 of the 250 mg/kg-day males and 2/13 of the 50 mg/kg-day males during treatment week 6. No other clinical signs of toxicity related to DTDP treatment were observed in this study.

Females in the 50 and 250 mg/kg-day groups showed significant decreases in body weight gain over the 15-day pre-mating period in relation to controls (Table 5.2). On day 15, at the end of the pre-mating period, body weight was significantly reduced relative to controls in the 250 mg/kg-day females (-5.0%). Mean body weight in the 50 mg/kg-day group was also lower than controls (-3.6%), but the difference was not statistically significant (Table 5.2). There were no significant treatment-related effects on mean maternal body weight or body weight gain during gestation or lactation (the study was continued to the fourth day of lactation). Body weight and body weight gain in males were not affected by exposure to DTDP. There were no effects of DTDP on food consumption in males or females (CIPC, 2010b, c).

Table 5.2. Body Weight Data for Female Sprague-Dawley Rats Administered DTDP by Gavage Prior to Mating				
Dose (mg/kg-day)	**0**	**10**	**50**	**250**
Body weight gain (g)				
Treatment days 1–8	20.2 ± 6.3[a]	16.2 ± 6.0	12.8 ± 7.4[b]	13.6 ± 8.1
Treatment days 8–15	17.2 ± 3.8	15.2 ± 6.5	15.2 ± 5.6	11.4 ± 6.1
Treatment days 1–15	37.3 ± 8.7	31.3 ± 9.3	28.0 ± 6.6[b]	25.0 ± 10.4[c]
Body weight (g)				
Treatment day 1	208.7 ± 6.4	209.3 ± 6.5	208.9 ± 6.1	208.6 ± 6.3
Treatment day 8	228.9 ± 7.5	225.5 ± 10.2	221.8 ± 9.1	222.1 ± 8.8
Treatment day 15	246.0 ± 8.6	240.6 ± 12.3	237.0 ± 10.1	233.6 ± 11.9[b]

[a]Mean ± standard deviation; n=13.
[b]Significantly different from control, $p < 0.05$ (adjusted for multiple comparisons).
[c]Significantly different from control, $p < 0.01$ (adjusted for multiple comparisons).

Source: CIPC (2010b, c).

5.8. Hematology

Hematology results in the combined repeated-dose oral toxicity and reproductive/toxicity screening test (CIPC, 2010b, c) were reported only for male rats. There were no significant changes in red blood cell count, hemoglobin, hematocrit, platelets, or white blood cell count or differential. There were slight, statistically significant reductions in calculated mean cell hemoglobin (MCH; -3.7%) and mean cell hemoglobin concentration (MCHC; -1.5%) in the 250 mg/kg-day group (Table 5.3), but the toxicological significance of these changes in the absence of changes in the measured variables from which they are derived is questionable.

Table 5.3. Selected Results of Hematology Evaluations for Male Sprague-Dawley Rats Exposed to DTDP by Gavage for 42 Days				
Dose (mg/kg-day)	**0**	**10**	**50**	**250**
MCH (pg)	19.7 ± 0.5[a]	19.2 ± 0.6	19.4 ± 0.5	19.0 ± 0.5[b]
MCHC (%)	34.1 ± 0.4	34.0 ± 0.6	33.8 ± 0.4	33.6 ± 0.6[c]

[a]Mean ± standard deviation; n=13.
[b]Significantly different from control, $p < 0.01$ (statistical test not specified in Table 17 of the original study report).
[c]Significantly different from control, $p < 0.05$ (statistical test not specified in Table 17 of the original study report).

Source: CIPC (2010c).

5.9. Thymus

Gross pathologic examinations in the CIPC (2010b, c) study revealed what was described as "small thymus" in 2/13, 5/13, 5/13, and 8/13 female rats in the control, low-, mid-, and high-dose groups, respectively. Absolute and relative thymus weights for all exposure groups were less than control weights. Decrements were not statistically significant from control weights or visibly dose-related. The incidence and severity of thymic atrophy was marginally increased over controls, although these differences were not statistically significant or visibly dose-related. In the males, "small thymus" was reported in 2/13 animals from the mid-dose group, but no animals in the control, low-, or high-dose groups. Also in the males, absolute and relative thymus weights in the treated groups did not differ from controls, and histopathological thymus lesions were not observed at any dose (CIPC, 2010b, c).

