FOREWORD INTRODUCITON

Bis(2-ethylhexyl)adipate (DEHA)

CAS N°: 103-23-1

SIDS Initial Assessment Report

For

SIAM 10

Tokyo, Japan, 15-17 March 2000

1. Chemical Name: Bis(2-ethylhexyl)adipate (DEHA)

2. CAS Number: 103-23-1

3. Sponsor Country: United States

National SIDS Contact Point in Sponsor Country:
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4. Shared Partnership with:

5. Roles/Responsibilities of the Partners:

- Name of industry sponsor /consortium
- Process used

6. Sponsorship History

SIDS Dossier and Testing Plan were reviewed by the US EPA and the following SIDS Testing Plan was recommended:

no testing	(X)
testing	( )

This SIAR was reviewed at SIAM 8 (Paris, October, 1998) at which the human health assessment and conclusions were accepted and the environmental assessment was to be revised and discussed at a subsequent SIAM.

- How was the chemical or category brought into the OECD HPV Chemicals Programme ?

7. Review Process Prior to the SIAM:

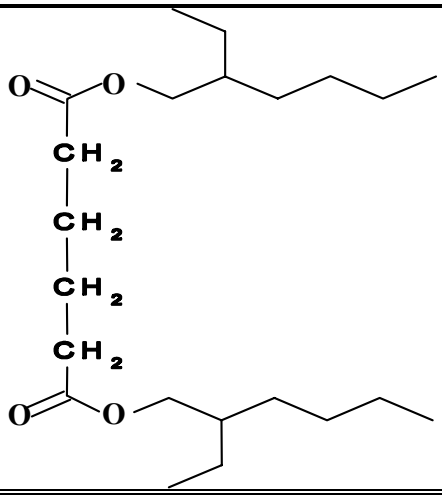
8. Quality check process:

9. Date of Submission:

10. Date of last Update:

11. Comments:

SIDS INITIAL ASSESSMENT PROFILE

CAS No.	103-23-1
Chemical Name	Bis(2-ethylhexyl)adipate
Structural Formula	

RECOMMENDATIONS

The chemical is a candidate for further work.

SUMMARY CONCLUSIONS OF THE SIAR

Human Health

DEHA exhibits low acute mammalian toxicity as seen by reported oral and dermal LD50s in rats of greater than 2 g/kg and no mortality in rodents exposed via inhalation for eight hours at levels up to saturation. Available data show that DEHA is not irritating to skin or eyes in animal studies and was not a dermal sensitizer in guinea pigs. Repeated-dose toxicity studies (up to 90-days) in rats and mice with DEHA in feed showed reduced body weight gains at levels of approximately 400 mg/kg and higher in rats and approximately 600 mg/kg and higher in mice (NOAELs of 189 mg/kg in rats and 451 mg/kg in mice). *In vitro* genotoxicity studies have been negative for mutations, unscheduled DNA synthesis and DNA interactions in bacterial and mammalian systems. *In vivo* genotoxicity studies have also been negative (two mouse micronucleus assays). DEHA has been evaluated for carcinogenicity in mice and rats, and there was no evidence of carcinogenicity in rats but there was evidence of liver cancer in female mice (significant incidence) and male mice (less significant). Tumors in mice were observed at high concentrations (3222 mg/kg for females and 2659 mg/kg in males). A one-generation reproductive toxicity test was performed in rats and there were no effects on reproduction although the body weight gains of pregnant dams and first generation pups was reduced at a dose level of approximately 3222 mg/kg. A developmental toxicity performed with DEHA in rats (animals treated orally via DEHA in feed on days 6-15 of gestation) demonstrated reduced maternal body weight gain at the highest dose (1080 mg/kg/d). There was evidence of pre-implantation fetal loss at the highest dose, but no gross, skeletal, or visceral abnormalities. A NOAEL for developmental toxicity was determined in rats at an estimated oral dose of 170 mg/kg/d, based on slight fetotoxicity from reduced ossification which was not statistically significant.

Environment

Experiments show that DEHA has no acute toxicity effects to aquatic organisms and a low bioaccumulation potential, and is readily degradable via abiotic (hydrolysis) and biotic processes. No acute aquatic toxic effects were noted at the apparent limit of DEHA solubility (0.0032 mg/L) and no effects were noted at concentrations several orders of magnitude greater than the solubility for most species. A chronic daphnid study did show effects at

concentrations slightly above the water solubility limit. There were no effects observed at the lowest concentration tested (0.014 mg/L). An *acceptable toxic concentration* of 0.035 mg/L was derived as the geometric mean of the NOEC (0.024 mg/L) and the LOEC (0.052 mg/L). A PNEC of 0.0035 mg/L has been established (0.035 divided by an assessment factor of 10).

Tests on terrestrial organisms (earthworm) have also been performed (LC50s of >1000 mg/kg and 865 mg/kg were reported after exposures of 7 and 14 days, respectively.

Exposure

DEHA is a plasticizer used primarily in food-contact wrapping. Approximately 10,000 to 50,000 tonnes are produced each year in closed systems. It is estimated that only 25-50 individuals in the US are involved in the manufacturing and handling process. Occupational exposures are low based on production in a closed system and its low vapor pressure. The estimated exposure levels to the general population via consumer products (migration of DEHA from food wraps – estimated exposures of 117 ug/kg/d) and the environment (highest measured surface water concentration was 0.001 mg/L) are considered to be quite low in several countries.

NATURE OF FURTHER WORK RECOMMENDED

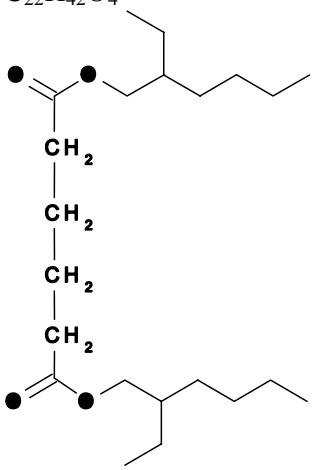
Because of the potential chronic hazard to the aquatic environment an exposure assessment is recommended with subsequent risk assessment, as appropriate.

SIDS Initial Assessment Report

1 IDENTITY

1.1 Identification of the Substance

CAS Number: 103-23-1
IUPAC Name: Bis(2-ethylhexyl)adipate
Molecular Formula: $C_{22}H_{42}O_4$
Structural Formula:



Molecular Weight: 370.64
Synonyms: DEHA

1.2 Purity/Impurities/Additives

DEHA is a clear liquid at room temperature which is manufactured as >99% pure. A minor impurity is 0.01-0.02% adipic acid.

1.3 Physico-Chemical properties

The physical properties of DEHA are shown in Table 1. DEHA is a colorless (Ashford, 1994) to light colored (Verschueren, 1983) liquid. DEHA is a liquid over a wide range of temperatures ranging from – 67.8°C (melting point) to 417 °C (boiling point). The density of DEHA is 0.922 g/mL, which is slightly less than water. This means that it will generally float on top of water, but also easily forms emulsions with water when a slight amount of energy is applied.

The aqueous solubility of DEHA has been reported in three studies, one of which yielded greatly different results from the other two. The first test utilized a separator column technique that yielded a solubility of <0.005 mg/L (Monsanto, 1982). The second test utilized a vigorous shake flask technique that stirred a mixture of water and DEHA (Monsanto, 1983) and resulted in aqueous solubilities of 0.78 mg/L and 0.23 mg/L in freshwater and saltwater, respectively. It is believed that these elevated values represent the presence of emulsions formed by the vigorous stirring. A third test has been performed that measured the aqueous solubility of DEHA using the slow stir method and reported a measured aqueous solubility of 0.0032 mg/L (Parkerton, TF et al., in review, *J. Chem. Eng. Data*).

The log of the octanol-water partition coefficient (log Kow) for DEHA was calculated from its structure. Log Kow values of >6.11 to 8.39 have been determined (Felder *et al*., 1986; US EPA, 1984; SRC, 1998). These values suggest that DEHA is a hydrophobic compound expected to have low solubility in water and a tendency to move out of water to other matrices.

The low water solubility (likely low ppb range) and high log Kow (greater than 6.0) values for DEHA can be supported by looking at analogous chemicals such as dibutyl adipate, dihexyl adipate, and diisononyl adipate. These chemicals have estimated or measured water solubilities of 16 mg/L (IUCLID, 1998a), 42 ug/L (Howard and Meylan, 1997), and "< 1 mg/L" (IUCLID, 1998b) and estimated log Kows of 3.8, 5.9, and 9.1 (USEPA,1994), respectively.

Table 1 Summary of physico-chemical properties

Property	Test Description	Results	Comments	Reference
Melting Point Boiling Point		-67.8 °C +417 °C		
Density (g/mL)		0.922		
Aqueous Solubility (µg/L)	Saturator column technique Slow stir technique	<5.0 3.2	Values of 230 to 780 µg/L were also reported using vigorous stir-flask method, which likely creates emulsions. Slow-stir techniques eliminate the formation of emulsions	Monsanto, 1982, 1983; Parkerton, 1999
Vapor Pressure (Pa)		1.1E-4 @ 20^{o} C	calculated from vapor pressure measured at 200 °C	Felder, 1986

2 GENERAL INFORMATION ON EXPOSURE

Di-(2-ethylhexyl) adipate (DEHA) is often referred to by the archaic name di-octyl adipate (DOA) in technical literature. The structure of DEHA is the result of the combination of 2-ethylhexanol with adipic acid in an esterification process. The resulting structure has a molecular weight of 370.64 g/mol and is shown below.

$$CH_3(CH_2)_3C(C_2H_5)HCH_2OC(=O)(CH_2)_4C(=O)OCH_2C(C_2H_5)H(CH_2)_3CH_3$$

DEHA is used as a plasticizer which is used primarily in food-contact wrapping. Occupational exposure to DEHA during manufacture is minimized through the use of good industrial hygiene practices which include personal protective equipment such as gloves and dust mask as appropriate. Based on a survey of manufacturers, it is estimated that no more than 25-50 individuals in the USA are involved in the manufacturing and handling process. Exposure by employees to DEHA during processing into final products is likely to be minimized through the use of good industrial hygiene practices. Occupational exposure to DEHA during formulation of end-use products cannot be estimated. Exposure by consumers has been estimated and is considered to be minimal based on the low migration of DEHA from wrapping materials into food.

2.1 Environmental Exposure and Fate

2.1.1 Environmental Fate Properties

The potential for accumulation of DEHA in aquatic organisms was examined (Felder *et al*., 1986) in a bioconcentration test using bluegill sunfish (*Lepomis macrochirus*). The test used a flow

through system that maintained the concentration of DEHA at about 250 µg/L using ^{14}C-labeled DEHA. The test was carried out for 42 days. Concentrations of DEHA in water, whole fish, viscera, and fillet were analyzed at intervals during the test. After the first 35 days of exposure, the remaining fish were exposed to clean water for an additional 14 days and concentrations of DEHA were measured in the fish at intervals.

A steady state concentration of DEHA in the whole fish was attained by day 7. A whole fish bioconcentration factor (BCF) of 27 was reported at day 35. Daily BCFs of 3.2 to 46, 1.3 to 19, and 4.1 to 66 were reported for whole fish, fillet, and viscera samples, respectively. Following exposure to clean water, a depuration rate for DEHA of 0.26/day ($t_{1/2}$ = 2.7 days) was determined. DEHA is apparently rapidly and extensively excreted from the fish, as it is for higher vertebrates such as rats. For instance, Takahashi (Takahashi *et al*., 1981) fed radio-labeled DEHA to rats and found no evidence of accumulation in any organs or tissues. After 96 hours, less than 0.14% of the administered radioactivity was found in any organ or tissue. Based on log Kow, a higher BCF in aquatic organisms may be expected for DEHA. However, the measured bioconcentration data show that DEHA likely has a low bioaccumulation potential in aquatic organisms.

A vapor pressure of 8.5 x 10^{-7} mm Hg (1.13 E-4 Pa) at 20 °C was reported for DEHA (Felder *et al*., 1986). This vapor pressure was calculated from a measured vapor pressure at 200 °C. The relative partitioning of DEHA between water and air is described by it's Henry's Law Constant (H_c). The H_c is determined by the combination of vapor pressure in units of atmospheres (P_v) and aqueous solubility in units of mol/m^3 (S_a) as follows (Eq.1).

$$H_c = P_v / S_a \qquad (1)$$

A Henry's Law Constant of 1.3 x 10^{-5} atm-m^3/mol was calculated from the vapor pressure of 1.13E-4 Pa and an aqueous solubility of 3.2 µg/L.

To further assess the relative volatilization potential of DEHA, the procedures of Lyman (Lyman *et al*., 1982) were used. The procedure assumes a shallow model river 1 m deep flowing at 1 m/sec with a wind velocity of 3 m/sec. A standard oxygen reaeration rate was assumed. If the only variable allowed to vary is the H_c, then a relative volatilization rate can be calculated. In such a model river system, a volatilization half-life of 1.1 days was calculated. This relatively rapid volatilization reflects the low aqueous solubility of DEHA. While the vapor pressure is very low, the Henry's Law Constant is relatively high due to the offsetting low aqueous solubility.

The sorption constant Koc is an estimate of a compound's capacity to sorb or bind to soil particles, sediment, or suspended solids. Measured Koc values are not available for DEHA, so Koc values were calculated in two ways using the procedures of Karickhoff (Karickhoff *et al*., 1979) as reported in Lyman (Lyman *et al*., 1982) and Mill (Mill *et al*., 1982) as reported in Felder (Felder *et al*., 1986). Karickhoff's approach used log Kow to calculate a Koc value (Eq. 2a), while Mill's approach used molar water solubility (Eq.2b).

$$\text{Log Koc} = 1.00 \log \text{Kow} - 0.21 \qquad (2a)$$

$$\text{Log Koc} = -0.792\ S_a\ (\text{molar}) - 0.27 \qquad (2b)$$

Koc values of 770,000 to 794,000 were calculated for DEHA. Using an assumed organic carbon content of 1% for either soil, sediment, or suspended solids, partition coefficients (Kd) of 7700 to 7940 were calculated. This indicates that soil, sediments, and suspended solids may be significant sinks for DEHA released to soil or surface waters.

Degradation characteristics of DEHA are presented in Table 2. Abiotic transformation processes can include photo-oxidation in the atmosphere, indirect photolysis in water, hydrolysis, and reaction with ozone or hydroxy radicals (HO-). DEHA reacts rapidly with hydroxy radicals with calculated

half-lives of 2.6 to 26 hours (Howard, 1991, calculated according to Atkinson, 1987). A hydrolysis half-life of <1 day in basic solutions was also reported (US EPA, 1984).

In studies by Saeger (Saeger *et al*., 1976), DEHA was tested for primary biodegradability using a semi-continuous activated sludge (SCAS) test. After a three week period, daily biodegradation rates of 92% loss per day and 73% loss per day using 5 mg DEHA daily addition and 20 mg DEHA daily addition, respectively. Boethling (Boethling *et al*., 1997) has shown that results from SCAS tests adequately predict treatability in real-world wastewater treatment plants when the SCAS test results in a >90% removal rate.

Saeger (Saeger *et al*., 1976) also measured the ultimate biodegradability of DEHA to carbon dioxide and water. Two test procedures were used. The first procedure was the modified Sturm test which is currently referred to as the OECD method 301B test, except that Saeger used an acclimated activated sludge microbial seed. In this test, DEHA was added to the standard BOD dilution water and microbial seed yielding a DEHA concentration of 20 mg/L and a microbial seed of 83 mL per liter total solution. The system was sealed, flasks were shaken on a rotary shaker in the dark at room temperature for 35 days. Carbon dioxide was trapped in barium hydroxide solutions, periodically collected and analyzed. By day 35, after correction with control data, 93.8% of the theoretical carbon dioxide ($ThCO_2$) was formed. The second test procedure was the Gledhill shake flask method that was set up similarly to the Sturm test but used smaller vessels and an initial DEHA concentration of 37 mg/L and 100 mL composite seed per 500 mL total solution (250 mL/L). By day 35, after correction for carbon dioxide formation in controls, 81.6% of the $ThCO_2$ was formed. Two recent tests by Huls AG (1996a,b) performed under GLP and using the modified Sturm test and ISO 10708 BOD test for insoluble substances reported that by day 28, 83% and 82% ThOD, respectively, were reported and the 10 day time windows were met, indicating that DEHA is readily biodegradable. Together, these data suggest that DEHA undergoes substantial primary and ultimate biodegradation under aerobic conditions.