5.10. Hepatotoxicity

The CIPC (2010b, c) study found evidence of liver effects of DTDP in both male and female rats (Table 5.4). Relative, but not absolute, liver weight was significantly increased in males at 250 mg/kg-day (+17%) and in females at 50 (+10%) and 250 mg/kg-day (+15%). Gross pathological examinations revealed no effects in males, but 2/13 females at 250 mg/kg-day had enlarged livers. Histological examinations revealed centrilobular hepatocellular hypertrophy in the livers of mid- and high-dose male and female rats; incidences were statistically significantly increased in males at 250 mg/kg-day and females at 50 and 250 mg/kg-day. Also in the liver in males, there was a significant decrease in incidence/severity of periportal fatty change. This change was not seen in females. Judging by the results reported, it appears that assays for liver catalase activity were conducted in only two males per group and no females. A "very slight" increase in incidence and size of catalase positive granules was reported in the two high-dose males tested. Serum chemistry evaluations found a significant increase in serum alkaline phosphatase (ALP) activity in the 250 mg/kg-day males (21% higher than that of controls) that may reflect an effect on the liver.

The lowest-observed-adverse-effect level (LOAEL) for effects on the liver in this study (CIPC, 2010b, c) is 50 mg/kg-day based on increased relative liver weight and increased incidence of centrilobular hepatocellular hypertrophy in female rats. The corresponding no-observed-adverse-effect level (NOAEL) for liver effects in female rats is 10 mg/kg-day. Male rats showed the same liver effects as the females, but were somewhat less sensitive.

Table 5.4. Liver Effects in Sprague-Dawley Rats Administered DTDP by Gavage in a Combined Repeated-Dose Toxicity and Reproductive/Developmental Toxicity Screening Test

Dose (mg/kg-day)	**0**						**10**						**50**						**250**					
Males																								
Serum chemistry																								
ALP (U/L)	201 ± 39[a]						181 ± 47						206 ± 33						243 ± 51[d]					
Liver weight																								
Final body weight (g)	501.7 ± 30.7						509.6 ± 32.1						498.9 ± 28.3						496.0 ± 56.1					
Absolute liver weight (g)	14.47 ± 1.32						14.48 ± 1.57						14.67 ± 1.36						16.86 ± 2.85					
Relative liver weight (g per 100 g body weight)	2.88 ± 0.17						2.84 ± 0.18						2.94 ± 0.16						3.38 ± 0.22[e]					
Histological lesions[b]	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**
Hypertrophy, hepatocyte, centrilobular	13	0	0	0	0	0	13	0	0	0	0	0	10	3	0	0	0	3	2	6	5	0	0	11[f,g]
Fatty change, periportal	0	2	3	8	0	13	0	0	4	9	0	13	0	1	5	7	0	13	3	7	3	0	0	10[g]
Increased/elongated catalase-positive granules[c]	2	0	0	0	0	0	2	0	0	0	0	0	2	0	0	0	0	0	0	2	0	0	0	2[g]
Females																								
Liver weight																								
Final body weight (g)	313.7 ± 24.9						307.2 ± 22.2						291.0 ± 16.8[h] (12)						296.7 ± 17.4 (11)					
Absolute liver weight (g)	13.16 ± 1.19						13.48 ± 1.23						13.50 ± 1.30 (12)						14.34 ± 1.08 (11)					
Relative liver weight (g per 100 g body weight)	4.20 ± 0.28						4.40 ± 0.37						4.63 ± 0.29[e] (12)						4.83 ± 0.19[e] (11)					
Gross lesions[b]	**N**			**P**			**N**			**P**			**N**			**P**			**N**			**P**		
Liver enlargement	13			0			13			0			13			0			11			2		
Histological lesions[b]	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**
Hypertrophy, hepatocyte, centrilobular	13	0	0	0	0	0	13	0	0	0	0	0	9	4	0	0	0	4[i]	0	9	4	0	0	13[f,g]

[a]Mean ± standard deviation; n=13 unless indicated otherwise in ().
[b]Severity of lesion: N = no lesion, 1 = very slight, 2 = slight, 3 = moderate, 4 = severe, P = the total number of positive responders (i.e., animals exhibiting severity grade 1, 2, 3, or 4 for the selected lesion).
[c]It appears from the data presented that only 2 males per group were evaluated for catalase activity.
[d]Significantly different from control, $p < 0.05$ (statistical test not specified in Table 18 of the original study report).
[e]Significantly different from control, $p < 0.01$ (adjusted for multiple comparisons).
[f]Significantly different from control, $p < 0.01$ (Fisher's exact test).
[g]Significantly different from control, $p < 0.01$ (Mann-Whitney U test).
[h]Significantly different from control, $p < 0.05$ (adjusted for multiple comparisons).
[i]Significantly different from control, $p < 0.05$ (Fisher exact test).

Source: CIPC (2010b, c).

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

5.11. Renal Toxicity

There is some evidence of renal effects of DTDP in the CIPC (2010b, c) study (Table 5.5). Absolute, but not relative, kidney weight was significantly increased in males at 250 mg/kg-day and gross pathology examination revealed kidney enlargement in 3/13 males from this same group. Histologically, there was a significant increase in the incidence and severity of eosinophilic bodies in renal tubular cells ("regeneration foci") in the 250 mg/kg-day males. None of these changes were seen in females, although one high-dose female had "slight" hyperplasia in the renal pelvic epithelium.

Table 5.5. Kidney Effects in Sprague-Dawley Rats Administered DTDP by Gavage in a Combined Repeated-Dose Toxicity and Reproductive/Developmental Toxicity Screening Test

Dose (mg/kg-day)	**0**						**10**						**50**						**250**					
Males																								
Serum chemistry																								
Potassium (mEq/L)	3.95 ± 0.24[a]						3.76 ± 0.11[c]						3.85 ± 0.19						3.69 ± 0.19[d]					
Kidney weight																								
Necropsy body weight (g)	501.7 ± 30.7						509.6 ± 32.1						498.9 ± 28.3						496.0 ± 56.1					
Absolute kidney weight (g)	3.00 ± 0.19						2.92 ± 0.19						3.07 ± 0.24						3.28 ± 0.35[e]					
Relative kidney weight (g per 100 g body weight)	0.60 ± 0.05						0.57 ± 0.03						0.62 ± 0.04						0.67 ± 0.09					
Gross lesions[b]	**N**			**P**			**N**			**P**			**N**			**P**			**N**			**P**		
Kidney enlargement	13			0			13			0			13			0			10			3		
Histological lesions[b]	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**
Eosinophilic bodies, tubular cells	10	0	1	2	0	3	11	1	1	0	0	2	8	3	1	1	0	5	4	2	4	2	1	9
Females																								
Kidney weight																								
Final body weight g)	313.7 ± 24.9						307.2 ± 22.2						291.0 ± 16.8[e] (12)						296.7 ± 17.4 (11)					
Absolute kidney weight (g)	1.80 ± 0.21						1.87 ± 0.20						1.75 ± 0.12 (12)						1.85 ± 0.15 (11)					
Relative kidney weight (g per 100 g body weight)	0.57 ± 0.04						0.61 ± 0.04						0.60 ± 0.05 (12)						0.62 ± 0.05 (11)					
Histological lesions[b]	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**
Hyperplasia, pelvic epithelium	13	0	0	0	0	0	13	0	0	0	0	0	13	0	0	0	0	0	12	0	1	0	0	1

[a]Mean ± standard deviation; n=13 unless indicated otherwise in ().
[b]Severity of lesion: N = no lesion, 1 = very slight, 2 = slight, 3 = moderate, 4 = severe, P = the total number of positive responders (i.e., animals exhibiting severity grade 1, 2, 3, or 4 for the selected lesion).
[c]Significantly different from control, $p < 0.05$ (statistical test not specified in Table 18 of the original study report).
[d]Significantly different from control, $p < 0.01$ (statistical test not specified in Table 18 of the original study report).
[e]Significantly different from control, $p < 0.05$ (adjusted for multiple comparisons).
[f]Significantly different from control, $p < 0.05$ (Fisher's exact test).

Source: CIPC (2010b, c).

Serum chemistry evaluations in the males rats showed significantly reduced serum potassium levels in the low- and high- (but not mid-) dose males. The lack of a clear dose-response suggests that this change was likely not related to DTDP treatment. The incidence of males with elevated urine pH was high in all groups, including controls, but there were no treatment-related effects on urinalysis in either males or females.

These data identify the kidney as a target for DTDP in male rats, with a LOAEL of 250 mg/kg-day and NOAEL of 50 mg/kg-day based on increased kidney weight and tubular lesions. The toxicological significance of the finding of slight renal pelvic hyperplasia in one high-dose female is uncertain.

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

5.12. Urinary Bladder Toxicity

Transitional cell hyperplasia of "very slight" severity was observed by CIPC (2010b, c) in the urinary bladder of 2/13 high-dose female rats; this lesion was not observed at any severity in control, low-, or mid-dose females or males at any dose. The toxicological significance of this finding in two high-dose female rats is uncertain.

5.13. Endocrine Activity

Estrous cyclicity was not affected in female Sprague-Dawley rats administered DTDP by gavage at doses up to 250 mg/kg-day in the previously-described combined repeated-dose toxicity and reproductive/developmental toxicity screening test (CIPC, 2010b, c). Harris et al. (1997) assessed the estrogenic activity of DTDP and other phthalate esters in an *in vitro* screening test in recombinant yeast cells in which the human estrogen receptor had been

integrated into the yeast genome and in assays for stimulation of proliferation in human breast cancer cell lines MCF-7 and ZR-75. Although weakly positive results were reported for the original sample of DTDP tested, this was subsequently determined by the researchers to be a result of contamination with bisphenol A. Results were negative for uncontaminated DTDP, and the researchers concluded that DTDP was not estrogenic in this study.