Table 2 Environmental Fate Properties

Property	Test Description	Results	Comments	Reference
Partitioning Properties Henry's Law Constant ($atm\text{-}m^3/mol$)		1.3E-4	Calculated from solubility of 3.2 µg/L and vapor pressure	
Log Octanol-Water Partition Coefficient (log Kow)	Calculated from structure	>6.11 8.0 8.39		Felder *et al*., 1986; US EPA, 1984; SRC, 1998
Bioconcentration Factor (BCF)	Measured in bluegill sunfish with ^{14}C-DEHA, 28-d uptake and 14-d depuration, EPA/ASTM methods	27 (whole fish BCF at 28-d)	Depuration $t_{1/2}$ <1 day (k2=0.26 $d^{-1)}$, extensive metabolism and excretion, daily BCFs 3.2 to 46 for whole fish, 1.3 to 19 for fillet, 4.1 to 66 for viscera	Felder *et al*., 1986
Soil Partition Constant (Koc)	Calculated from log Kow Calculated from water solubility (WS) of 0.005 mg/L	794,000 770,000	log Koc=(1.00*log Kow) - 0.21 log Koc=(-0.782*molar WS) - 0.27	Karickhoff *et al*., 1979; Mill *et al*., 1982
Abiotic Transformation Photo-oxidation	Reaction with hydroxy radicals (HO-)	$t_{1/2}$ = 2.6 to 26 hours	range in polluted and unpolluted air, estimated according to Atkinson, 1987	Howard, 1991
Biotic Transformation Primary biodegradation	Semi-continuous activated sludge (SCAS)	92% loss/day 73% loss/day	Based on 5 mg/da feed rate Based on 20 mg/da feed rate	Saeger *et al*., 1976
Ultimate biodegradation	CO_2 evolution: modified Sturm test and Gledhill shake flask test	93.8% $ThCO_2$ 81.6% $ThCO_2$	35-d tests, rapid conversion of DEHA to CO_2 observed, acclimated activated sludge	Saeger *et al*., 1976
Primary biodegradation	Oxygen demand, modified Sturm test, performed using GLP	83% ThOD in 28 days	10 day time window met, readily biodegradable, 55.1% $ThCO_2$	Huls AG, 1996a
Primary biodegradation	BOD test for insoluble substances, ISO 10708	82% ThOD in 28 days	10 day time window met	Huls AG, 1996b

Potential environmental distributions of DEHA between environmental compartments were estimated using Mackay level 1 procedures (Mackay and Paterson, 1981, 1985). The Mackay Level 1 modeling approach calculates environmental distributions within a hypothetically sized "unit-world". The unit-world has compartments of an atmosphere, soil, surface water, sediment, suspended solids (any solid matter in suspension within the water column) and aquatic biota. Relative volumes of each compartment and density characteristics were designated for soil, sediments, suspended solids, and biota based on model default values. The values used for the critical physical properties of aqueous solubility, vapor pressure, soil and sediment distribution coefficient (Kd) and biota bioconcentration factor (BCF) are also shown. Any dispersive, degradation, or other loss processes were ignored for these calculations. Results are shown in Table 3.

Table 3 Mackay Level I Environmental Distribution Modeling

Compartment	Volume (m^3)	Media Density (kg/m^3)	Distribution (%)
Air	6.0×10^9	1.19	0.6
Soil	4.5×10^4	2400	51.3
Water	7.0×10^6	1000	0.2
Biota	7	1000	<0.1
Suspended Solids	35	1500	0.5
Sediments	2.1 E+4	2400	47.8
Model Parameters	Values		
Molecular Weight	370.64 g/mol		
Aqueous Solubility	0.005 mg/L		
Henry's Law Constant	1.3E-4 atm-m^3/mol		
Koc	782,000		
Kd – soil	15,640[a]		
Kd – sus. solids	31,280[b]		
Kd – sediment	31,280[b]		
BCF	27		

a Kd-soil = Koc x foc, where, foc=organic carbon content, foc=2% for soil

b Kd-soil = Koc x foc, where, foc=4% for suspended solids and sediments

The calculations suggest that only about 0.6% of the DEHA in the hypothetical system would be in the atmosphere and only 0.2% in the water column. Most of the DEHA in the hypothetical system would be associated with the soil and sediment (51.3% and 47.8%, respectively). Negligible amounts of DEHA would be associated with suspended solids or biota. These results are reflective of DEHA's low water solubility, high log Kow, and high calculated sorption constants. Actual environmental characteristics, dispersive, and degradation processes would contribute to the definition of environmental concentrations. These calculated distributions are substantially different from that reported by Felder (Felder *et al*., 1986). Felder reported that up to 65% of DEHA in a theoretical environment at equilibrium would be in the air and 34% in the water. While Felder used slightly different environmental compartment dimensions than used here, most of the difference is related to the aqueous solubility used. Felder assumed that the aqueous solubility of DEHA was 0.780 mg/L, rather than the 0.005 mg/L used here. As discussed above, the log Kow values calculated for DEHA based on structure, range from >6.11 to 8.39. This supports the lower solubility and higher soil and sediment partitioning that are used here.

2.1.2 Predicted Environmental Concentrations

Regional PECs - Environmental monitoring data were presented by Felder (Felder *et al*., 1986) and reported by Hicks (Hicks and Michael, 1983). The analyses were performed with GC/MS with selected ion monitoring (SIM). During this study, water and sediment samples were collected at 24 selected sites in the US. All samples were collected and analyzed under a rigorous Good Laboratory Practices (GLP) program. Only six of the 85 water samples exceeded the detection limit of 0.2 µg/L with a maximum reported value of 1.0 µg/L from one of four replicates at one site (the other three replicates were all ≤0.2 µg/L). The site was in the Mississippi River just south of the city of St. Louis, Missouri. Based on an average recovery of 66% for the laboratory and field spiked samples, the geometric mean concentration of DEHA in waters of the US is <0.3 µg/L. None of the 32 sediment samples exceeded the analytical detection limit of 0.10 mg/kg dry. The field and laboratory spiked sediment recoveries of DEHA was a relatively low 12.5%. This is likely attributable to the inherent difficulties in spiking sediments, mixing to achieve homogenous concentrations, preserving of the sediment, followed by extraction and analysis of the DEHA in the samples. Nevertheless, correcting upwards for the recovery, the geometric mean concentration of DEHA in US sediments was <0.8 mg/kg dry weight. Pore water concentrations in the sediments were estimated using the mean calculated Koc value of 782,000 and an organic carbon content of 1%. A Kd value of 7820 results from multiplying the Koc and the organic carbon content. Based on the geometric mean sediment concentration of <0.8 mg/kg dry weight, a pore water concentration of <0.1 µg/L is expected.

The low surface water concentrations of DEHA and the low frequency of detection (7%) within all samples and in 5 of 24 or 21% of all sites, attests to the low amounts of DEHA released to surface waters. The complete absence of detectable concentrations of DEHA in US sediments at a 0.10 mg/kg dry weight detection limit further supports the negligible releases of DEHA to surface waters. This finding is especially important as Mackay Level 1 distribution modeling suggests that substantial sorption to sediments would occur in the absence of degradation.

Local PECs - Local predicted exposure concentrations can be estimated from release data for US manufacturers and processors of DEHA. Releases to the environment of DEHA were reported under the Toxics Release Inventory (TRI) from 1987 to 1994. DEHA was removed from the TRI list based on a de-listing petition to which was granted in 1996. The basis for the delisting was the low toxicity to humans and the environment. In 1994, the last available reporting year for DEHA, 12 waste streams containing a total of 557 kg DEHA were released directly to surface waters. Another 20 waste streams totaling about 10,000 kg were sent to publicly owned treatment works (POTWs) for treatment, prior to release to surface waters. A procedure developed by the US EPA (Nabholz *et al*., 1993) assumes that the releases are mixed into the receiving stream's annualized low flow to obtain a reasonable worst case calculated in-stream concentration of DEHA. The low flows used were the lower 90^{th} percentile flow for all US receiving streams for organic manufacturers and processors (6.55×10^6 L/day) and POTWs (7.5×10^6 L/day). Direct releases were assumed to mix with the receiving stream low flow instantly with no further dilution or any other dispersion or degradation processes considered. Indirect releases were assumed to be reduced by an assumed treatment plant efficiency prior to discharge. The treatment plant efficiency assumed was 82.5%, which is the average of the SCAS test removal rates measured by Saeger (Saeger *et al*., 1976).

For direct releases to surface waters, the calculated range of DEHA concentrations was 16 to 7198 µg/L, while the geometric mean concentration based on the distribution of direct releases was 235 µg/L. For indirect releases to surface waters following treatment in the POTWs, the calculated range of DEHA concentrations was 3 to 21,692 µg/L, while the geometric mean concentration based on the distribution of indirect releases was 261 µg/L. These concentrations would only be possible during low flow conditions. They are also considerably higher than actual measured

surface water concentrations in the US, likely due to the very conservative nature of the calculations.

2.2 Human Exposure

2.2.1 Occupational Exposure

It is estimated that no more than 25-50 individuals in the USA are involved in the manufacturing and handling process. Occupational exposure during formulation of final products is likely to be minimized through good industrial hygiene practices; however, no data are available on occupational exposure levels. Exposure to vapors is unlikely because the vapor pressure for DEHA is low (1.13×10^{-4} Pa at 20°C or 2.4 mm Hg at 200°C) unless the product is heated. Based on the understanding of DEHA producers, incorporation of DEHA into products does not require heating to temperature of greater than 300°F. Exposure to an aerosol is possible during drumming and transport, although the likelihood of significant inhalation or dermal exposure is reduced through the use of good industrial hygiene practices (personal protective equipment such as gloves and dust mask if the worker deems it appropriate).

2.2.2 Consumer Exposure

Consumer exposure to DEHA via food wrap has been reported for human populations in the US (Till *et al*., 1982), Canada (Page and LaCroix, 1995), the UK (MAFF, 1987; 1990), and elsewhere (Harrison, 1988; Kozyrod and Ziaziaris, 1989; Petersen *et al*., 1995). DEHA can migrate from cling film wrappings to food. In general, higher levels of DEHA are found in food with high fat content (Harrison, 1988). Temperature also affects migration (Startin *et al*., 1987). Decreasing the storage temperature to 5°C inhibits the migration by 22%, and decreasing the temperature to -18°C inhibits the migration by up to 75%. Conversely, increasing the temperature to 50°C enhances the migration up to 400%. Microwave cooking of foods can also enhance migration. The extent to which high temperature enhances migration of DEHA into foods is dependent on the area in contact and the fat content of the food.

Estimates of total DEHA consumption are low with the MAFF reporting an estimated total consumption of 8.2 mg/person/day (117 µg/kg/day for a 70 kg person). Studies in humans indicate that this value may be an upper limit with a median exposure of 2.7 mg/person/day determined from a limited sampling (Loftus *et al*., 1994).

2.2.3 Indirect exposure via the environment

Indirect exposure that may arise from biomagnification through food-chains or from depositing from the air to plants or soil are unlikely since the measured BCF for fish was 27 after 35 days of exposure and depuration is rapid, and rapid metabolism and excretion was reported in mammals. In addition, no accumulation between trophic levels is expected so no secondary effects are expected. While DEHA is released to the atmosphere and some may reach plants or soil via rainfall or attached to particles, no terrestrial effects are expected, so no secondary effects are expected.

3 HUMAN HEALTH HAZARDS

3.1 Effects on Human Health

3.1.1 Acute Toxicity

The effects of acute exposure to DEHA are summarized in the SIDS Profile. Oral treatment of rats with DEHA has not resulted in mortality or clinical signs of toxicity except at dose levels greater than the current 2000 mg/kg limit dose level (Smyth et al., 1951; Kolmar Research Center, 1967; Mason Research Laboratories, 1976a). Data for acute inhalation exposure are limited, but available information shows no mortality to rats exposed to air saturated with DEHA for 8 hours (Smyth et al., 1951). Dermal exposure does not result in lethality or clinical signs of toxicity even at dose levels in excess of the current limit dose level of 2 000 mg/kg (Smyth et al., 1951; Kolmar Research Center, 1967). Dermal irritation following prolonged exposure (24 hours) was slight (Kolmar Research Center, 1967), but a shorter exposure period did not result in any signs of irritation (Smyth et al., 1951). DEHA was not irritating to the eye and was not a dermal sensitizer (Kolmar Research Center, 1967).

3.1.2 Repeated Dose Toxicity

DEHA has been evaluated for systemic toxicity in 14, 21, and 90 day oral feeding studies of rats and mice. Although none of the studies were conducted according to current testing guidelines, the information available suggests that repeated exposure of animals to DEHA (up to 90 days) resulted in reduced body weight gain for rats at dose levels of 6300 ppm in feed and higher, and for mice 3100 ppm in feed and higher (Mason Research Laboratories, 1976b; National Toxicology Program, 1980). Hepatic hypertrophy and increased peroxisomal enzyme activity can occur in rats and mice within 1 week of treatment with 12,000 ppm in feed, but there were no adverse effects on the liver (Chemical Manufacturers Association, 1982a; 1986; 1995). No effects were observed at 2500 ppm in feed, which is equivalent to 189 mg/kg for rats and 451 mg/kg for mice (Chemical Manufacturers Association, 1995).

3.1.3 Mutagenicity

Genotoxicity studies *in vitro* have been negative for mutations, unscheduled DNA synthesis, and DNA interaction. No mutations were found when DEHA was incubated with TA-98, -100, -1535, -1537, or -1538 strains of *Salmonella* with and without metabolic activation (CMA unpublished studies, 1982b; Zeiger *et al*., 1985). In addition, there was no evidence of mammalian cell mutation in mouse lymphoma assays conducted with and without metabolic activation (CMA unpublished studies, 1982c; MacGregor *et al*., 1988). No chromatid exchange was observed in an SCE assay using Chinese hamster ovary cells with or without activation (Galloway *et al*., 1987), and there was no induction of unscheduled DNA synthesis in primary rat hepatocytes incubated with DEHA (CMA unpublished studies, 1982d).

In vivo studies for genotoxicity have also been negative for micronucleus and dominant-lethal assays. Two independent mouse micronucleus assays were conducted without evidence of interaction with DNA even at a dose level of 5 000 mg/kg (CMA unpublished studies, 1982e; Shelby *et al*., 1993). In a dominant-lethal study using mice, DEHA did not demonstrate decreases in litter size that might suggest adverse effects on spermatogenesis (Singh *et al*., 1975).

3.1.4 Carcinogenicity

DEHA has been evaluated for its carcinogenic potential in rats and mice. No evidence of carcinogenicity was found in rats following oral treatment of 12 000 or 25 000 ppm DEHA in feed, but a significantly higher incidence of hepatocellular neoplasms was found in female mice with an insignificantly higher incidence observed in male mice at dose levels of 12 000 and 25 000 ppm in the diet (estimated dose 3222 and 8623 mg/kg for female mice, and 2659 and 6447 mg/kg for male mice; National Toxicology Program, 1980). A significantly higher incidence of hepatocellular adenomas was observed only in male mice at 25 000 ppm in the diet (6447 mg/kg). There was no carcinogenic activity when DEHA was applied to the skin of mice (Hodge *et al*., 1966). Repeated-dose studies in mice and rats suggest that species sensitivity to the carcinogenic effect of DEHA may be related to differences in cell proliferation (Lake *et al*., 1997). Based on the available data, the US EPA has classified DEHA as a Class C carcinogen (possible human carcinogen) and IARC has classified DEHA as category 3 (not classifiable).

3.1.5 Toxicity for Reproduction

Effects on Fertility

The reproductive toxicity of DEHA was evaluated in a guideline one-generation reproduction study using rats (CEFIC, 1988a). There were no effects on reproduction although the body weight gains of pregnant dams and F_1 pups was reduced at a dose level of 12 000 ppm in feed. There were no adverse effects on the reproductive organs of either generation. The NOEL was 1,800 ppm (200 mg/kg; average over a 10 week period). Singh *et al*. (1975) reported decreased fertility in male mice treated with a single dose of DEHA by ip injection during a dominant lethal study. Doses of 5 ml/kg (4610 mg/kg) and 10 ml/kg (9220 mg/kg) resulted in a decrease in fertility, fewer implants, and higher fetal mortality compared with the control group. A dose level of 922 mg/kg had no effect.

Developmental Toxicity

The developmental toxicity of DEHA has been evaluated following intraperitoneal injection and oral exposure in the diet. Singh *et al*. (1973) treated pregnant rats with DEHA by i.p. injection on gestation days 5, 10, and 15. There was a slight decrease in the mean fetal weight at the two highest dose levels, but no effect on gross, skeletal, or visceral abnormalities. The NOEL was 4.6 mg/kg/d. Another study in which pregnant rats were treated with DEHA in the diet throughout gestation also demonstrated reduced maternal body weight gain at the high dose of 1080 mg/kg/d (CEFIC, 1988b). There was evidence of pre-implantation fetal loss at 1080 mg/kg/d, but no gross, skeletal, or visceral abnormalities. A dose of 170 mg/kg/d was considered to be slightly fetotoxic resulting in reduced ossification. A dose of 28 mg/kg/d was determined to be the NOEL by the study authors. The US EPA identified a NOEL = 170 mg/kg/d for purposes of deriving an oral RfD (US EPA, 1992).

DEHA is hydrolyzed to adipic acid and 2-ethylhexanol, the latter of which is oxidized to ethylhexanoic acid (EHA). EHA should be considered when addressing the developmental toxicity of DEHA since EHA is a known potent developmental toxicant. The OECD SIDS program has reviewed both 2-ethylhexanol (SIAM 3) and ethylhexanoic acid (EHA) (SIAM 9).

3.2 Initial Assessment for Human Health

Significant human health risk from inhalation exposure is expected to be minimal based upon an inhalation toxicity study in animals (Smyth *et al*., 1951). Significant human health risk from dermal exposure is minimal based upon dermal toxicity studies (Smyth *et al*., 1951; Kolmar Research

Center, 1967) and dermal irritation studies (Smyth *et al*., 1951; Kolmar Research Center, 1967) in animals which indicate that DEHA is virtually non-toxic and non-irritating to the skin.

Review of the data for DEHA indicates that this substance has a low potential for toxicity from acute exposure, and that repeated exposure of high dose levels may result in adaptive responses in the liver, but no irreversible changes.