5.14. Reproductive Toxicity

In the combined repeated-dose toxicity and reproductive/developmental toxicity *screening* test (CIPC, 2010b, c), there were no significant treatment-related effects on estrous cyclicity, mating, fertility, corpora lutea, implantation sites, or gestation in the dams, and no effects on number born, sex ratio, body weight, or viability through post-natal day 4 in the pups.

A statistically significant decrease in live birth index ([number of live pups on day 0 / number of pups born] × 100) was reported in the 250 mg/kg-day group compared to controls (87.7 ± 28.4 versus 99.6 ± 1.6 for controls; $p < 0.05$, including adjustment for multiple comparisons according to the study authors). However, independent analysis of the live birth index results from the study report using an unpaired t-test did not confirm that the difference was statistically significant (p=0.14). This result is also of questionable toxicological significance because neither of the measured values contributing to the index (number of live pups on day 0 and number of pups born) differed significantly from controls.

The available English summary (CIPC, 2010b) of this Japanese study report (CIPC, 2010c) stated that poor lactation was observed in the 250 mg/kg-day dose group; however, additional details and supporting data were not provided. There was no apparent effect on body weight or viability of the pups in this group.

Organ weight measurements and gross and histopathological evaluations of the testes and epididymides from the males and ovaries from the females (pathology only) revealed no evidence of DTDP treatment-related effects. Gross pathology examination revealed an epididymal nodule in 1/13 high-dose male rats, but no control or lower-dose males. Histopathological evaluation revealed spermatic granuloma in both a control and a high dose rat. The incidence for this lesion is not considered to be related to DTDP treatment. The researchers concluded that no testicular toxicity was observed in any group (CIPC, 2010b).

The researchers identified the high-dose of 250 mg/kg-day as a NOAEL for reproductive effects in males in this study. They identified 50 mg/kg-day as a NOAEL for reproductive effects in females, apparently based on the reported poor lactation in the 250 mg/kg-day females, although this was not explicitly stated in the available English language summary.

The lack of comprehensive reproductive toxicity studies using DTDP as a test substance supported the conclusion that there was "inadequate evidence" for the designation of DTDP as a "reproductive toxicant".

5.15. Prenatal, Perinatal, and Post-natal Toxicity

The CIPC (2010b, c) developmental toxicity screening study found no significant treatment-related effects on pup number born, sex ratio, body weight, viability or external/malformations through post-natal day 4. The study did not include assessment of pups for possible treatment-related effects on developmental endpoints from post-partum day 4 through adulthood.

As discussed in Section 5.14 above, a small decrease in live birth index was reported to be statistically significant, but could not be verified independently and was considered to be of questionable toxicological significance. The researchers considered the high dose of 250 mg/kg-day to be a NOAEL for developmental effects in pups in this study.

The lack of comprehensive developmental toxicity studies using DTDP as a test substance supported the conclusion that there was "inadequate evidence" for the designation of DTDP as a "developmental toxicant".

5.16. Carcinogenicity

The lack of comprehensive carcinogenicity, genotoxicity, or initiation/promotion studies using only DTDP as a test substance supported the conclusion that there was "inadequate evidence" for the designation of DTDP as a "carcinogen."

Genotoxicity

DTDP was not mutagenic in *Salmonella typhimurium* strains TA98, TA100, TA1535, or TA1537 or *Escherichia coli* WP2 uvrA with or without exogenous metabolic activation in two

studies conducted using the preincubation modification of the Ames test (CIPC, 2010b, d; Zeiger et al., 1985, 1982). The CIPC (2010b, d) study used dimethyl sulfoxide (DMSO) as the solvent, and tested six concentrations ranging from 156 to 5,000 µg/plate, in addition to negative and positive controls. Visible precipitate was reported at the end of the exposure period at the three highest concentrations (1,250, 2500, 5,000 µg/plate), and growth inhibition was reported in TA1537 at 5,000 µg/plate. When assessing the three non-precipitating test concentrations (156, 313, 625 µg/plate), no statistically significant changes in the mean number of revertants were observed for any strain. The Zeiger et al. (1985, 1982) study tested DTDP in acetone and included five concentrations ranging from 100 to 10,000 µg/plate in addition to negative and positive controls. No precipitate or toxicity was reported.

DTDP did not induce chromosomal aberrations in Chinese hamster lung cells in vitro with or without exogenous metabolic activation (CIPC, 2010b, e). The study used DMSO as the solvent and included three DTDP exposure levels (1,188–4,750 µg/mL) in addition to negative and positive controls. A visible precipitate was present at the end of the exposure period at all DTDP concentrations tested, but not the blank or positive controls (CIPC, 2010e). Presence of a precipitate in all DTDP groups complicates the interpretation of data from this study.

Initiation and Promotion

No initiation or promotion studies were located for DTDP.

Carcinogenicity Studies

No carcinogenicity or chronic studies were located for DTDP.

Refer to Section 5.11 for information regarding renal pelvis epithelial hyperplasia and Section 5.12 for information regarding urinary bladder transitional cell hyperplasia in female rats administered DTDP by gavage in the CIPC (2010b, c) study.

6. EXPOSURE

Exposure to HMWPEs is believed to be primarily in the workplaces where manufactured. The primary workplace exposure in manufacturing activities would be dermal and may be potential for formation of aerosol during some applications (OECD, 2004). Because HMWPEs are handled only in industrial manufacturing facilities, minimal consumer exposure is expected

(OECD, 2004). The consumer is exposed indirectly through use of the products that may contain the HMWPEs and uptake is expected to be low (OECD, 2004). Exposure data specific to DTDP were not found.

7. DISCUSSION

Appendix A provides a summary of the NOAELs and LOAELs for organ-specific endpoints for DTDP, most of which were derived from the oral combined repeated-dose toxicity and reproductive/developmental toxicity screening test in rats (CIPC, 2010c).

Overall Uncertainty

The hazard database for DTDP consisted of one subchronic toxicity study and other studies of various duration, all of which were not well described (or in a foreign language).

There is considerable uncertainty in the NOAEL and LOAEL values due to limitations in study design, including short duration for systemic endpoints (6 weeks) and screening-level assessment of reproductive and developmental effects (small group sizes, limited endpoints, no assessment of reproductive performance in F_1 pups, no assessment of pup development from lactation day 4 onward) with no consideration of endpoints known to be affected by other phthalates (e.g., anogenital distance, etc.). These limitations must be taken into account when comparing repeated-dose, reproductive, and developmental effects for DTDP with similar endpoints for other phthalate esters.

Overall Acceptable Daily Intakes

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 DTDP relevant exposure durations for the general population or for other sensitive subpopulations because confirmatory data on toxicological endpoints and discrete methodological details of the reporting study were not available.

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Appendix A. Summary of Endpoints by Organ System

Species (Gender)	Exposure Route	Dose (Number of Animals per Dose Group)	Dose Duration	Toxicological Endpoint	Toxicological Basis	Citation	Effect Category
Sprague-Dawley rat (M&F)	Oral gavage in corn oil	0, 2,000 mg/kg (5/group)	Once	NOAEL=2,000 mg/kg	No deaths	CIPC, 2010a, b	Mortality
				NOAEL=2,000 mg/kg	No clinical signs of toxicity, body weight changes, or autopsy findings		General
Sprague-Dawley rat (M&F)	Oral gavage in corn oil	0, 10, 50, 250 mg/kg-day (13 M, 13 F per group)	(M) Once daily for 6 weeks (F) Once daily for 14 days premating to PND 4 (~6 weeks)	NOAEL=250 mg/kg-day	No deaths	CIPC, 2010b, c	Mortality
				NOAEL=250 mg/kg-day	**Small decreases in body weight in females during the pre-mating period not considered toxicologically significant**		General
				NOAEL=250 mg/kg-day	No toxicologically significant effects		Hematology
				NOAEL=250 mg/kg-day	No toxicologically significant effects		Thymus
				NOAEL=10 mg/kg-day LOAEL=50 mg/kg-day	Increased liver weight; centrilobular hepatocellular hypertrophy		Liver
				NOAEL=50 mg/kg-day LOAEL=250 mg/kg-day	Increased kidney weight and eosinophilic bodies in renal tubular cells (males only); "slight" renal pelvic hyperplasia in one high-dose female is of uncertain toxicological significance		Kidney
				NOAEL=250 mg/kg-day	"Very slight" hyperplasia of transitional cell epithelium in two high-dose female is of uncertain toxicological significance		Urinary bladder
				NOAEL=250 mg/kg-day	No toxicologically significant effects. Study authors considered the high dose of 250 mg/kg-day to be a LOAEL for females, apparently based on "poor lactation", but provided no supporting data.		Reproduction
				NOAEL=250 mg/kg-day	No toxicologically significant effects		Development/ fetus

Appendix B. Critical Study Reviews

CIPC (2010b, c)

The oral toxicity of DTDP was assessed in a combined repeated-dose toxicity and reproductive/developmental toxicity screening test (CIPC, 2010b, c). Groups of male and female Sprague-Dawley rats (13 per sex per group) were administered DTDP (purity 93.7–100%) by gavage in corn oil at 0, 10, 50, or 250 mg/kg-day. Males were treated for 42 days (6 weeks) and sacrificed on study day 43; females were treated from 14 days prior to mating through lactation day 3 and sacrificed on day 4 of lactation. The animals were monitored for clinical signs of toxicity, body weight, and food consumption during the in-life portion of the study. For the reproduction/developmental toxicity screen, females were monitored for frequency of estrus, mated pairs were observed for copulations and fertility, pregnant females were observed for reproductive success, and pups were evaluated for body weight, viability, and external and visceral malformations through day 4 of lactation.