Animal data indicate that the liver is a target for DEHA toxicity, and that repeated exposure of rodents to DEHA results in the induction of enzymes associated with the organelles called peroxisomes (CMA, 1982a, 1986, 1989; Lake *et al*., 1997). The increased size and number of these organelles (peroxisome proliferation) has been associated with carcinogenesis in rodents (Hsia, 1990). This mechanism may be linked to induction of enzymes that produce hydrogen peroxide (oxidative stress), or it may be linked to mitogenic responses (cell proliferation) or interference with programmed cell death (apoptosis). In any case, the applicability of rodent liver cancer at high dose levels to humans via this mechanism has been questioned (Cattley *et al*., 1998) because primates (Rhodes *et al*., 1986; Short *et al*., 1987; Kurata *et al*., 1998) and human cells (Elcombe and Mitchell, 1986) are refractory to the effects of other peroxisome proliferators. Recent studies have shown that a nuclear receptor (PPARα) plays a central role in the carcinogenic process (Peters *et al*., 1997) and the expression of this receptor in human liver is lower than in rodent liver suggesting that receptor density is a key element for species specificity (Tugwood *et al*., 1996, Palmer *et al*., 1998). DEHA is a weaker peroxisome proliferator compared with di(2-ethylhexyl)phthalate and other peroxisome proliferators (Barber *et al*., 1987).

Singh *et al*. (1975) suggested a potential effect of DEHA on the testes using a dominant lethal study design. Since only male mice were treated in this study, decreases in fertility, and pre- and post-implantation losses, can only be ascribed to effects on the testes. However, the effect levels were high (≥ 4610 mg/kg), the route of administration was intraperitoneal, and the testes of male mice were not examined histologically. In addition, treatment of rats with 25 000 ppm (2604 mg/kg/d) for 3 weeks did not result in histopathologic lesions in the testes. Therefore, it appears that a NOEL level of 2604 mg/kg/d can be established for effects on reproduction. The ability of DEHA to bind and activate the estrogen receptor has been tested (Jobling *et al*., 1996). DEHA bound to the receptor at a concentration of 1 mM, but did not activate the receptor. The ability of DEHA to elicit a uterotrophic response *in vivo* was also negative (JPIA, 1998).

Effects by DEHA on development have been identified using i.p. injection at dose levels that exceed present regulatory guidelines for limit doses (Singh *et al*., 1975). Reduced fetal weight occurred at high dose levels (≥ 4610 mg/kg). A subsequent developmental screening study conducted at a high dose of 1080 mg/kg/d found no teratogenic effect, but reduced litter size was observed at 1080 mg/kg/d. In that study, animals were treated via the diet. A dose of 1800 ppm (170 mg/kg/d) was considered to be fetotoxic resulting in reduced ossification which was not statistically significant. A dose of 28 mg/kg/d was determined to be the NOEL by the authors, but since the changes at 170 mg/kg were not statistically significant this dose level may be more appropriate for a NOAEL.

Although the effect on the liver and the potential for hepatocellular neoplasms might appear to be the critical effect for risk assessment, an analysis of the possible non-genotoxic mechanisms of carcinogenicity is outside of the scope of the OECD SIDS Program. Developmental toxicity may be more appropriate as a critical effect for assessment based on dose levels at which effects are observed. The U.S. EPA has also identified developmental toxicity as the critical endpoint in its Integrated Risk Information System (IRIS) (US EPA, 1992). Using the NOAEL of 170 mg/kg/d for developmental toxicity, an Estimated Level of Low Concern (EDLC) can be calculated using uncertainty factors of 10 for interspecies extrapolation and 10 for intraspecies extrapolation.

$$EDLC = \frac{170}{100} = 1.7 \text{ mg/kg/d}$$

The greatest potential for human exposure is via food in contact with cling-film wrap. Migration rates of DEHA from cling-film wrap are variable depending on the fat content of the food. However, based on the consumption of certain food groups by the general population in Europe (MAFF, 1990), the total dose received by humans has been calculated to be 117 µg/kg/d. This value is lower than the EDLC for developmental toxicity.

The EC Scientific Committee for Food has established a tolerable daily intake (TDI) of 0.3 mg/kg/day for DEHA based on a NOEL of 30 mg/kg/d from the developmental toxicity study. Comparing this value to the amount of DEHA ingested from food, the exposure does not exceed the TDI. However, Petersen *et al*., (1995) suggested that a small portion of the Scandanavian population might be exposed to higher levels of DEHA based on the assumption of excessive consumption of cheese (greater than that consumed by 80% of the population). These consumption values have not been verified by any survey of food consumption. Therefore, the assertion that small portions of the population are exposed to levels of DEHA that exceed the TDI cannot be confirmed. In addition, the basis for the TDI is overly conservative based on the lack of statistically significant changes at the dose level of 170 mg/kg/d.

4 HAZARDS TO THE ENVIRONMENT

4.1 Aquatic Effects

Acute Toxicity Test Results

The acute toxicity of DEHA was measured with several species of aquatic organisms, including fish, invertebrates, and algae. Both benthic and pelagic organisms were tested as were both freshwater and marine species. The available data are shown in Table 4.

Toxicity test data on four freshwater fish species are available, including bluegill sunfish (*Lepomis macrochirus*), fathead minnow (*Pimephales promelas*), rainbow trout (*Oncorhynchus mykiss*), and carp (*Cyprinus carpio*). For invertebrates, the pelagic daphnid *Daphnia magna* and the benthic midge larvae *Chironomus tentans* and *C. riparius*, respectively, were tested. Also tested were the amphipod *Gammarus fasciatus* and the isopod *Asellus* species. Marine invertebrates tested were mysid shrimp (*Mysidopsis bahia*), the grass shrimp (*Paleomonetes pugio*), and *Ampelisca abdita*. Green algae toxicity tests using *Selenastrum capricornutum* and *Scenedesmus subspicatus* were also tested (Table 4).

No acute effects at or above its water solubility limit were reported for any species tested with DEHA except for a study with rainbow trout (Hrudey *et al,*1976) exposed to emulsions of DEHA reported 30 to 55% mortality across the range of concentrations tested 54 to 420 mg/L. Such observations have been noted to be a methodological difficulty with floating oily liquids that have a low aqueous solubility and easily form emulsions with water (Adams *et al*., 1995; Rhodes *et al*., 1995; Staples *et al*., 1997).

Chronic Toxicity Test Results

The chronic toxicity of DEHA has been studied with two species, the invertebrate daphnid *Daphnia magna* and the green algae *Selenastrum capricornutum*. No effects were reported for the green algae. A 96-h NOEC of =780 µg/L was reported, which is well in excess of the aqueous solubility

of DEHA. Felder et al. (1986) reports of a study in which daphnids were exposed to DEHA under flow-through conditions for 21 days at measured concentrations of 0.014, 0.024, 0.052, 0.087, and 0.180 mg/L.

Chronic effects to daphnids were reported including immobilization, reduced lengths, and reduced young per adult per day at measured test concentrations above 0.052 mg/L. Thus, the authors concluded that the geometric mean of the NOEC (0.024) and LOEC (0.052) was 0.035 mg/L.

Structure activity relationship (SAR) analyses supports the findings reported in Felder et al. ECOSAR analyses with the analogous chemicals dibutyl adipate, dihexyl adipate, and diisononyl adipate suggests that chronic effects to aquatic organisms would be expected for dibutyl adipate (log Kow of 3.9) at relatively high concentrations and for dihexyl adipate (log Kow of 5.9) at relatively low concentrations; but not for diisononyl adipate (log Kow of 9.1) for which no chronic aquatic toxicity is expected. The log Kow for DEHA is estimated to be 7.8; in between that of dihexyl adipate and diisononyl adipate. Thus, a certain level of chronic toxicity might be expected, as is shown by the Felder et al. study.

Taken together, the Felder et al. and QSAR analysis suggest that further chronic aquatic toxicity studies are not necessary to meet the SIDS-level requirements.

Based on the available data, a concern concentration (U.S. EPA term) or predicted no effect concentration (PNEC) for aquatic organisms for DEHA is one-tenth of the chronic value in the Felder et al. study (0.0035 mg/L, or 3.5 ug/L).

Table 4 Acute and Chronic Toxicity to Aquatic Organisms.

Organism	Test Type	Results	Comments	Reference
Fish				
Bluegill sunfish *Lepomis macrochirus*	96-h static, freshwater	No effects at 780 µg/L	greatly exceeds aqueous solubility[1] based on EPA (1977) methods	Felder *et al*., 1986
Fathead minnow *Pimephales promelas*	96-h static, freshwater	No effects at 780 µg/L	greatly exceeds aqueous solubility based on EPA (1977) methods	Felder *et al*., 1986
Rainbow trout *Oncorhynchus mykiss*	96-h static, freshwater	No effects at 780 µg/L	greatly exceeds aqueous solubility based on EPA (1977) methods	Felder *et al*., 1986
Rainbow trout *Oncorhynchus mykiss*	96-h static, freshwater	30 to 55% mortality at 54 to 110 mg/L	authors note physical effects due to coating by test material	Hrudey *et al*., 1976
Carp Cyprinus carpio	96-h semi-static, freshwater	No effects at any concentration, 96-h LC50>1.6 mg/L (only concentration tested)	GLP	Huls AG (1996c)
Invertebrates				
Water flea *Daphnia magna*	48-h, static, freshwater	No effects, 48-h EC50>1.6 mg/L (only concentration tested)	greatly exceeds aqueous solubility	Huls AG, 1996d
Water flea *Daphnia magna*	21-d chronic, reproduction	Five concentrations tested (from 0.014 mg/L to 0.180 mg/L). Maximum acceptable toxicant concentration (MATC) reported to be 0.024 to 0.052 mg/L	Lowest measured value (0.014 mg/L) is within a factor of 5 of the two measured water solubility values (0.005 and 0.0032)	Felder et al., 1986
Midge larvae *Chironomus tentans*	96-h static, freshwater	No effects at 780 µg/L	greatly exceeds aqueous solubility, based on EPA (1977) methods	Felder *et al*., 1986
Midge larvae Chironomus riparius	96-h flow-through, freshwater	No effects at 730 µg/L	greatly exceeds aqueous solubility	Springborn Life Sciences, 1989a
Amphipod *Gammarus fasciatus*	96-h flow-through, freshwater	No effects at 730 µg/L	greatly exceeds aqueous solubility	Springborn Life Sciences, 1989a

[1] Aqueous solubility measured to be 0.0032 mg/L.

Isopod *Assellus sp.*	96-h flow-through, freshwater	No effects at 730 µg/L	greatly exceeds aqueous solubility	Springborn Life Sciences, 1989a
Mysid shrimp *Mysidopsis bahia*	96-h flow-through, marine	No effects at 230 µg/L	greatly exceeds aqueous solubility	Springborn Life Sciences, 1989b
Grass shrimp *Paleomonetes pugio*	96-h flow-through, marine	No effects at 230 µg/L	greatly exceeds aqueous solubility	Springborn Life Sciences, 1989b
Algae				
Green algae *Selenastrum capricornutum*	96-h static, growth inhibition, freshwater	No effects at 780 ug/L	greatly exceeds aqueous solubility	Felder *et al*., 1986
Green algae *Scenedesmus subspicatus*	72-h, static, growth inhibition	No effects at only concentration tested, 72-h EC50>1.4 mg/L	greatly exceeds aqueous solubility	Huls AG, 1996e
Microorganisms				
Activated sludge	effects on ability of activated sludge organisms to biodegrade DEHA	No effects at 352 mg/L, highest concentration tested	greatly exceeds aqueous solubility	Huls AG, 1996f

4.2 Terrestrial Effects

Tests using the earthworm (*Eisenia foetida foetida*). Earthworms were exposed to DEHA amended quartz sand and soil (method 88 / 302 EWG). LC_{50}s of >1000 mg/kg and 865 mg/kg were reported after 7 and 14 day exposures, respectively (Huls, 1996g).

4.3 Other Environmental Effects

Secondary effects include those that may arise from biomagnification through food-chains or from depositing from the air to plants or soil. Since the measured BCF for fish was reported to be 27 after 35 days of exposure, although it may have been higher, and depuration is rapid, and rapid metabolism and excretion was reported in mammals, so accumulation in aquatic organisms is not expected. Therefore, secondary effects in fish consumers is not expected. While DEHA is released to the atmosphere and some may reach plants or soil via rainfall or attached to particles, based on the earthworm tests no terrestrial effects are expected, so no secondary effects are expected.

No effects on benthic organisms are expected from DEHA. The toxicity of DEHA to benthic organisms was tested with three species of invertebrates, including the midges *Chironomus tentans* and *C. riparius*, and the amphipod *Gammarus fasciatus*. No acute effects were reported for any sediment-dwelling species when tested at concentrations of DEHA up to several orders of magnitude greater than its aqueous solubility. DEHA is readily biodegradable and has rapid photolysis and hydrolysis rates, indicating that this chemical will not accumulate in sediment.

Concentrations of DEHA that were used in SCAS tests and ultimate biodegradation tests ranged from 3300 to 37,400 µg/L. No inhibition of either primary or ultimate biodegradation of DEHA was reported. Concentrations of DEHA up to 352 mg/L had no inhibitory effect on activated sludge organisms (Huls, 1996f). Therefore, no inhibition of the treatability of DEHA in wastewater treatment plants is expected.

4.4 Initial Assessment for the Environment

DEHA poses negligible hazard to the aquatic environment based on the absence of acute effects to aquatic organisms, low bioaccumulation potential, very low environmental measured concentrations, and is readily degradable via abiotic (hydrolysis) and biotic processes. No acute aquatic toxic effects were noted at the apparent limit of DEHA solubility (0.0032 mg/L) and no effects were noted at concentrations several orders of magnitude greater than the solubility for most species. A chronic daphnid study did show effects at concentrations slightly above the water solubility limit. There were no effects observed at the lowest concentration tested (0.014 mg/L).

The ratio of predicted exposure concentrations (PECs) to predicted no effect concentrations (PNECs) is a simple and straight forward characterization of potential adverse risks to exposed populations posed by a particular chemical. The PEC / PNEC ratio requires that both PECs and PNECs be determined. Due to the absence of acute toxicity to aquatic organisms at levels of saturation and the absence of expected toxicity to terrestrial organisms, no PEC / PNEC ratios can be calculated for DEHA under these conditions. However, for chronic toxicity to aquatic organisms, a PNEC of 3.5 ug/L is compared with the highest measured DEHA concentration in water (1.0 ug/L) for a PEC/PNEC ratio of 0.29. PEC/PNEC ratios of less than 1 suggest there is no need for further work in the OECD HPV Chemicals Programme. Nevertheless, because of the potential chronic hazard to the aquatic environment member countries are invited to perform a more in-depth exposure assessment with subsequent risk assessment, as appropriate.

5 RECOMMENDATIONS

The chemical is a candidate for further work.

Because of the potential chronic hazard to the aquatic environment, member countries are invited to perform a more in-depth exposure assessment with subsequent risk assessment, as appropriate.

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SIDS DOSSIER

CAS-No.: 103-23-1
EINECS-No.: 203-090-1
IUPAC-Name: Bis(2-ethylhexyl)adipate

Producer related part
Company: Chemical Manufacturers Association
Creation date: 03/02/94

Substance related part
Company: Chemical Manufacturers Association
Creation date: 03/02/94

Printing date: 31/05/94
Revision date: 03/02/94
Date of last Update: 31/05/98

1.04 OECD and Company Information

Type lead organisation
Company Chemical Manufacturers Association
Street 2501 M St., NW
Town 20037 Washington, DC
Country United States
Phone 202-887-1207 Telex
Telefax 202-887-1237 Cedex

1. General Information

1.1 General Substance Information

Molecular Formula: c22-h42-o4
Molecular Weight: 370.64
Smiles Code: CCCCC(CC)COC(=O)CCCCC(=O)OCC(CC)CCCC

Physical Status	Purity organic			
liquid	>	99	..	% w/w

1.2 Synonyms

Bis(2-ethylhexyl) adipate
BEHA
Di(2-ethylhexyl) adipate
DEHA
Dioctyl adipate
DOA

1.3 Impurities

IUPAC-Name	Value
adipic acid	0.01 ..0.02 % w/w

1.5 Quantity

Quantity produced or imported

produced 10 000 - 50 000 tonnes in year 1993

==========

Indicate if the substance has been produced during the 12 months after adoption of the regulation: yes

Indicate if the substance has been imported during the 12 months after adoption of the regulation: no data

1.6.1 Labelling

Labelling no labelling required (no data available)
Specific Limits no data
Symbols
Nota

1.7 Use Pattern

Type of Use	Category
type	Non dispersive use
industrial	Polymers industry
use	other
type	Use resulting in inclusion into or onto matrix

Remark Plasticizer.

1.9 Source of Exposure

Remark Di-(2-ethylhexyl) adipate is manufactured by esterification of adipic acid with 2-ethylhexanol. The process occurs in a closed system at elevated temperature. Unreacted acid is neutralized, while unreacted alcohol is steam-stripped. The ester (DEHA) is transferred to closed containers for storage and transport.

Remark Because DEHA is manufactured in a closed system, the exposure potential of workers is low. Exposure during storage and transport is also low due to the low volatility of the test substance and use of protective equipment. It is estimated that only 25-50 individuals are involved in the manufacture, storage, and transport of the test substance.