Blood was collected from males on day 42 of the study for hematology (red blood cell count, hemoglobin, hematocrit, platelets, white blood cell count and differential, mean cell volume, MCH, and MCHC) and serum chemistry (alanine aminotransferase, aspartate aminotransferase, ALP, gamma glutamyl transferase, bilirubin, blood urea nitrogen, creatinine, cholesterol, triglyceride, glucose, protein, and electrolytes) analyses. Urine was collected for urinalysis (pH, protein, glucose, ketone, bilirubin, occult blood, and urobilinogen) from males on day 42 of the study and from females on day 14 of pregnancy. All animals were necropsied upon sacrifice. The liver, kidneys, adrenals, and thymus were weighed in both sexes; the testes and epididymides were weighed in males. The brain, heart, liver, spleen, thymus, kidney, urinary bladder, adrenal gland, and selected sex organs (testes and epididymides for males, ovaries for females) were examined for histopathology.

All control and DTDP-treated rats survived until terminal sacrifice. Salivation following dosing was observed in 1/13 of the 250 mg/kg-day males during treatment weeks 3 and 5 and in 2/13 of the 50 mg/kg-day males and 7/13 of the 250 mg/kg-day males during treatment week 6. No other clinical signs of toxicity related to treatment were observed. There were no effects on food consumption in males or females, or body weight or body weight gain in males. Females in the 50 and 250 mg/kg-day groups showed significant decreases in body weight gain over the 15-day pre-mating period in relation to controls (Table B.1). On day 15, at the end of the pre-mating period, body weight was significantly reduced relative to controls in the 250 mg/kg-day females (-5.0%). Mean body weight in the 50 mg/kg-day group was also lower than controls

(-3.6%), but the difference was not statistically significant (Table B.1). There were no significant treatment-related effects on mean maternal body weight or body weight gain during gestation or the first 3 days of lactation.

Table B.1. Body Weight Data for Female Sprague-Dawley Rats Administered DTDP by Gavage During 15 Days of Pre-Mating Treatment in the Combined Repeated-Dose Toxicity and Reproductive/Developmental Toxicity Study

Dose (mg/kg-day)	0	10	50	250
Body weight gain (g)				
Treatment days 1–8	20.2 ± 6.3[a]	16.2 ± 6.0	12.8 ± 7.4[b]	13.6 ± 8.1
Treatment days 8–15	17.2 ± 3.8	15.2 ± 6.5	15.2 ± 5.6	11.4 ± 6.1
Treatment days 1–15	37.3 ± 8.7	31.3 ± 9.3	28.0 ± 6.6[b]	25.0 ± 10.4[c]
Body weight (g)				
Treatment day 1	208.7 ± 6.4	209.3 ± 6.5	208.9 ± 6.1	208.6 ± 6.3
Treatment day 8	228.9 ± 7.5	225.5 ± 10.2	221.8 ± 9.1	222.1 ± 8.8
Treatment day 15	246.0 ± 8.6	240.6 ± 12.3	237.0 ± 10.1	233.6 ± 11.9[b]

[a]Mean ± standard deviation; n=13.
[b]Significantly different from control, $p < 0.05$ (adjusted for multiple comparisons).
[c]Significantly different from control, $p < 0.01$ (adjusted for multiple comparisons).

Source: CIPC (2010b, c).

Hematology and serum chemistry results were reported only for males. Hematological evaluations found no significant changes in red blood cell count, hemoglobin, hematocrit, platelets, or white blood cell count or differential. There were slight, statistically significant reductions in calculated MCH (-3.7%) and MCHC (-1.5%) in the 250 mg/kg-day group (Table B.2), but the toxicological significance of these changes in the absence of changes in the measured variables from which they are derived is questionable. Serum chemistry changes were limited to a significant increase in ALP in the 250 mg/kg-day group (+21%) and decreases in potassium in the 10 and 250 mg/kg-day groups (-5–6%), but not in the 50 mg/kg-day group (Table B.2). The incidence of males with elevated urine pH was high in all groups, including controls. There were no treatment-related effects on urinalysis in either males or females.