2.1 Melting Point

Value =	-67.8	..	degree C
Decomposition	no		
Sublimation	no		
Method	other		
Year			
GLP	no data		

2.2 Boiling Point

Value =	417	..	degree C
Pressure			
Decomposition	no		
Method			
Year	1966		
GLP	no		

2.3 Density

Type	relative density	
Value =	0.922	..
Temperature	25	degree C
Method		
Year	1976	
GLP	no	

2.4 Vapour Pressure

Value =	1.13E-6	..	hPa
Temperature	20	degree C	
Method			
Year			
GLP	no data		
Remark	Reported in Felder, et al. 1986. Env. Tox. & Chem. 5:777-784. Calculated from vapor pressure measured at 200 C.		
Remark	Value reported in Felder (1986) was 8.5E-7 mm Hg.		

Value =		..	hPa
Temperature	200	degree C	
Method			
Year			
GLP	no data		
Remark	Reported in Felder, et al. 1986. Env. Sci. & Chem. 5:777-784. Original source unknown. Methods unknown.		
Remark	Value reported in Felder (1986) was 2.3 mm Hg.		

2.5 Partition Coefficient

log Pow >	6.11	..
Temperature		degree C
Method	other (calculated)	
Year	1986	
GLP	no data	
Reference	Felder et al. 1986. Env.Tox. & Chem. 5:777-784.	

2.6 Water Solubility

Value =	0.0032	mg/l	at 22	degree C	
pH	Concentration	at	degree C		

pKa at 25 degree C
Descr. insoluble
Method
Year 1999
Remark Aqueous solubility measured using slow-stir method for low soluble compounds
GLP yes
Reference Parkerton, TF. 1999. Submitted to J. Chemical Engineering Data.

Value = <0.0005 mg/l at 22 degree C
pH Concentration at degree C
pKa at 25 degree C
Descr. insoluble
Method
Year 1986
GLP no data
Reference Felder, J.D., Adams, W.J., and Saeger, V.W. (1986). Assessment of the Safety of Dioctyl Adipate in Freshwater Environments, Environ. Toxicol. Chem. 5:777-784

Value = 0.78 mg/l at 22 degree C
pH Concentration at degree C
pKa at 25 degree C
Descr. slightly soluble
Method
Year 1986
Remark Measured using rapid stir, shake flask method that results in emulsions of low soluble liquid compounds
GLP no data
Reference Felder, J.D., Adams, W.J., and Saeger, V.W. (1986). Assessment of the Safety of Dioctyl Adipate in Freshwater Environments, Environ. Toxicol. Chem. 5:777-784

2.11 Oxidizing Properties

Result other
Method
Year
GLP
Remark DEHA is unreactive to radicals RO2.
DEHA is unreactive to ozone O3
DEHA reacts rapidly with OH, half-lives = 2.6 to 26 hrs (estimated using procedures of Atkinson, 1987)
Reference Howard, PH. (1991) Handbook of Environmental Degradation Rates, Lewis Publishers, Inc. Chelsea, MI

3.1.2 Stability in Water

A hydrolysis half-life of <1 day in basic solutions was also reported (US Environmental Protection Agency (1984). Chemical Hazard Information Profile for Diethylhexyl Adipate. Draft Report, September 28, 1984, Washington, DC).

3.2 Monitoring Data (Environment)

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Delaware River, Wilmington, DE, USA
Remark
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Delaware River (mouth), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Delaware River(mouth), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Chesapeake Bay (estuary), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Baltimore Harbor (Sparrow's Point), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Ohio River (Gallipolis Ferry), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark 0.00025 mg/L (1 sample), <0.0002 mg/L (3 samples), Ohio River (Pittsburgh, PA, USA)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water

Remark <0.0002 mg/L, 4 samples, Kanawha River, Nitro, WV, USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Kanawha River (Winfield Dam), WV, USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Southern Lake Michigan, Chicago, IL, USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Northern Lake Michigan, Grand Traverse Bay, USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Lake Huron (Saginaw Bay), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Saginaw River at Lake Huron, USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Eastern Lake Superior (Sault Ste. Marie)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark 0.00030 mg/L (1 sample), <0.0002 mg/L (2 samples), Lake Ontario (Four Mile Creek)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Niagara River, Sandy Beach, NY, USA
GLP
Reference Felder, et al. 1986. Env. Sci. & Chem. 5:777-784.

Type of Measurement background concentration

Media surface water
Remark <0.0002 mg/L, 3 samples, Detroit River (mouth), USA
GLP
Reference Felder et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 3 samples, Mississippi River (above St. Louis, MO), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark 0.0010 mg/L (1 sample), 0.00020 mg/L (1 sample), <0.0002 mg/L (2 samples), Mississippi River (below St. Louis, MO), USA
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark 0.00030 mg/L (1 sample), <0.0002 mg/L (2 samples), Mississippi River (Memphis, TN, USA)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Missouri River (St. Louis, MO, USA)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark <0.0002 mg/L, 4 samples, Alabama River (Mobile, AL, USA)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media surface water
Remark 0.0007 mg/L (1 sample), <0.0002 mg/L (3 samples), San Francisco Bay (Brooks Island, CA, USA)
GLP
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP) DELAWARE RIVER (MOUTH), USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
DELAWARE RIVER (ESTUARY), USA

<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
CHESAPEAKE BAY (ESTUARY), USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
OHIO RIVER (GALLIPOLIS FERRY)
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
CECOS/CER LANDFILL, WILLIAMSBURGH, OH, USA
<0.10 MG/KG, 3 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO ES-83-SS-24
FALL 1982 SAMPLING (GLP)
KANAWHA RIVER, WV, USA (WINFIELD DAM)
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
SOUTHERN LAKE MICHIGAN, CHICAGO, IL, USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
NORTHERN LAKE MICHIGAN, GRAND TRAVERSE BAY, MI,
USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
LAKE HURON, SAGINAW BAY, MI, USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
EASTERN LAKE SUPERIOR, SAULT STE MARIE, MI, USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24

FALL 1982 SAMPLING (GLP)
NIAGARA RIVER, SANDY BEACH, NY, USA
<0.10 MG/KG

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
MISSISSIPPI RIVER, MEMPHIS, TN, USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
MISSOURI RIVER, ST. LOUIS, MO, USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONSANTO CO. REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
ALABAMA RIVER, MOBILE, AL, USA
<0.10 MG/KG, 2 samples

Type of Measurement background concentration
Media sediment
Remark MONASANTO CO, REPORT NO. ES-83-SS-24
FALL 1982 SAMPLING (GLP)
SAN FRANCISCO BAY, (BROOKS ISLAND), CA, USA
<0.10 MG/KG, 4 samples

3.3.1 Transport between Environ. Compart.

Type adsorption
Media water - soil
Method other
Year
Remark Soil Partition Constant (Koc) = 770,000 L/kg, calculated from aqueous solubility: log Koc=-0.27-0.782(log molar solubility).
Reference Mill, et al. 1982. USEPA, Athens, GA, USA. EPA-600/3-82-022.

3.3.2 Distribution

Media air - biota - sediment(s) - soil - water
Method Calculation according Mackay, Level I
Year
Remark Mackay Model Level I - Percent Distribution
Air: 0.6%
Water: 0.2%
Soil: 51.3%
Sediment: 47.8%
Biota: 0.5%
Reference Trent University, Environmental Modeling Center, EQC Level I Model

3.5 Biodegradation

Type	aerobic
Inoculum	activated sludge
Concentration	21.0 mg/l related to test substance
Degradation	= 83 .. % thOD after 28 day
Results	met 10 day window (reached >60% within 10 days of achieving the initial 10%), readily biodegradable
Method	Modified Sturm test
Year	1996
GLP	yes
Test substance	Vestinol OA, di (2-ethylhexyl) adipate
Remark	"readily biodegradable"
Remark	equaled 15.0 mg C/L
Remark	55.1% carbon dioxide formation
Remark	Sodium benzoate control reached 89% in 28 day
Reference	Huls AG. 1996. Bestimmung der biologischen Abbaubarkeit von Vestinol OA in Modifizierten Sturm-Test. EG-Richtlinie 92/69/EWG C.4-C. Abschlussbericht ST-113/96.

Type	aerobic
Inoculum	activated sludge
Concentration	= 52 mg/l related to test substance
Degradation	= 82 % thOD after 28 day
Results	met 10 day window (reached >60% within 10 days of achieving the initial 10%), readily biodegradable
Method	ISO 10708 BOD Test for Insoluble Substances
Year	1996
GLP	yes
Test substance	Vestinol OA, di (2-ethylhexyl) adipate
Remark	"readily biodegradable"
Reference	Huls AG. 1996. Bestimmung der biologischen Abbaubarkeit von Vestinol OA im Blok Test (BOD-Test for Insoluble Substances). Abschlussbericht BO - 89 / 42.

Type	aerobic
Inoculum	activated sludge
Concentration	3.3 mg/l related to test substance
Degradation	= 88 ..96 % after 1 day
Results	other
Kinetic	1 day = 88 .. 96 % .. .. ..
Method	
Year	
GLP	
Testsubstance	
Remark	"substantial primary biodegradation"
Remark	Addition rate of 5 mg/24 hr
Reference	Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.
Reference	Saeger, et al. 1976. Appl. Environ. Microbiol. 31:29-34. (method)

Type	aerobic
Inoculum	activated sludge
Concentration	13.3 mg/l related to Test substance
Degradation	= 65 ..81 % after 1 day
Results	other

```
Kinetic         1   day   =     65     .. 81     %
                          ..
                          ..
                          ..
                          ..
Method
Year
GLP
Testsubstance
Remark          "undergoing substantial primary biodegradation"
Remark          addition rate of 20 mg/24 hr
Reference       Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.
Reference       Saeger, et al. 1976. Appl. Environ. Microbiol.
                31:29-34.  (method)

Type            aerobic
Inoculum        aerobic microorganisms
Concentration                          related to
Degradation     =     94      ..         % after 35     day
Results             other
Kinetic                               ..
                          ..
                          ..
                          ..
                          ..
Method
Year
GLP
Testsubstance
Remark      CO2 evolution test (ultimate biodegradation) based on
            measured concentrations
Remark      "undergoing substantial ultimate biodegradation   (ie.
            conversion to CO2)"
Reference   Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.
Reference   Thompson & Duthie. 1968. J. Water Pollut. Control. Fed.
            40:306-319. (method)
Reference   Sturm, R.M. 1973. J. Am. Oil Chem. Soc. 50:159-167.
            (method)

Type
Inoculum        Aspergillus niger
Concentration                           related to Test substance
Degradation                  ..          % after
Results         other
Kinetic                               ..
                          ..
                          ..
                          ..
                          ..
Method              other
Year
GLP                 no
Testsubstance   as prescribed by 1.1 - 1.4
Remark      Mycelium homogenates of Aspergillun niger   hydrolyze
            DEHA.
Reference   Anisimov, et al. 1981. reported in Chem Abstracts
            95:182143r

Type
Inoculum        Penicillium sp.
Concentration                           related to
Degradation                  ..          % after
```

Results other
Kinetic ..
..
..
..
..
Method other
Year
GLP no
Testsubstance as prescribed by 1.1 - 1.4
Remark Penicillium funiculosum will degrade the plasticizer in the presence of other carbon sources.
Reference Bochkareva, et al. 1976, reported in Chem Abstracts 85:109318r.

3.7 Bioaccumulation

Species Lepomis macrochirus
Exposure Period 28 day
Temperature degree C Concentration 0.25 mg/l
BCF = 27 ..
Elimination yes
Method other
Year 1979
GLP yes
Testsubstance as prescribed by 1.1 - 1.4
Reference Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

4.1 Acute/Prolonged Toxicity to Fish

Type	semi-static
Species	Cyprinus carpio
Unit	mg/l
Exposure Period	96 hour
NOEC	..
LC0	>1.60 ..
LC50	> 1.60 ..
LC100	..
Analyt. Monitoring	yes
Method	Semistatischer Test entsprechend der EG-Vorschrift 92/69 EWG Teil C 1
Year	1996
GLP	yes
Testsubstance	Vestinol OA, di-(2-ethylhexyl) adipate
Reference	Huls AG, 1996, Bestimmung der akuten Wirkungen von Vestinol OA gegenuber Fischen (nach EG 92/69 C 1), Abschlussbericht FK 1353.

Type	static
Species	Lepomis macrochirus
Unit	mg/l
Exposure Period	96 hour
NOEC	> 0.78 ..
LC0	..
LC50	> 0.78 ..
LC100	..
Analyt. Monitoring	yes
Method	other
Year	1977
GLP	no data
Testsubstance	as prescribed by 1.1 - 1.4
Reference	Felder, J.D., Adams, W.J., and Saeger, V.W. (1986). Assessment of the safety of dioctyl adipate in freshwater environments. *Environ Toxicol Chem* **5**: 777-784

Type	static
Species	Pimephales promelas
Unit	mg/l
Exposure Period	96 hour
NOEC	> 0.78 ..
LC0	..
LC50	> 0.78 ..
LC100	..
Analyt. Monitoring	yes
Method	other
Year	1977
GLP	no data
Testsubstance	as prescribed by 1.1 - 1.4
Reference	Felder, J.D., Adams, W.J., and Saeger, V.W. (1986). Assessment of the safety of dioctyl adipate in freshwater environments. *Environ Toxicol Chem* **5**: 777-784

Type	static
Species	*Oncorhynchus mykiss*
Unit	mg/l
Exposure Period	96 hour
NOEC	> 0.78 ..
LC0	..

LC50	> 0.78	..
LC100	..	
Analyt. Monitoring	yes	
Method	other	
Year	1977	
GLP	no data	
Testsubstance	as prescribed by 1.1 - 1.4	
Reference	Felder, J.D., Adams, W.J., and Saeger, V.W. (1986). Assessment of the safety of dioctyl adipate in freshwater environments. *Environ Toxicol Chem* **5**: 777-784	

Type	static	
Species	Salmo gairdneri	
Unit	mg/L	
Exposure Period	96 hour	
NOEC	..	
LC0	..	
LC50	ca. 54	..110
LC100	..	
Analyt. Monitoring	no data	
Method		
Year		
GLP	no	
Testsubstance		
Reference	LC50 estimated, Hrudey, et al. 1976. Proc. 11th Canadian Symp., Water Pollut. Res. Canada:34-45.	

4.2 Acute Tox. to Aquatic Invertebrates

Species	Daphnia magna	
Unit	mg/L	
Exposure Period	48 hour	
NOEC	..	
EC0	..	
EC50	= >1.6	..
EC100	..	
Analyt. Monitoring	yes	
Method	Statischer Test entsprechend der EG-Vorschrift EG/92/69/EWG	
Year	1996	
GLP	yes	
Testsubstance	Vestinol OA, di-(2-ethylhexyl) adipate	
Reference	Huls AG, 1996, Bestimmung der Auswirkungen von Vestinol OA auf das Schwimmverhalten von Daphnia magna (nach EG-Richtlinie 92/69/EWG) Abschlussbericht DK-677.	

Species	Daphnia magna	
Unit	mg/L	
Exposure Period	48 hour	
NOEC	< 0.32	..
EC0	..	
EC50	= 0.66	..
EC100	..	
95% CI	=0.48	..0.85
Analyt. Monitoring	yes	
Method	other	
Year	1977	
GLP	no data	
Testsubstance	as prescribed by 1.1 - 1.4	
Reference	Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.	

Species	other		
Unit	mg/l		
Exposure Period	96	hour	
NOEC		..	
EC0		..	
EC50	> 0.78	..	
EC100		..	
Analyt. Monitoring	yes		
Method	other		
Year	1977		
GLP	no data		
Testsubstance	as prescribed by 1.1 - 1.4		
Remark	Chironomus tentans		
	GLP		
Reference	Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.		

Species	other		
Unit	mg/l		
Exposure Period	96	hour	
NOEC	= 0.73	..	
EC0		..	
EC50	> 0.73	..	
EC100		..	
Analyt. Monitoring			
Method			
Year			
GLP	yes		
Testsubstance			
Remark	Chironomus riparius		
Remark	USEPA/OTS guidelines, ASTM (1980)		
Reference	Monsanto Co. Springborn Life Sciences. Feb 23, 1989. Report #88-12-2997.		

Species	Gammarus fasciatus		
Unit	mg/l		
Exposure Period	96	hour	
NOEC	= 0.73	..	
EC0		..	
EC50	> 0.73	..	
EC100		..	
Analyt. Monitoring	yes		
Method	other		
Year	1980		
GLP	yes		
Testsubstance	as prescribed by 1.1 - 1.4		
Remark	USEPA/OTS guidelines, ASTM (1980)		
Reference	Monsanto Co. , Springborn Life Sciences, Inc. Feb. 23, 1989, Report #88-12-2897		

Species	Asellus sp.		
Unit	mg/L		
Exposure Period	96	hour	
NOEC	= 0.73	..	
EC0		..	
EC50	> 0.73	..	
EC100		..	
Analyt. Monitoring	yes		
Method	other		
Year	1980		
GLP	yes		
Testsubstance	as prescribed by 1.1 - 1.4		
Remark	USEPA/OTS protocol, ASTM (1980)		

Reference	Monsanto Co., Springborn Life Sciences, Inc., Feb. 23, 1989, Report #88-12-2897
Species	Mysidopsis bahia
Unit	mg/L
Exposure Period	96 hour
NOEC	= 0.23 ..
EC0	..
EC50	> 0.23 ..
EC100	..
Analyt. Monitoring	yes
Method	other
Year	1980
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	USEPA/OTS protocol, ASTM (1980)
Reference	Monsanto Co., Springborn Life Sciences, Inc. May 12, 1989, Report #88-12-2894

Species	Palaemonetes pugio
Unit	mg/L
Exposure Period	96 hour
NOEC	= 0.23 ..
EC0	..
EC50	> 0.23 ..
EC100	..
Analyt. Monitoring	yes
Method	other
Year	1980
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	USEPA/OTS protocol, ASTM (1980)
Reference	Monsanto Co., Springborn Life Sciences, Inc., May 12, 1989, Report #88-12-2894

Species	other
Unit	mg/L
Exposure Period	96 hour
NOEC	= 0.23 ..
EC0	..
EC50	> 0.23 ..
EC100	..
Analyt. Monitoring	yes
Method	other
Year	1980
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Ampelisca abdita
Remark	USEPA/OTS protocol, ASTM (1980)
Reference	Monsanto Co., Springborn Life Sciences, Inc., May 12, 1989, Report #88-12-2894

4.3 Toxicity to Aquatic Plants e.g. Algae

Species	Selenastrum capricornutum
Endpoint	growth rate
Unit	mg/L
Exposure Period	96 hour
NOEC	> 0.78 ..
LOEC	..
EC0	..

EC10	..	
EC50	> 0.78	..
Analyt. Monitoring	yes	
Method	other	
Year	1978	
GLP	no data	
Testsubstance	as prescribed by 1.1 - 1.4	
Reference	Protocol: Miller, et al. 1978. EPA-600/9-78-018. USEPA, Corvallis, OR	
Reference	Reported in Felder, et al. 1986. Env. Tox. & Chem.5:777-784.	

Species	Scenedesmus subspicatus	
Endpoint	cell growth, growth rate inhibition	
Unit	mg/L	
Exposure Period	72	hour
NOEC	> 1.4	..
LOEC	..	
EC0	..	
EC10	..	
EC50	> 1.4	..
Analyt. Monitoring	yes	
Method	algal growth inhibition test according to 92/69/EEC	
Year	1996	
GLP	yes	
Testsubstance	as prescribed by 1.1 - 1.4	
Reference	Huls AG, 1996, Bestimmung der Auswirkungen von Vestinol OA, aud das Wachstum von Scenedesmus subspicatus 86.81.SAG (Algenwachstumshemmtest nach Richtlinie 92/69/EWG)	

Species	Activated sludge	
Endpoint	inhibition of test substance biodegradation	
Unit	mg/L	
Exposure Period	3 Stunde	
NOEC	> 352	..
LOEC	..	
EC0	..	
EC10	..	
EC50	> 352	..
Analyt. Monitoring	yes	
Method	EG-Nr. L 133 / 118 vom 30.5.1988	
Year	1996	
GLP	yes	
Test substance	Vestinol OA (di-(2-ethylhexyl) adipate)	
Reference	Huls AG, 1996, Bestimmung der Atmungshemmung von Belebtschlamm (EG-Nr. L 133 / 118 vom 30.5.1988) VESTINOL OA Abschlussbericht BH - 96 / 03.	

4.5.2 Chronic Tox. to Aquatic Invertebrates

Species	Daphnia magna	
Endpoint	other	
Unit	mg/L	
Exposure Period	21	day
NOEC	>0.77	..
LOEC	>0.77	..
EC50	..	
Analyt. Monitoring	yes	

Method	OECD Guideline 202 Teil II, 21-d semistatic reproduction test
Year	1996
GLP	yes
Testsubstance	Vestinol OA, di-(2-ethylhexyl) adipate
Remark	Endpoints were adult mortality and reproduction
Reference	Huls AG, 1996, Bestimmung der Auswirkungen von Vestinol OA auf die Reproduktionsrate von Vestinol OA (nach OECD-Guideline 202 Teil II), Abschlussbericht DL-167)

Species	Daphnia magna
Endpoint	other
Unit	mg/L
Exposure Period	21 day
NOEC	..
LOEC	..
EC50	..
MATC	= 0.024 ..0.052
Analyt. Monitoring	yes
Method	other
Year	1981
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Endpoints were adult mean length, mortality, and reproduction
Reference	Protocol: ASTM, 1981. Committee E47.01.
Reference	Reported in Felder, et al. 1986. Env. Tox. & Chem. 5:777-784.

5.1.1 Acute Oral Toxicity

Type	LD50
Species	rat
Value	= 9110 .. mg/kg
Method	other
Year	1951
GLP	no
Testsubstance	no data
Remark	Non-fasted animals (no number per group specified) were treated by oral gavage and observed for 14 days.
Reference	Smyth, H.F., Carpenter, C.P., and Weil, C.S. (1951). Range-Finding Toxicity Data: List IV, AMA Arch. Ind. Hyg. Occ. Med. 4:119-122.

Type	LD50
Species	rat
Value	> 20000 .. mg/kg
Method	other
Year	1976
GLP	no
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Similar method to Guideline 401. Five animals per sex per group were treated by oral gavage with 80, 160, 310, 630, 1250, 2500. 5000, 10000, or 20000 mg/kg of test substance in corn oil. Animals were observed for 14 days.
Remark	Mortality was 2/5 males at 10000 mg/kg. At 20000 mg/kg, 1/5 males and 1/5 females died. All surviving animals gained weight. Lethargy was noted in all animals. Loss of use of hind extremities occurred in 3/5 male and 3/5 female rats at 10000 mg/kg, and 1/5 male and 4/5 female rats at 20000 mg/kg. Five animals at 10000 mg/kg (3 male and 2 female), and 2 animals at 20000 mg/kg (one of each sex) were prostrate. Labored breathing was noted in 1 male at 5000 mg/kg, 1 male at 10000 mg/kg, and 1 male and 3 females at 20000 mg/kg. Increased lacrimation and wetting of hair at neck and hind quarters were noted in most animals at 10000 and 20000 mg/kg, and a few animals at 2500 and 5000 mg/kg. No indication of severity, day of onset, or duration of the condition was given. Excretion of bright yellow urine was noted in all groups given 630 mg/kg and higher doses. No gross pathology was reported.
Reference	Mason Research Institute Unpublished report. Single Dose Acute Toxicity Test of Di(2-ethyl hexyl) Adipate in Fischer 344 Rats and B6C3F1 Mice, Report No. MRI-TRA 16-76-34, 1976.

Type	LD50
Species	mouse
Value	= 10000 ..20000 mg/kg
Method	other
Year	1976
GLP	no
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Similar to Guideline 401. Five animals per sex per group were treated by oral gavage with 1250,

	2500, 5000, 10000, or 20000 mg/kg of test substance in corn oil. Animals were observed for 14 days post-treatment.
Remark	Mortality was 2/5 males at 10000 mg/kg, and 1/5 females at 20000 mg/kg. All surviving animals gained weight. Lethargy was noted in 2/5 males at 1250 mg/kg, 1/5 males at 2500 mg/kg, and all males at higher doses. Three of five female mice at 5000 and 1/5 females at 10000 and 20000 mg/kg were lethargic. Prostration was noted in 1 male and 1 female at 10000 mg/kg. One male at 10000 mg/kg had labored breathing. Wetness of the fur at the hindquarters was seen in all animals at 10000 and 20000 mg/kg. Most animals at 5000 mg/kg, 4/5 males at 10000 mg/kg, and all male and 1 female mouse at 20000 mg/kg had unsteady gait. There was no indication of the severity, onset, or duration of these conditions. No gross pathologic results were reported.
Reference	Mason Research Institute Unpublished report. Single Dose Acute Toxicity Test of Di(2-ethyl hexyl) Adipate in Fischer 344 Rats and B6C3F1 Mice, Report No. MRI-TRA 16-76-34, 1976.

Type	LD50
Species	rat
Value	> 7380 .. mg/kg
Method	other
Year	1967
GLP	no data
Testsubstance	no data
Remark	A group of 5 male and 5 female rats (strain not specified)were given a single oral dose of 7380 mg/kg of the neat test substance. Animals were observed daily for two weeks. Body weights were recorded prior to dosing and at the end of the observation period.
Remark	One animal died on Day 14. All other animals gained weight. No clinical signs of toxicity were reported. No gross pathology was reported.
Reference	Kolmar Research Center Unpublished report. The Toxicological Examination of Di-a-ethyl-hexyl-adipate (Wickenol 158), 1967.

5.1.2 Acute Inhalation Toxicity

Type	LC0
Species	rat
Exposure Time	8 hour
Value	..
Method	other
Year	1951
GLP	no
Testsubstance	no data
Remark	Animals (no number or sex specified) were exposed to a saturated vapor for 8 hours with no mortality.
Reference	Smyth, H.F., Carpenter, C.P., and Weil, C.S. (1951). Range-Finding Toxicity Data: List IV, AMA Arch. Ind. Hyg. Occ. Med. 4:119-122.

5.1.3 Acute Dermal Toxicity

Type	LD50
Species	rabbit
Value	= 15029 .. mg/kg
Method	other
Year	1951
GLP	no
Testsubstance	no data
Remark	The shaved skin of rabbits was treated with the neat test substance. There was no indication of occluding the area or the length of treatment. The observation period was 14 days post-treatment. No data were provided other than the estimated LD50.
Reference	Smyth, H.F., Carpenter, C.P., and Weil, C.S. (1951). Range-Finding Toxicity Data: List IV, AMA Arch. Ind. Hyg. Occ. Med. 4: 119-122.

Type	LD50
Species	rabbit
Value	> 8670 .. mg/kg
Method	other
Year	1967
GLP	no
Testsubstance	no data
Remark	The intact skin of one male and the abraded skin of one female rabbit (2.2-3.5 kg) were treated with 0, 3600, 5530, or 8660 mg/kg for 24 hours under an occlusive patch. Body weights, food consumption, and clinical signs of toxicity were checked daily. Urine was checked for the presence of protein, reducing substances, or blood pigments. Blood morphology was also evaluated.
Remark	Minimal irritation (erythema) was observed; the duration of the reaction was dose-dependent. No clinical signs of toxicity were noted. Feed consumption and body weights were reduced at the highest dose level. No protein, blood, or reducing substances were noted in the urine. Hematologic parameters were in the normal range.
Reference	Kolmar Research Center Unpublished report. The Toxicological Examination of Di-a-ethyl-hexyl-adipate (Wickenol 158), 1967.

5.1.4 Acute Toxicity, Other Routes

Type	LD50
Species	rat
Route of Administration	i.p.
Exposure Time	
Value	> 47619 .. mg/kg
Method	other
Year	1946
GLP	no
Testsubstance	no data
Remark	Thirty-eight rats (sex and strain not specified) were treated with doses of 8076-47619 mg/kg by i.p. injection. No mortality was noted. Clinical observations were not reported.
Reference	Eastman Kodak Company, Unpublished report, 1946.

5.2.1 Skin Irritation

Species	rabbit
Result	not irritating
Classification	not irritating
Method	Draize-Test
Year	1951
GLP	no
Testsubstance	no data
Reference	Smyth, H.F., Carpenter, C.P., and Weil, C.S. (1951). Range-Finding Toxicity Data: List IV, AMA Arch. Ind. Hyg. Occ. Med. 4:119-122.

Species	rabbit
Result	slightly irritating
Classification	irritating
Method	other
Year	1967
GLP	no
Testsubstance	no data
Remark	The neat test substance was applied to the abraded and intact skin of 6 albino rabbits for 24 hours and the irritation observed for 48 hours after patch removal. The entire trunk was wrapped with a rubberized cloth.
Remark	Slight erythema was observed when the patch was removed, and the severity decreased in all animals by 72 hours. There was no apparent difference between abraded and intact skin. The Primary Irritation Index was 0.83.
Reference	Kolmar Research Center Unpublished report. The Toxicological Examination of Di-a-ethyl-hexyl-adipate (Wickenol 158), 1967.

5.2.2 Eye Irritation

Species	rabbit
Result	slightly irritating
Classification	not irritating
Method	other
Year	1951
GLP	no
Testsubstance	no data
Remark	Rabbit eyes were treated with 0.5 ml of undiluted test substance and the area of corneal necrosis scored.
Reference	Smyth, H.F., Carpenter, C.P., and Weil, C.S. (1951). Range-Finding Toxicity Data: List IV, AMA Arch. Ind. Hyg. Occ. Med. 4:119-122.

Species	rabbit
Result	not irritating
Classification	not irritating
Method	Draize-Test
Year	1967
GLP	no
Testsubstance	no data
Remark	A volume of 0.1 ml was instilled into one eye of 6 albino rabbits. The other eye remained untreated. The eyes were examined at 24, 48, and 72 hours

	after treatment.
Remark	No irritation was observed at any timepoint.
Reference	Kolmar Research Center Unpublished report. The Toxicological Examination of Di-a-ethyl-hexyl-adipate (Wickenol 158), 1967.

5.3 Sensitization

Type	other
Species	guinea pig
Result	not sensitizing
Classification	not sensitizing
Method	other
Year	1967
GLP	no
Testsubstance	no data
Remark	Ten male guinea pigs were treated with intracutaneous injections of 0.1% DEHA in olive oil. Animals were injected three times per week for 3 weeks and were challenged after a two week rest period.
Remark	The area and height of the reaction was measured 24 hours after the challenge dose. The average area and height of the reaction at challenge were smaller than during induction.
Reference	Kolmar Research Center Unpublished report. The Toxicological Examination of Di-a-ethyl-hexyl-adipate (Wickenol 158), 1967.

5.4 Repeated Dose Toxicity

Species	rat
Strain	Fischer 344
Sex	male/female
Route of Administration	oral feed
Exposure Period	14-days
Frequency of Treatment	daily
Post Exposure Observ. Period	1 day
Doses	0.31 (males only), 0.63, 1.25, 2.50, 5.00, 10.00 (females only) %
Control Group	yes, concurrent vehicle
NOEL =	2.5 .. %
LOEL =	5 .. %
Method	other
Year	1980
GLP	no
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Five animals per sex per group were given the test substance in the feed for 14 consecutive days. Animals were observed for twice daily during the treatment period and for one day after dosing terminated. Body weights were measured at initiation and termination. Feed consumption was measured weekly. A complete gross necropsy was performed on each animal.
Remark	Details not provided on frequency of feed consumption measurements, but weekly measurements assumed. Histopathology was not performed.

Result	One female rat at the 10% dose level died. Weight gain was decreased at least 25% in male rats fed 5.00% and in female rats fed 2.50% test substance. Female rats at the 10.00% dose level lost weight, and feed consumption was reduced at the 5.00 and 10.00% dose levels. All animals fed 5.00% and 10.00% test substance in the diet looked emaciated. These animals also appeared to consume minimal amounts of food. Female rats in the 10.00% group were lethargic with signs of piloerection, increased wetness of hair, and ataxia. At necropsy, gray-white casts were observed in the liver of 2 10.00% females. Atrophy of the spleen was also noted in one 10.00% animal. Gray-white casts were noted in one 2.50% and 5.00% female. None of these findings were considered to be compound-related.
Reference	Mason Research Institute Unpublished report. Repeated Dose Acute Toxicity Test of Di(2-ethylhexyl) Adipate in Fisher 344 Rats and B6C3F1 Mice, Report No. MRI-TRA 31-76-54, 1976.

Species	mouse
Strain	B6C3F1
Sex	male/female
Route of Administration	oral feed
Exposure Period	14 days
Frequency of Treatment	daily
Post Exposure Observ. Period	1 day

Doses	0.31 (males only), 0.63, 1.25, 2.50, 5.00, 10.00 (females only) %
Control Group	yes, concurrent vehicle

NOEL =	0.63	..	%
LOEL =	2.5	..	%

Method	other
Year	1980
GLP	no
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Five animals per sex per group were given feed with the test substance for 14 days. Animals were observed twice daily and observed for 1 day post dosing for signs of toxicity. Body weights were measured at initiation and termination. Feed consumption was measured (presumably) weekly. A complete gross necropsy was performed at termination.
Result	All female mice died at the 10.00% dose level. Prior to death, all 10.00% animals were lethargic and emaciated with hunched posture and increased wetness of hair. Weight loss occurred in male mice at 5.00%, and in female mice at 2.50%. Feed consumption was reduced in female mice given 10.00% test substance. No compound-related gross pathology was observed at necropsy.
Reference	Mason Research Insitute Unpublished report. Repeated Dose Acute Toxicity Test of Di(2-ethylhexyl) Adipate in Fischer 344 and B6C3F1 Mice, Report No. MRI-TRA 31-76-54, 1976.

Species	rat		
Strain	Fischer 344		
Sex	male/female		
Route of Administration	oral feed		
Exposure Period	13 weeks		
Frequency of Treatment	daily		
Post Exposure Observ. Period	NA		
Doses	0.16, 0.31, 0.63, 1.25, 2.5 %		
Control Group	yes, concurrent vehicle		
NOEL =	0.63	..	%
LOEL =	1.25	..	%
Method	other		
Year	1980		
GLP	no		
Testsubstance	as prescribed by 1.1 - 1.4		
Remark	Ten animals per sex per group were fed diets containing the test substance for 13 weeks. Clinical observations were performed twice daily. Body weights and feed consumption were measured weekly. At termination, selected tissues (not specified) were preserved and evaluated histologically.		
Result	One female rat died at 0.16%. This was not considered to be compound-related. Weight gain in male and female rats was decreased by at least 11% at the 2.5% dose level, and by at least 11% in male rats at the 0.63 and 1.25% dose levels. Neither compound-related histopathology nor reduction in feed consumption were noted (data not shown).		
Reference	National Toxicology Program. Carcinogenesis Bioassay of Di(2-ethylhexyl) Adipate, Technical Report No. 212, 1980.		