Table B.2. Selected Results of Hematology and Serum Chemistry Evaluations for Male Sprague-Dawley Rats Exposed to DTDP by Gavage for 42 Days

Dose (mg/kg-day)	0	10	50	250
Hematology				
MCH (pg)	19.7 ± 0.6[a]	19.2 ± 0.6	19.4 ± 0.5	19.0 ± 0.5[b]
MCHC (%)	34.1 ± 0.4	34.0 ± 0.6	33.8 ± 0.4	33.6 ± 0.6[c]
Clinical chemistry				
Potassium (mEq/L)	3.95 ± 0.24	3.76 ± 0.11[c]	3.85 ± 0.19	3.69 ± 0.19[b]
ALP (U/L)	201 ± 39	181 ± 47	206 ± 33	243 ± 51[c]

[a]Mean ± standard deviation; n=13.
[b]Significantly different from control, $p < 0.01$ (statistical test not specified in Table 17 or 18 of the original study report).
[c]Significantly different from control, $p < 0.05$ (statistical test not specified in Table 17 or 18 of the original study report).

Source: CIPC (2010c).

Organ weight changes were observed only in the liver and kidney. These data are summarized in Table B.3. Relative, but not absolute, liver weight was significantly increased in males at 250 mg/kg-day (+17%) and in females at 50 (+10%) and 250 mg/kg-day (+15%). Absolute kidney weight was significantly increased in males at 250 mg/kg-day, but there was no change in relative weight and no corresponding change in females.

Table B.3. Mean Terminal Body Weights and Absolute and Relative Weights of Liver and Kidney from Sprague-Dawley Rats Administered DTDP by Gavage in the Combined Repeated-Dose Toxicity and Reproductive/Developmental Toxicity Study

Dose (mg/kg-day)	0	10	50	250
Males	n=13	n=13	n=13	n=13
Necropsy body weight (g)	501.7 ± 30.7[a]	509.6 ± 32.1	498.9 ± 28.3	496.0 ± 56.1
Liver, absolute weight (g)	14.47 ± 1.32	14.48 ± 1.57	14.67 ± 1.36	16.86 ± 2.85
Liver, relative weight (g per 100 g body weight)	2.88 ± 0.17	2.84 ± 0.18	2.94 ± 0.16	3.38 ± 0.22[b]
Kidney, absolute weight (g)	3.00 ± 0.19	2.92 ± 0.19	3.07 ± 0.24	3.28 ± 0.35[c]
Kidney, relative weight (g per 100 g body weight)	0.60 ± 0.05	0.57 ± 0.03	0.62 ± 0.04	0.67 ± 0.09
Females	n=13	n=13	n=12	n=11
Necropsy body weight (g)	313.7 ± 24.9	307.2 ± 22.2	291.0 ± 16.8[c]	296.7 ± 17.4
Liver, absolute weight (g)	13.16 ± 1.19	13.48 ± 1.23	13.50 ± 1.30	14.34 ± 1.08
Liver, relative weight (g per 100 g body weight)	4.20 ± 0.28	4.40 ± 0.37	4.63 ± 0.29[b]	4.83 ± 0.19[b]
Kidney, absolute weight (g)	1.80 ± 0.21	1.87 ± 0.20	1.75 ± 0.12	1.85 ± 0.15
Kidney, relative weight (g per 100 g body weight)	0.57 ± 0.04	0.61 ± 0.04	0.60 ± 0.05	0.62 ± 0.05

[a]Mean ± standard deviation.
[b]Significantly different from control, $p < 0.01$ (adjusted for multiple comparisons).
[c]Significantly different from control, $p < 0.05$ (adjusted for multiple comparisons).

Sources: CIPC (2010b, c).

Gross pathological evaluations of male rats showed kidney enlargement in 3/13 high-dose (250 mg/kg-day) males and 0/13 in each of the control, low-, and mid-dose groups, but no changes in the liver or other organs (Table B.4). In females, gross pathological findings were enlarged liver in 2/13 high-dose animals (versus no animals in the control or lower-dose groups) and small thymus, which was reported in 2/13, 5/13, 5/13, and 8/13 female rats of the control, low-, mid-, and high-dose groups, respectively (Table B.4).