Species	mouse		
Strain	B6C3F1		
Sex	male/female		
Route of Administration	oral feed		
Exposure Period	13 weeks		
Frequency of Treatment	daily		
Post Exposure Observ. Period	NA		
Doses	0.16, 0.31, 0.63, 1.25, and 2.50 %		
Control Group	yes, concurrent vehicle		
NOEL =	0.16	..	%
LOEL =	0.31	..	%
Method	other		
Year	1980		
GLP	no		
Testsubstance	as prescribed by 1.1 - 1.4		
Remark	Ten animals per sex per group were fed diets containing the test substance for 13 weeks. Clinical observations were performed twice each day. Body weights and feed consumption were measured weekly. At termination, selected tissues (not specified) were preserved and examined		

	microscopically. No clinical pathology was performed.
Result	One female mouse died accidentally. Weight gain was decreased by at least 10% at the 0.31% and higher dose levels. Neither compound-related histopathologic effects nor a reduction in feed consumption were noted (data not shown).
Reference	National Toxicology Program. Carcinogenesis Bioassay of Di(2-ethylhexyl) Adipate, Technical Report No. 212, 1980.

Species	rat
Strain	no data
Sex	no data
Route of Administration	oral feed
Exposure Period	90 days
Frequency of Treatment	daily
Post Exposure Observ. Period	NA

Doses	160, 610, 2920, 4740 mg/kg
Control Group	no data specified

NOEL =	610	..	mg/kg
LOEL =	2920	..	mg/kg

Method	other
Year	1951
GLP	no
Testsubstance	no data
Remark	Ten animals per group (number per sex not specified) were given feed containing the test substance for 90 days. Body weight and feed consumption determined, but the frequency was not specified. Liver and kidney weights were measured at necropsy. The liver, kidneys, spleen, and testis were examined microscopically.
Result	Mortality occurred at 4740 mg/kg, but the number of deaths was not specified. Reduced growth and appetite were observed at 2920 mg/kg. Altered organ weight (either kidney or liver) was noted. Microscopic lesions occurred in at least one tissue at 2920 mg/kg. No details or data shown for any results were provided.
Reference	Smyth, H.F., Carpenter, C.P., and Weil, C.S. (1951). Range-Finding Toxicity Data: List IV, AMA Arch. Ind. Hyg. Occ. Med. 4:119-122.

Species	rat
Strain	Fischer 344
Sex	male/female
Route of Administration	oral feed
Exposure Period	3 weeks
Frequency of Treatment	daily
Post Exposure Observ. Period	2 weeks

Doses	0.1, 1.2, 2.5 %
Control Group	Yes, concurrent vehicle

NOEL =	0.1	..	%
LOEL =	1.2	..	%

Method	other

Year	1982
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Groups of 12 male and 12 female rats were treated with the test substance in the diet. Animals were treated for up to 3 weeks followed by 2 weeks of recovery. Body weights and feed consumption were measured weekly. Clinical signs of toxicity were recorded at least once daily. Four rats per sex per group were sacrificed after 1 week; five rats per sex per group were sacrificed after 3 weeks; and the remaining animals were sacrificed after 5 weeks. At sacrifice, blood was collected for clinical pathology (alanine transaminase activity, alkaline phosphatase activity, total cholesterol levels, and triglyceride levels). The liver was perfused with saline and portions retained for histopathology, cytochemical localization of catalase, electron microscopy, and determinations of catalase and carnitine acetyltransferase activities. The kidneys, brain, spleen, testes, and thyroid gland were weighed and preserved. Bone marrow from the femur was also preserved.
Result	Male rats treated with the mid- and high-doses of the test substance had significantly lower body weights, especially during recovery. The body weights of female rats were unaffected. Feed consumption by the high-dose male rats was also significantly lower than the control group during week one. No other differences in feed consumption occurred. No clinical signs of toxicity were noted. Relative (to body weight) liver weights of mid- and high-dose rats at 1 and 3 weeks were significantly higher than for the control group. No differences were noted at 5 weeks (following recovery), and no changes were noted in the low-dose group. Changes in relative kidney weight at 1 and 3 weeks were considered to be incidental. Triglyceride levels in mid- and high-dose male rats at 1 week, and high-dose male rats at 3 weeks were significantly lower than in the control group. No differences were noted at 5 weeks or in other male or female groups. Cholesterol levels in the mid- and high-dose groups (both male and female) at 1 week and high-dose at 3 weeks were also significantly lower than in the control group. No differences were noted at 5 weeks or in the low-dose group. No other changes in clinical pathology were noted. Catalase activity was significantly higher in high-dose animals after 1 week, but was at control levels by 5 weeks. CAT activity was significantly higher in mid- and high-dose groups beginning at 1 week. Activity remained high even after recovery. Hepatocellular hypertrophy was noted in high-dose male rats at 1 and 3 weeks. Hypertrophy was noted only in a few high-dose female rats at 3 weeks. No lesions were found after recovery.
Reference	CMA Unpublished report. Toxicological Effects of Diethylhexyl Adipate (MRI Project 7343-B), 1982.

Species	rat
Strain	Fischer 344
Sex	male/female
Route of Administration	oral feed
Exposure Period	3 weeks
Frequency of Treatment	daily
Post Exposure Observ. Period	NA
Doses	0.1, 0.6, 1.2, 2.5 %
Control Group	Yes, concurrent vehicle
NOEL =	0.1 .. %
LOEL =	0.6 .. %
Method	other
Year	1986
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Groups of 5 male and 5 female rats were treated for 3 weeks with the test substance in the diet. Body weights and feed consumption were measured weekly. Clinical signs of toxicity were recorded at least once daily. All animals were sacrificed after 3 weeks, and blood was collected for clinical pathology (total cholesterol levels and triglyceride levels). The liver, kidneys, and testes were excised and weighed. Portions of each tissue were preserved in buffered formalin for histopathology. Another portion of the liver was fixed in osmium tetroxide for electron microscopy, while a third portion was homogenized for biochemical analyses (palmitoyl CoA oxidation, lauric acid 11-, 12- hydroxylation, and microsomal protein).
Result	Male and female rats fed 2.5% DEHA had lower lower body weights compared with controls. Male rats fed 2.5% DEHA had lower feed consumption compared with controls. Occasional lower body weights were also noted for male rats fed 0.1 and 1.2% DEHA. Absolute and relative (to body weight) liver weights for the 1.2 and 2.5% male, and 0.6, 1.2, and 2.5% female rats were higher than for the controls. Absolute kidney weights for the 2.5% male group, and relative (to body weight) kidney weights for the 1.2 and 2.5% male and female groups were higher than for the controls. No significant differences in testes weights were noted. Triglyceride and cholesterol levels for the treated groups were not significantly different than for the controls. Palmitoyl CoA oxidation was significantly higher for the 1.2% male, and 2.5% male and female groups. Lauric acid 11-hydroxylase activity was unchanged, but 12-hydroxylase activity was significantly higher for the 0.6, 1.2, and 2.5% male groups and 2.5% female group. Microsomal protein was significantly higher for the 2.5% male group. Reduced cytoplasmic basophilia was seen for the 1.2% males and 2.5% males and females. Increased cytoplasmic eosinophilia was noted for the 2.5% male group. Increase mitotic activity and focal

	necrosis was limited to the 2.5% groups. Slightly increased peroxisome proliferation was evident for the 0.6% male and female animals. No lesions were observed for the kidneys or testes.
Reference	CMA Unpublished report. A 21-Day Feeding Study Of Diethylhexyl Adipate To Rats: Effects On The Liver And Liver Lipids (BIBRA Project 3.0542), 1986.

Species	rat
Strain	Fischer 344
Sex	female
Route of Administration	oral feed
Exposure Period	3 weeks
Frequency of Treatment	daily
Post Exposure Observ. Period	14 days

Doses	0.012, 0.12, 1.2, 2.5 %
Control Group	Yes, concurrent vehicle

NOEL =	0.012	..	%
LOEL =	0.12	..	%

Method	other
Year	1989
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Groups of 20 female rats were treated for 3 weeks with the test substance in the diet. An additional 10 rats were treated with 2.5% DEHA for 21 days followed by control diet for 14 days. A control group of 10 rats was also maintained for this time. Body weights and feed consumption were measured weekly. Clinical signs of toxicity were recorded at least once daily. Five animals per group were sacrificed after 3, 7, 21, or 35 days for cell proliferation measurements following injection with ^{3}H-thymidine. An additional 5 animals per group were sacrificed after 21 and 35 days for histopathology, clinical chemistry, and biochemical analyses. Blood was collected for analysis of total cholesterol, triglyceride, total bilirubin levels, alanine transaminase activity, and alkaline phosphatase activity. The liver, kidneys, and spleens were excised and weighed. Portions of the liver were preserved in buffered formalin for histopathology. Another portion of the liver was fixed in osmium tetroxide for electron microscopy, while a third portion was homogenized for biochemical analyses (β-galactosidase, succinate dehydrogenase, palmitoyl CoA oxidation, lauric acid 11, 12 hydroxylation, and microsomal protein).
Result	Female rats fed 2.5% DEHA had lower feed consumption and lower body weights compared with the controls. Female rats fed 1.2% DEHA occasionally had lower body weights than did the controls. Weight gain during recovery period was higher than controls. Absolute and relative (to body weight) liver weights for the 0.012, 1.2 and 2.5% rats at day 21 were higher than for the controls. No differences in absolute kidney

weight were seen, but the 2.5% group had significantly higher relative kidney weights than did controls. The 1.2 and 2.5% groups had lower absolute spleen weights, but there were no differences in relative spleen weight. After recovery, no differences were seen in absolute organ weight, but relative kidney, liver, and spleen weights were higher in the 2.5% group than in the controls.
Triglyceride and cholesterol levels for the treated groups were not significantly different than for the controls. Total bilirubin was decreased for the 2.5% group, but the biological significance of this change is not clear. There was no change in alanine transaminase activity, and a decrease in alkaline phosphatase activity for the 2.5% group, the biological significance of which is not clear. There were no differences after the animals were allowed to recover.
After 3 days on test, there was no increase in cell proliferation although there was an increase in DNA content in the liver per 100 g body weight for the 2.5% group. However, liver weight was also increased for this group. After 7 days on test, the DNA content of the liver per 100 g body weight was increased for the 1.2 and 2.5% groups. Relative liver to body weights were also increased for these groups. There was a decrease in ^{3}H-thymidine incorporation into the liver for the 1.2 and 2.5% groups. Similar results were reported for animals sacrificed after 21 days on test. After 14 days of recovery the DNA content per 100 grams body weight was higher for the 2.5% group, and absolute and relative liver weights were higher. However, the amount of ^{3}H-thymidine incorporation was significantly lower than for the controls.
Palmitoyl CoA oxidation was higher for the 1.2 and 2.5% groups, but only the 2.5% dose level was statistically significant. Lauric acid 11- and 12-hydroxylase activity were significantly higher for the 2.5% group. Succinate dehydrogenase was higher for the 2.5% group, but there was no consistent change in galactosidase activity. Cytochrone P-450 content was unchanged, but there were increases in 7-ethoxycoumarin O-deethylase and NADPH-cytochrome c reductase activities.
Periportal vacuolization was reduced for the 1.2 and 2.5% groups, and cytoplasmic basophilia was reduced for the 2.5% group. No abormalities were seen for the kidneys and spleen. Slightly increased peroxisome proliferation was evident for the one 0.12% rat, and all 1.2 and 2.5% rats. These were not apparent after 14 days of recovery.

Reference: CMA Unpublished report. A Study Of The Hepatic Effects Of Diethylhexyl Adipate In The Mouse And Rat (BIBRA Project 3.0709), 1989.

Species: mouse
Strain: B6C3F1
Sex: female
Route of Administration: oral feed

Exposure Period	3 weeks
Frequency of Treatment	daily
Post Exposure Observ. Period	14 days
Doses	0.012, 0.12, 1.2, 2.5 %
Control Group	Yes, concurrent vehicle
NOEL =	0.12 .. %
LOEL =	1.2 .. %
Method	other
Year	1989
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Groups of 30 female mice were treated for 3 weeks with the test substance in the diet. An additional 30 mice were treated with 2.5% DEHA for 21 days followed by control diet for 14 days. A control group of 30 mice was also maintained for this time. Body weights and feed consumption were measured weekly. Clinical signs of toxicity were recorded at least once daily. Five animals per group were sacrificed after 3, 7, 21, and 35 days for cell proliferation measurements following injection with ^{3}H-thymidine. An additional 15 animals per group were sacrificed after 21 and 35 days for histopathology, clinical chemistry, and biochemical analyses. Blood was collected for analysis of total cholesterol, triglyceride, total bilirubin levels, alanine transaminase activity, and alkaline phosphatase activity. The liver, kidneys, and spleens were excised and weighed. Portions of the liver were preserved in buffered formalin for histopathology. Another portion of the liver was fixed in osmium tetroxide for electron microscopy, while a third portion was homogenized for biochemical analyses (β-galactosidase, succinate dehydrogenase, palmitoyl CoA oxidation, lauric acid 11, 12 hydroxylation, and microsomal protein).
Result	Female mice fed 2.5% DEHA had lower body weights, although food consumption was occasionally higher than for the controls. Weight gain during recovery period was higher than controls. Absolute and relative (to body weight) liver weights for the 1.2 and 2.5% mice at day 21 were higher than for the controls. Relative kidney weight for the 1.2 and 2.5% groups were significantly higher than controls, and absolute and relative (to body weight) spleen weights for the 2.5% group were significantly lower than controls. After recovery, the absolute organ weight for liver, kidney, and spleen were lower than control, but relative kidney, liver, and spleen weights were no significantly different. Triglyceride were lower and cholesterol levels higher for the 1.2 and 2.5% groups compared with the controls. There was no change in total bilirubin. Alanine transaminase activity for the 1.2 and 2.5% groups, and alkaline phosphatase activity for the 0.12 and 2.5% groups were higher than for the controls. Alkaline phosphatase

activity was significantly higher than the controls after the end of the recovery period. After 3 days on test, the 1.2 and 2.5% groups had significant lower DNA content per gram of liver, and the 1.2% group had an increase in cell proliferation (^{3}H-thymidine incorporation). The absolute and relative liver weights were higher for the 1.2 and 2.5% groups. After 7 days on test, the 1.2 and 2.5% groups also had lower DNA content per gram liver, but the 2.5% group had higher DNA per gram body weight. Both groups had significantly increased cell proliferation. The absolute and relative liver weights were higher for the 1.2 and 2.5% groups. After 21 days on test, the 0.12, 1.2, and 2.5% groups had significant lower DNA content per gram of liver, and the DNA content per gram body weight was lower for the 0.12% group. Cell proliferation was higher for the 1.2 and 2.5% groups. The absolute and relative liver weights were higher for the 1.2 and 2.5% groups. After 14 days of recovery, the DNA content per 100 grams of body weight was higher for the 2.5% group. There were no other differences.

Palmitoyl CoA oxidation, β-galactosidase, and 7-ethoxyresorufin O-deethylase activities were higher for the 1.2 and 2.5% groups compared with the control. Lauric acid 11- and 12-hydroxylase activity were significantly higher for the 0.12, 1.2, and 2.5% groups. Succinate dehydrogenase activity was unchanged. Cytochrone P-450 content was higher for the 0.12, 1.2, and 2.5% groups. Microsomal and homogenate protein were higher for the 2.5% group. Glucose-6-phosphatase activity was significantly lower for the 0.012, 1.2, and 2.5% groups compared with the control. After the recovery period, only β-galactosidase activity was higher for the treated group. No other significant differences were observed. Periportal vacuolization was reduced for the 1.2 and 2.5% groups. No abormalities were seen for the kidneys and spleen. Increased peroxisome proliferation was evident for all 1.2 and 2.5% rats. These were not apparent after 14 days of recovery.

Reference: CMA Unpublished report. A Study Of The Hepatic Effects Of Diethylhexyl Adipate In The Mouse And Rat (BIBRA Project 3.0709), 1989.

Species	rat
Strain	Fischer 344
Sex	female
Route of Administration	oral feed
Exposure Period	6 weeks
Frequency of Treatment	daily
Post Exposure Observ. Period	14 days
Doses	0.025, 0.12, 0.25, 1.2, 2.5 %
Control Group	Yes, concurrent vehicle

NOEL =	<0.025	..	%
LOEL =	0.025	..	%

Method	other
Year	1995
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Groups of 25 female rats were treated for up to 6 weeks with the test substance in the diet. Groups of 5 rats were sacrificed on days 3, 7, 14, 28, and 42. An additional 5 rats were treated with 2.5% DEHA for 42 days followed by control diet for 14 days. A control group of 5 rats was also maintained for this time. Body weights and feed consumption were measured weekly. Clinical signs of toxicity were recorded at least once daily. At each time point, animals were sacrificed for cell proliferation measurements following implantation of osmotic pumps with BrdU. Blood was collected for determination of alanine transaminase (ALT) and aspartate transaminase (AST) activities. The liver, kidneys, and spleen were excised and weighed; sections of these organs were preserved for histopathology. Liver samples were collected for determination of cell proliferation and peroxisomal enzyme proliferation (palmitoyl CoA oxidation). Samples were also prepared for transmission electron microscopy.
Result	All groups of rats fed DEHA had lower body weights, although food consumption for these groups was not different from the controls. Weight gain during recovery period was not different from controls. Absolute and relative (to body weight) liver weights for the 2.5% rats were higher than for the controls on Days 7, 14, 28, and 42. Absolute liver weights for the 1.2% rats were higher than for the controls on Day 42, and relative liver weights were higher on Days 7, 14, 28, and 42. No differences in absolute kidney weight were seen, but relative kidney weight was higher for the 1.2% group on Days 14 and 42, and for the 2.5% group on Day 42. Relative kidney weight for the 0.025% group was lower than control on Day 3. Absolute spleen weights were higher than control for the 0.25% group on Day 3, and for the 2.5% group on Day 14. Absolute spleen weight was lower than control for the 2.5% group on Day 42. Relative spleen (to body weight) weight was higher than control for the 0.25% group on Days 3 and 14. Relative spleen weights for the 0.12, 1.2, and 2.5% groups were also significantly higher than control on Day 14. There were no significant differences in organ weight after recovery. There were no significant differences in AST activity among groups, and a significant increase in ALT activity for the 1.2% group on Day 28. No differences were observed after recovery. After 3 days on test, the 1.2% group had significant higher palmitoyl CoA (Pal CoA) oxidation activity per μg DNA than did the control. On Day 7, Pal CoA oxidation per gram liver and per μg DNA for the 2.5% group was significantly higher than for the control. On Day

14, Pal CoA activity per gram tissue for all treated groups was significantly higher than for the controls, and Pal CoA activity per µg DNA was higher than control for the 0.25 and 1.2% groups. On Day 28, Pal CoA activity per gram tissue and per µg DNA was higher than control for the 0.25, 1.2, and 2.5% groups. No significant differences in Pal CoA activity were seen on Day 42 or after recovery.
Evidence of cell proliferation in the liver was observed on Day 3 for the 1.2 and 2.5% groups (all liver lobes; median lobe for 1.2% group; caudate lobe for the 2.5% group), and on Day 7 for the 0.12% group (all liver lobes; median lobe). The labeling index for the 2.5% group was significantly lower than control on Day 14 (all liver lobes; median lobe; lateral lobe) and on Days 28 and 42 for the right lateral lobe. There was no evidence of cell proliferation after recovery.
Hepatocellular hypertrophy was observed in 1/5 animals of the 0.025% group on Days 3, 7, and 28. Hypertrophy was observed for at least one animal from the 0.12% group on Days 3, 7, 14, and 42, and hyperplasia was seen in one animal on Day 3 and 28. Hypertrophy was seen in all animals of the 0.25, 1.2, and 2.5% groups on Day 3, most animals on Day 7, and an occasional animal at the later time points. Hyperplasia was seen in on animal from the 1.2% group on Day 14, and one animal from the 1.2 and 2.5% groups on Day 42. No lesions were observed after recovery.

Reference: CMA Unpublished report. Studies Of The Hepatic Effects Of Diethylhexyl Adipate (DEHA) In The Mouse And Rat (SRI Project 2759-S01-91), 1995.

Species	mouse
Strain	B6C3F1
Sex	female
Route of Administration	oral feed
Exposure Period	6 weeks
Frequency of Treatment	daily
Post Exposure Observ. Period	14 days
Doses	0.025, 0.12, 0.25, 1.2, 2.5 %
Control Group	Yes, concurrent vehicle
NOEL =	<0.025 .. %
LOEL =	0.025 .. %
Method	other
Year	1995
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4

Remark: Groups of 25 female mice were treated for up to 6 weeks with the test substance in the diet. Groups of 5 mice were sacrificed on days 3, 7, 14, 28, and 42. An additional 5 mice were treated with 2.5% DEHA for 42 days followed by control diet for 14 days. A control group of 5 mice was also maintained for this time. Body weights and feed consumption were measured weekly. Clinical signs

	of toxicity were recorded at least once daily. At each time point, animals were sacrificed for cell proliferation measurements following implantation of osmotic pumps with BrdU. Blood was collected for determination of alanine transaminase (ALT) and aspartate transaminase (AST) activities. The liver, kidneys, and spleen were excised and weighed; sections of these organs were preserved for histopathology. Liver samples were collected for determination of cell proliferation and peroxisomal enzyme proliferation (palmitoyl CoA oxidation). Samples were also prepared for transmission electron microscopy.
Result	Significantly lower body weights for the 2.5% group were noted at Weeks 3, 4, 5, and 6. No differences were seen after recovery. The mean body weight for the 1.2% group was significantly higher than control at Week 2. Weight gains for the 0.25, 1.2, and 2.5% groups were higher than for control at Week1, but lower than control at Week 3. No differences in feed consumption were seen among groups except for the 2.5% group which had higher feed consumption at Week 4 compared with the control. Significantly higher absolute and relative (to body weight) liver weights for the 1.2 and 2.5% were seen at each time point. In addition, absolute liver weight for the 0.25% group was higher than control on Day 14, and relative liver for this group was higher on Days 14 and 42. Absolute liver weight for the 0.12% group was higher than for control on Days 7, 14, and 28, and relative liver weight for this group was higher on Days 7 and 28. Absolute liver weight for the 0.025% mice was higher than for the controls on Day 3, and relative liver weights were higher on Days 3 and 42. Higher absolute kidney weight were seen for an occasional group (0.12% group on Days 14 and 42; 0.25% group on Day 7; 1.2% group on Day 42; 2.5% group on Days 7 and 14). Higher mean relative (to body weight) kidney weight was noted for the 2.5% group at each time point, and for the 1.2% group on Days 7 and 14. Higher relative kidney weight was seen for the 0.25% group on Days 14 and 42, while higher relative kidney weight was seen for the 0.12% group on Day 14. Absolute and relative (to body weight) spleen weight for the 1.2% group was higher than for control on Day 42. No other differences in spleen weight were observed. There were no significant differences in organ weight after recovery. Mean AST and ALT activities for the 2.5% group were significantly higher than for controls on Day 42. No differences were observed after recovery. After 3 days on test, the 0.12% group had significant higher palmitoyl CoA (Pal CoA) oxidation activity per gram tissue compared with the control. On Day 7, Pal CoA oxidation per gram liver for the 2.5% group was significantly higher than for the control. On Day 14, Pal CoA activities per gram tissue, per µg DNA, and per mg protein for the 1.2 and 2.5% groups were

	significantly higher than for the controls. On Day 28, Pal CoA activities per gram tissue, per µg DNA, and per mg protein for all treated groups were higher than control. Although on Day 42, the Pal CoA activities for all groups were also higher than controls, significant differences were noted only for the 2.5% group when expressed per gram tissue and mg protein, and for the 1.2 and 2.5% groups when expressed per µg DNA. No significant differences in Pal CoA activity were seen after recovery. Evidence of cell proliferation in the liver was observed on Day 7 for the 2.5% group (all liver lobes; left lateral lobe), on Day 14 for the 1.2% group (median lobe), on Day 28 for the 0.025 and 0.12% groups (caudate lobe), and on Day 42 for the 2.5% group (median lobe). There was no evidence of cell proliferation after recovery. Hepatocellular hypertrophy or karyomegaly was observed in all animals of the 1.2 and 2.5% groups at each time point. No lesions were observed in other groups. No lesions were observed after recovery.
Reference	CMA Unpublished report. Studies Of The Hepatic Effects Of Diethylhexyl Adipate (DEHA) In The Mouse And Rat (SRI Project 2759-S01-91), 1995.

Species	rat
Strain	Fischer 344
Sex	female
Route of Administration	dietary
Exposure Period	13 weeks
Frequency of Treatment	daily, 7 days per week
Post Exposure Observ. Period	NA
Doses	0.15, 0.3, 0.6, 1.2, 2.5, and 4.0%
Control Group	yes, concurrent vehicle
NOEL	= 0.15 … %
LOEL	= 0.3 … %
Method	other
Year	1997
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Female rats were treated with diets containing the test substance. A control group received diet without the test substance. Body weight and food consumption were measured. During Weeks 1, 4, and 13, 5-8 animals per group were implanted with osmotic pumps containing BrdU. At the end of Weeks 1, 4, and 13, these animals were sacrificed and sections of the liver evaluated for cell proliferation. Another section of liver was used for biochemical analysis (palmitoyl CoA oxidation, lauric acid 11- and 12-hydroxylase activity, cytocrhome P-450 content, and microsomal protein).
Result	Body weights and food consumption for the 2.5 and 4.0% groups were significantly lower than for the controls. Relative (to body weight) liver weights

	were higher than for the controls for the 1.2, 2.5, and 4.0% groups at Weeks 1 and 4, and for the 0.6, 1.2, 2.5, and 4.0% groups at Week 13. The cell labeling indices for the 2.5 and 4.0% groups were significantly higher than the controls at Week 1, but not thereafter. Pal CoA oxidation was higher than the controls for the 0.6, 1.2, 2.5, and 4.0% groups at each time period. Lauric acid 11-hydroxylation for the 0.3, 0.6, 2.5, and 4.0% groups were significantly higher than control at Week 1, with the 2.5 and 4.0% groups higher at Week 4, and the 4.0% group higher at Week 13. Lauric acid 12-hydroxylation for the 0.3, 0.6, 1.2, 2.5, and 4.0% groups were significantly higher than control at Weeks 1 and 13, with the 2.5 and 4.0% groups higher at Week 4. Cytochrome P-450 for the 4.0% group was higher than control at Week 1, but not thereafter.
Reference	Lake, B.G., Price, R.J., Cunninghame, M.E., and Walters, D.G. (1997). Comparison Of The Effects Of Di(2-Ethylhexyl)Adipate On Heptic Peroxisome Proliferation And Cell Replication In The Rat And Mouse, Toxicology 123: 217-226.

Species	mouse
Strain	B6C3F1
Sex	female
Route of Administration	dietary
Exposure Period	13 weeks
Frequency of Treatment	daily, 7 days per week
Post Exposure Observ. Period	NA

Doses	0.15, 0.3, 0.6, 1.2, and 2.5%
Control Group	yes, concurrent vehicle

NOEL	<	0.15	…	%
LOEL	=	0.15	…	%

Method	other
Year	1997
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Female mice were treated with diets containing the test substance. A control group received diet without the test substance. Body weight and food consumption were measured. During Weeks 1, 4, and 13, 5-8 animals per group were implanted with osmotic pumps containing BrdU. At the end of Weeks 1, 4, and 13, these animals were sacrificed and sections of the liver evaluated for cell proliferation. Another section of liver was used for biochemical analysis (palmitoyl CoA oxidation, lauric acid 11- and 12-hydroxylase activity, cytocrhome P-450 content, and microsomal protein).
Result	Body weights for the 1.2 and 2.5% groups were significantly lower than for the controls at Weeks 4 and 13. There were no differences in food consumption. Relative (to body weight) liver weights were higher than for the controls for the 0.6, 1.2, and 2.5% groups at Weeks 1 and 4, and for the 1.2 and 2.5% groups at Week 13. The cell labeling indices for the 0.6, 1.2, and 2.5% groups

were significantly higher than the controls at Week 1, and the indices for the 1.2 and 2.5% groups were higher at Weeks 4 and 13. Pal CoA oxidation was higher than the controls for the 0.6, 1.2, and 2.5% groups at Weeks 1 and 4, and the pal CoA activities for all treated groups were higher than controls at Week 13. Lauric acid 11-hydroxylation for the 0.6, 1.2, and 2.5% groups were significantly higher than control at Weeks 1 and 4, and the activities for the 0.3, 0.6, 1.2, and 2.5% groups were higher than control at Week 13. Lauric acid 11-hydroxylation for the 0.6, 1.2, and 2.5% groups were significantly higher than control at Week 1, and the activities for the 0.3, 0.6, 1.2, and 2.5% groups were higher than control at Weeks 4 and 13. Cytochrome P-450 content for the 1.2 and 2.5% groups was higher than control at Weeks 1 and 4, and the P-450 content for the 0.6, 1.2, and 2.5% groups was higher at Week 13 compared with the control.

Reference: Lake, B.G., Price, R.J., Cunninghame, M.E., and Walters, D.G. (1997). Comparison Of The Effects Of Di(2-Ethylhexyl)Adipate On Heptic Peroxisome Proliferation And Cell Replication In The Rat And Mouse, Toxicology 123: 217-226.

Species: rabbit
Strain: no data
Sex: male
Route of Administration: dermal
Exposure Period: two weeks
Frequency of Treatment: daily, 5 days per week
Post Exposure Observ. Period: NA

Doses: 410, 2060 mg/kg
Control Group: yes, concurrent vehicle

NOEL	<	410	..	mg/kg
LOEL	=	410	..	mg/kg

Method: other
Year: 1962
GLP: no
Testsubstance: no data

Remark: Four male rabbits were treated with 410 or 2060 mg/kg of the neat test substance. A control group received mineral oil. Animals received 10 applications to the shaved skin of the abdomen over a two week period. The area was not occluded, but ingestion was restricted by a plastic Elizabethan collar. Animals were observed daily for signs of toxicity or dermal irritation. Prior to testing and 24 hours after the last administration, blood and urine were collected for analysis. The blood was analyzed for total erythrocyte count, total and differential leukocyte count, and hematocrit. Urine was analyzed for appearance, pH, specific gravity, sugar, acetone, protein, bilirubin, and occult blood. The sediment was evaluated microscopically. The brain, thyroid, lungs, heart, liver, kidneys, adrenals, skin, and bone

	marrow were preserved. The liver and kidneys were examined microscopically.
Result	Three animals in the 410 mg/kg group appeared normal and gained weight. One animal appeared normal during the first week, but had diarrhea and was found dead after the first treatment of the second week. All animals in the 2060 mg/kg group survived. One animal gained weight, while the other three did not gain weight. These animals had labored breathing and were lethargic during the second week. The 410 mg/kg group had slight to moderate erythema and slight desquamation, while the 2060 mg/kg group had moderate erythema and desquamation. No hematologic changes occurred in the 410 mg/kg group, but the 2060 mg/kg group had lower hematocrit values and erythrocyte counts. These animals also had a higher percentage of neutrophils. The differences do not appear to be clinically important, however. No differences in urinalysis were noted. One animal in the 2060 mg/kg group had slightly altered cytology of the liver parenchymal cells (basophilic granulation with enlarged and hyperchromatic nuclei. No other microscopic changes were noted.
Reference	Hazleton Laboratories, Inc. Unpublished report. Repeated Dermal Application - Albino Rabbits, 1962.

5.5 Genetic Toxicity in Vitro

Type	Ames test
System of Testing	TA-98, TA-100, TA-1535, TA-1537, TA-1538
Concentr.	0.15, 0.47, 1.50, 4.74, 15.00, 47.4, 150.0 ug/plate
Metabolic Activation	with and without
Result	negative
Method	OECD Guide-line 471
Year	1982
GLP	no
Testsubstance	no data
Remark	No toxicity was observed in preliminary studies with TA-100. Metabolic activation did not influence the results.
Reference	CMA Unpublished report. Mutagenicity Evaluation of Di(2-ethylhexyl) Adipate (DEHA) in the Ames Salmonella/microsome Plate Test (LBI Project 20988), 1982.

Type	Ames test
System of Testing	TA-98, TA-100, TA-1535, TA-1537
Concentr.	100, 333, 1000, 3333, and 10000 ug/plate
Metabolic Activation	with and without
Result	negative
Method	OECD Guide-line 471
Year	1985
GLP	no
Testsubstance	as prescribed by 1.1 - 1.4
Reference	Zeiger, E., Haworth, S., Mortelmans, K., and

	Speck, W.(1985). Mutagenicity Testing of Di(2-ethylhexyl)phthalate and Related Chemicals in Salmonella, Environ. Mutagen. 7:213-232.
Type	Sister chromatid exchange assay
System of Testing	Chinese hamster ovary
Concentr.	40, 130, 400 ug/ml
Metabolic Activation	with and without
Result	negative
Method	OECD Guide-line 473
Year	1987
GLP	no data
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Negative without activation, equivocal with activation.
Reference	Galloway, S.M., Armstrong, M.J., Reuben, C., Colman, S., Brown, B., Cannon, C., Bloom, A.D., Nakamura, F., Ahmed, M., Duk, S., Rimpo, J., Margolin, B.H., Resnecik, M.A., Anderson, B., and Zeiger, E. (1987). Chromosome Aberrations And Sister Chromatid Exchanges in Chinese Hamster Ovary Cells: Evaluations of 108 Chemicals, Environ. Mol. Mutagen. 10, Suppl. 10:1-175.
Type	Mouse lymphoma assay
System of Testing	mouse lymphoma cell line L5178Y TK+/-
Concentr.	15.6, 31.3, 62.5, 125.0, 250.0, 500.0, 1000.0 nl/ml
Metabolic Activation	with and without
Result	negative
Method	OECD Guide-line 476
Year	1982
GLP	no
Testsubstance	no data
Remark	Concentrations of 62.5, 125, 250, 500, and 1000 nl/ml were used without activation. Substantial reduction in growth (>50%) was observed at the two highest concentrations. With activation, concentrations of 15.6, 31.3, 62.5, 125, 250, or 25, 50, 75, 100, 150, and 200 nl/ml were used. Growth was substantially reduced at 250 nl/ml, but not at other concentrations. No change in mutation frequency occurred at any concentration.
Reference	CMA Unpublished report. Mutagenicity Evaluation of DEHA in the Mouse Lymphoma Forward Mutation Assay (LBI Project 20989), 1982.
Type	Mouse lymphoma assay
System of Testing	mouse lymphoma cell L5178Y tk+/tk-
Concentr.	312.5, 625, 1000, 1250, 1800, 2000, 2500, 2600, 3000, 4000, 4200, 5000 ug/ml
Metabolic Activation	with and without
Result	negative
Method	OECD Guide-line 476
Year	1988
GLP	no data
Testsubstance	as prescribed by 1.1 - 1.4
Reference	McGregor, D.B., Brown, A., Cattanach, P., Edwards, I., McBride, D., Riach, C., and Caspary, W.J. (1988). Responses of the L5178Y tk+/tk- Mouse Lymphoma Cell Forward Mutation Assay:III. 72 Coded Chemicals, Environ. Mol. Mutagen. 12:85-154.