Table B.4. Incidence Data for Selected Lesions in Sprague-Dawley Rats Administered DTDP by Gavage in the Combined Repeated-Dose Toxicity and Reproductive/Toxicity Study

Dose (mg/kg-day)	**0**						**10**						**50**						**250**					
Males																								
Gross lesions[a]	**N**				**P**		**N**				**P**		**N**				**P**		**N**				**P**	
Kidney, enlargement	13				0		13				0		13				0		10				3	
Females																								
Gross lesions[a]	**N**				**P**		**N**				**P**		**N**				**P**		**N**				**P**	
Thymus, small	11				2		8				5		8				5		5				8	
Liver, enlargement	13				0		13				0		13				0		11				2	
Males																								
Histological lesions[a]	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**
Liver, hypertrophy, hepatocyte, centrilobular	13	0	0	0	0	0	13	0	0	0	0	0	10	3	0	0	0	3	2	6	5	0	0	11[b,c]
Liver, fatty change, periportal	0	2	3	8	0	13	0	0	4	9	0	13	0	1	5	7	0	13	3	7	3	0	0	10[c]
Liver, increased/elongated catalase-positive granules[e]	2	0	0	0	0	0	2	0	0	0	0	0	2	0	0	0	0	0	0	2	0	0	0	2[c]
Kidney, eosinophilic bodies, tubular cells	10	0	1	2	0	3	11	1	1	0	0	2	8	3	1	1	0	5	4	2	4	2	1	9[d]
Females																								
Histological lesions[a]	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**	**N**	**1**	**2**	**3**	**4**	**P**
Liver, hypertrophy, hepatocyte, centrilobular	13	0	0	0	0	0	13	0	0	0	0	0	9	4	0	0	0	4[d]	0	9	4	0	0	13[b,c]
Kidney, hyperplasia, pelvic epithelium	13	0	0	0	0	0	13	0	0	0	0	0	13	0	0	0	0	0	12	0	1	0	0	1
Urinary bladder, hyperplasia, transitional cell	13	0	0	0	0	0	13	0	0	0	0	0	13	0	0	0	0	0	11	2	0	0	0	2

[a]Severity of lesion: N = no lesion, 1 = very slight, 2 = slight, 3 = moderate, 4 = severe, P = the total number of positive responders (i.e., animals exhibiting severity grade 1, 2, 3, or 4 for the selected lesion).
[b]Significantly different from control, $p < 0.01$ (Fisher's exact test).
[c]Significantly different from control, $p < 0.01$ (Mann-Whitney U test).
[d]Significantly different from control, $p < 0.05$ (Fisher's exact test).
[e]It appears from the data presented that only 2 males per group were evaluated for catalase activity.

Sources: CIPC (2010b, c).

Histological examinations revealed centrilobular hepatocellular hypertrophy in the livers of mid- and high-dose male and female rats; incidences were statistically significantly increased in males at 250 mg/kg-day and females at 50 and 250 mg/kg-day (Table B.4). Also in the liver in males, there was a significant decrease in incidence/severity of periportal fatty change. This change was not seen in females. Judging by the results reported, it appears that assays for liver catalase activity were conducted in only two males per group and no females. A "very slight" increase in incidence and size of catalase positive granules was reported in the two high-dose males tested. In the kidney, there was a significant increase in the incidence of eosinophilic bodies in renal tubular cells in 250 mg/kg-day males. The change was not seen in females. One high-dose female had "slight" hyperplasia in the epithelium of the kidney pelvis and two had "very slight" hyperplasia in the transitional epithelium of the urinary bladder.

There were no significant treatment-related effects on estrous cyclicity, mating, fertility, corpora lutea, implantation sites, or gestation in the dams, and no effects on number born, sex ratio, body weight, viability, or external/visceral malformations through post-natal day 4 in the pups. The study did not include assessment of pups for possible treatment-related effects on developmental endpoints from post-partum day 4 through adulthood.

A statistically significant decrease in live birth index ([number of live pups on day 0 / number of pups born] × 100) was reported in the 250 mg/kg-day group compared to controls (87.7 ± 28.4 versus 99.6 ± 1.6 for controls; $p < 0.05$, including adjustment for multiple comparisons according to the study authors). However, independent analysis of the live birth index results from the study report using an unpaired t-test did not confirm that the difference was statistically significant (p=0.14). This result is also of questionable toxicological significance because neither of the measured values contributing to the index (number of live pups on day 0 and number of pups born) differed from controls.

The available English summary (CIPC, 2010b) of this Japanese study report (CIPC, 2010c) stated that poor lactation was observed in the 250 mg/kg-day dose group; however, additional details and supporting data were not provided. There was no apparent effect on body weight or viability of the pups in this group.

Organ weight measurements and gross and histopathological evaluations of the testes and epididymides from the males and ovaries from the females (pathology only) revealed no evidence of DTDP treatment-related effects. Gross pathology examination revealed an epididymal nodule in 1/13 high-dose male rats, but no control or lower-dose males. Histopathological evaluation showed the nodule to be a spermatic granuloma. One other rat, a

control male, was found to have spermatic granuloma. Therefore, the incidence for this lesion was identical in control and high-dose rats, and the lesion is not considered to be related to DTDP treatment. The researchers concluded that no testicular toxicity was observed in any group in this study and did not mention the epididymal findings in the English language summary of the paper (CIPC, 2010b).