Type	Unscheduled DNA synthesis
System of Testing	primary rat hepatocyte
Concentr.	5, 10, 25, 50, 100, 250, 500, and 1000 nl/ml
Metabolic Activation	no data
Result	negative
Method	OECD Guide-line 482
Year	1982
GLP	no
Testsubstance	no data
Remark	Weak toxicity was observed at a concentration of 1000 nl/ml. Other concentrations had no effect on cell survival. No increase in nuclear labelling was noted.
Reference	CMA Unpublished report. Evaluation of DEHA in the Primary Rat Hepatocyte Unscheduled DNA Synthesis Assay (LBI Project 20991), 1982.

5.6 Genetic Toxicity in Vivo

Type	Micronucleus assay
Species	mouse
Strain	B6C3F1
Sex	male/female
Route of Administration	i.p.
Exposure Period	30 hours after single injection; 24 hours after second injection of 2 daily doses.
Doses	0 and 5 g/kg
Method	other
Year	1982
GLP	no
Testsubstance	no data
Remark	Six male and six female mice were given single injections of the test substance in corn oil, and the bone marrow harvested 30 hours after injection. A second group of animals was treated with 2 injections 24 hours apart and bone marrow harvested 24 hours after the last injection.
Result	All animals survived treatment. Four animals per sex per group were chosen for evaluation of bone marrow. There was no significant difference in the numbers of micronucleated erythrocytes between the treated and negative control groups. Mitotic indices were similar in treated and control groups. Based on these results, the test substance did not induce micronuclei.
Reference	CMA Unpublished report. Mutagenicity Evaluation of DEHA in the Mouse Micronucleus Test (LBI Project 20996), 1982.

Type	Micronucleus assay
Species	mouse
Strain	B6C3F1
Sex	male
Route of Administration	i.p.
Exposure Period	Three consecutive days
Doses	0, 375, 750, 1500, 2000 mg/kg
Method	other
Year	1993

GLP no data
Testsubstance as prescribed by 1.1 - 1.4
Remark Five to seven animals per group were treated with varying doses of test substance for 3 consecutive days and the bone marrow harvested 24 hours after the last treatment.
Result The test was negative at all doses up to 2000 mg/kg.
Reference Shelby, M.D., Erexson, G.L., Hook, G.J., and Tice, R.R.(1993). Evaluation of a Three-Exposure Mouse Bone Marrow Micronucleus Protocol: Results with 49 Chemicals, Environ. Mol. Mutagen. 21:160-179.

Type Dominant lethal assay
Species mouse
Strain ICR
Sex male
Route of Administration i.p.
Exposure Period single injection
Doses 0.5, 1.0, 5.0, 10.0 ml/kg
Method OECD Guide-line 478
Year 1975
GLP no
Testsubstance no data
Result The test substance did not affect late fetal deaths. The 10 ml/kg dose lowered the overall percentage of pregnancies with substantially lower percentages at weeks 1 and 5. There was no effect on the total numbers of implants, but early fetal deaths were more common in the 5 and 10 ml/kg dose groups. There was no time-dose relationship, however. The number of live fetuses was significantly reduced at the 10 mg/kg dose compared with the control group. The effects appear to be postmeiotic rather than premeiotic.
Reference Singh, A.R., Lawrence, W.H., and Autian, J. (1975). Dominant Lethal Mutations and Antifertility Effects of Di-2-Ethylhexyl Adipate and Diethyl Adipate in Male Mice, Toxicol. Appl. Pharmacol. 32:566-576.

5.7 Carcinogenicity

Species rat
Strain Fischer 344
Sex male/female
Route of Administration oral feed
Exposure Period 103 weeks
Frequency of Treatment daily
Post Exposure Observ. Period 1-3 weeks
Doses 1.20, 2.50 %
Control Group yes, concurrent vehicle
Method OECD Guide-line 451
Year 1980
GLP no
Testsubstance as prescribed by 1.1 - 1.4
Result Body weights at the 2.5% dose level were lower than in the control group (data not provided). Treatment did not affect survival. Several neoplasms which are common to this strain were noted in both treated and control animals. No

	compound-related increase in tumor incidence was noted. No hematology was performed at termination
Reference	National Toxicology Program. Carcinogenesis Bioassay of Di(2-ethylhexyl) Adipate, Technical Report No. 212, 1980.

Species	mouse
Strain	B6C3F1
Sex	male/female
Route of Administration	oral feed
Exposure Period	103
Frequency of Treatment	daily
Post Exposure Observ. Period	1-3
Doses	1.20, 2.50 %
Control Group	yes, concurrent vehicle
Method	OECD Guide-line 451
Year	1980
GLP	no
Testsubstance	as prescribed by 1.1 - 1.4
Result	Body weights of high-dose male mice were substantially lower than other groups, and body weights of low- and high-dose female mice were lower than in the control group (no data shown). Survival in the low-dose male group was significantly lower than in the other groups, but was comparable among female groups. The incidence of hepatocellular carcinoma was higher in treated male groups (12% vs 7% in controls) and significantly higher in treated female groups (12-14%) compared with the control group (1%). The incidence of hepatocellular adenomas was also higher in treated groups compared with control groups, but a significant difference was noted only in the high-dose male group. Combined tumor (carcinoma and adenoma) incidences in the treated groups were 41 and 56% in males (vs 26% in controls), and 38 and 37% in females (vs 6% in controls) for low- and high-dose groups, respectively. These results were considered to indicate that the test substance is a hepatocarcinogen in rodents. Hematology was not performed.
Reference	National Toxicology Program. Carcinogenesis Bioassay of Di(2-ethylhexyl) Adipate, Technical Report No. 212, 1980.

Species	rat
Strain	no data
Sex	no data
Route of Administration	oral feed
Exposure Period	two years
Frequency of Treatment	daily
Post Exposure Observ. Period	
Doses	0.1, 0.5, 2.5%
Control Group	yes, concurrent vehicle
Method	other
Year	1966
GLP	no
Testsubstance	no data
Remark	Rats (strain, number, and sex not specified) were fed the test substance in the diet for two years.

	Tissues were examined for the presence of tumors. No hematology was performed. Body weights and feed consumption were not reported. The tissues preserved and examined were not specified.
Result	Of the total of 33 tumors found, most were lymphomata and adenomata. No compound-related increase in tumor incidence was noted.
Reference	Hodge, H.C., Maynard, E.A., Downs, W.L., Ashton, J.K., and Salerno, L.L. (1966). Tests on Mice for Evaluating Carcinogenicity, Toxicol. Appl. Pharmacol. 9:583-596.

Species	mouse
Strain	C3H
Sex	male/female
Route of Administration	dermal
Exposure Period	18 months
Frequency of Treatment	weekly
Post Exposure Observ. Period	
Doses	0.1, 10 mg/animal
Control Group	yes, concurrent vehicle
Method	other
Year	1966
GLP	no
Testsubstance	no data
Remark	Fifty male and female mice per group were given weekly treatments of test substance in 0.2 ml acetone. Animals were treated until death occurred.
Result	There was no evidence of tumor formation on the skin of treated mice.
Reference	Hodge, H.C., Maynard, E.A., Downs, W.L., Ashton, J.K., and Salerno, L.L. (1966). Tests on Mice for Evaluating Carcinogencity, Toxicol. Appl. Pharmacol. 9:583-596.

5.8 Toxicity to Reproduction

Type	One generation study
Species	rat
Strain	other
Sex	male/female
Route of Administration	oral feed
Exposure Period	Premating exposure only
Frequency of Treatment	daily
Premating Exposure Period	
male	10 weeks
female	10 weeks
Duration of Test	10 weeks premating plus 36 days post partum
Doses	0.03, 0.18, 1.20 %
Control Group	yes, concurrent vehicle
NOEL Parental	= 0.18 .. %
NOEL F1 Offspring	= 0.18 .. %
NOEL F2 Offspring	..
Method	OECD Guide-line 415
Year	1988
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	Dosing terminated prior to mating. Litters were not culled to 4 male and 4 female pups as

	specified in the Guideline. Strain used was Alpk:APfSD (Wistar derived).
Result	No clinical signs of toxicity were noted in males or females. No changes in body weight or feed consumption occurred during the premating exposure period, but the high-dose dams had reduced body weight gain during gestation compared with the control group. No treatment-related effects on fertility (male or female) were noted. Mean pup weight gain, total litter weight, and litter size in the high-dose group were reduced throughout the post-partum period. No other treatment groups were affected. The percentage of live-birth litters and their survival to Day 22 post-partum were comparable among groups. At necropsy, liver weights (absolute and relative to body weight) were increased in high-dose males and females, but not in other groups. No treatment-related microscopic changes occurred in the reproductive tissues of the P generation. No treatment-related gross pathologic changes occurred in the F1 generation.
Reference	CEFIC Unpublished report. Di-(2-ethylhexyl) Adipate (DEHA)Fertility Study in Rats (CTL Study RR0374), 1988.

5.9 Developmental Toxicity/Teratogenicity

Species	rat
Strain	Sprague-Dawley
Sex	female
Route of Administration	i.p.
Exposure Period	during gestation
Frequency of Treatment	Day 5, 10, and 15 of gestation
Duration of Test	Day 20 of gestation
Doses	0.9, 4.6, and 9.2 mg/kg
Control Group	yes, concurrent vehicle

NOEL Maternal Toxicity	=	4.6	mg/kg bw/day
NOEL Teratogenicity	=	9.2	mg/kg bw/day

Method	other
Year	1973
GLP	no
Testsubstance	no data
Remark	Five female rats per group were treated by ip injection on Days 5, 10, and 15 of gestation at the doses specified. The test substance was administered neat. Dams were sacrificed on Day 20, and the uterine horns removed. The numbers of corpora lutea, resorption sites, viable and dead fetuses were counted. Individual fetal weights were measured, and all fetuses (dead and viable) were examined for gross anomalies. Half the fetuses were examined for skeletal anomalies and the other half for visceral anomalies.
Result	Maternal body weights were significantly lower in the mid- and high-dose groups. No increases in early or late fetal deaths were noted. Mean fetal weights in the mid- and high-dose groups were significantly lower than in the control group. There was no teratogenicity or increase in skeletal anomalies.

Reference	Singh, A.R., Lawrence, W.H., and Autian, J. (1973). Emrbyonic-Fetal Toxicity and Teratogenic Effects of Adipic Acid Esters in Rats, J. Pharm. Sci. 10:1596-1600.
Species	rat
Strain	other
Sex	female
Route of Administration	oral feed
Exposure Period	Days 1-22 of gestation
Frequency of Treatment	daily
Duration of Test	Day 22 of gestation
Doses	.03, 0.18, 1.2 %
Control Group	yes, concurrent vehicle
NOEL Maternal Toxicity	= 0.18 .. %
NOEL Teratogenicity	= 1.2 .. %
Method	Chernoff-Kavlok teratogenicity screening test
Year	1988
GLP	yes
Testsubstance	as prescribed by 1.1 - 1.4
Remark	The mid-dose was considered to be fetotoxic with the low-dose clearly a no effect level. Strain used was Alpk:APfSD (Wistar derived).
Result	Maternal body weights and feed consumption in the high-dose group were significantly lower than in the control group. A slight decrease in litter size occurred in the high-dose group, but this was not statistically significant. There was no effect on fetal weight. No major abnormalities or minor visceral were noted. Mid- and high-dose groups had slightly higher incidences of minor skeletal abnormalities which were attributed to fetotoxicity.
Reference	CEFIC Unpublished report. Di(2-ethylhexyl) Adipate: Teratogenicity Study in the Rat (CTL Study RR0372), 1988.

5.10 Other Relevant Information

Type	Chemobiokinetics general studies
Remark	Male Sprague-Dawley rats received both radiolabelled materials in either corn oil or DMSO by oral gavage. Blood, bile, and urine were collected for 450 min and the amount of radioactivity determined. Male Sprague-Dawley rats and male NMRI mice were treated with both radiolabelled materials by oral gavage and an animal sacrificed 20 min, 1, 4, 24 hrs, or 4 days after treated. These animals were imbedded, frozen, and sagital sections prepared for whole-body autoradiography.
Remark	In rats, absorption of DEHA from corn oil was 2-4 times lower than from DMSO, and total radioactivity was greater when the carbonyl-14C labelled test substance was used. Peak absorption from the DMSO preparation occurred within 2 hours. Absorption from corn oil continued to rise during the 450 min sampling period. Biliary excretion of [carbonyl-14C]DEHA was less than 1% of the dose, whereas 10-41% of the [2-ethylhexyl-1-14C]DEHA was found in the bile. The DMSO vehicle enhanced the biliary excretion

(41% of dose vs 10% for corn oil). Enterohepatic cycling was indicated, although the data were not shown. Urinary excretion was >= 1% of the dose of [carbonyl-14C]DEHA, and was 2.9 and 6.6% for [2-ethylhexyl-1-14C]DEHA during the 450 min sampling period. Autoradiography of a mouse and rat 24 hr after oral gavage of [carbonyl-14C]DEHA indicated distribution in the liver and kidneys of mice, and bone marrow, brown fat, adrenal cortex, salivary glands, and forestomach of rats. Traces of radioactivity were observed in mice 4 days after dosing. After oral dosing of [2-ethylhexyl-1-14C]DEHA, radioactivity was observed in the liver, kidneys, and intestine of rats, and in the liver of mice.

Test substance [Carbonyl-14C]DEHA (radiochemical purity 99%) and [2-ethylhexyl-1-14C]DEHA (radiochemical purity 97%) were synthesized and used for this study.

Reference Bergman, K., and Albanus, L. (1987). Di-(2-ethylhexyl) Adipate: Absorption, Autoradiographic Distribution and Elimination in Mice and Rats, Fd. Chem. Toxic. 25:309-316.

Type Chemobiokinetics general studies

Remark Male and female B6C3F1 mice, F-344 rats, and Cynomolgus monkeys were treated by oral gavage with [hexyl-2-14C]DEHA. Doses of 50, 500, and 5000 mg/kg were used. Elimination of radioactivity in urine, feces and expired air was determined during the 24-48 hours posttreatment. The extent of DEHA hydrolysis and absorption were also determined in mice at 1, 3, and 6 hours postdosing. Excreta and liver samples were analyzed to determine the identity of metabolites.

Remark Disposition studies in mice indicated that 95-102% of the administered radioactivity from the low- and mid-dose groups was eliminated in the urine, feces, and expired air within 24 hours postdosing. Approximately 90% was excreted in the urine and 7-8% in the feces. At the high-dose, ~12% was recovered in the GI tract, 75% excreted in the urine, and 4% excreted in the feces. Tissue localization was dose-dependent with the adrenals and liver of low- and mid-dose groups containing detectable radioactivity. The blood, liver, skin, and fat of high-dose animals had detectable radioactivity. Rats and monkeys also eliminated the majority of radioactivity in the urine, but had a higher fecal elimination than did mice. Urinary metabolites consisted of 2-ethylhexanoic acid (EHA), its glucuronide conjugate, 5-hydroxy-EHA, and the diacid diEHA. Rats excreted less glucuronide conjugated metabolite than did mice or monkeys. Monkeys excreted more MEHA, EHA, and glucuronide conjugated metabolites.

Absorption studies in mice indicated rapid absorption after dosing with peak levels reached at 1 and 3 hours. The contents of the GI tract contained DEHA, MEHA, and 2-ethylhexanol.

Test substance [Hexyl-2-14C]DEHA was synthesized and determined to be 98% radiochemically pure.

Reference CMA Unpublished report. Metabolism and Disposition of Di-2-ethylhexyl Adipate (MRI Project 7550-B), 1984.

Type Chemobiokinetics general studies

Remark Male Wistar rats were treated by oral gavage with a single dose of 500 mg/kg of DEHA in DMSO. Urine, feces, and expired air were collected for 2 days post dosing. Other animals were given the same dose and groups of 3 sacrificed after 6, 12, 24, 48, and 96 hours to determine tissue distribution. Five rats were also given a single oral dose of 100 mg of unlabelled DEHA in DMSO. The appearance of metabolites MEHA and adipic acid (AA) in the urine, blood, stomach, intestine, and liver were determined at 1, 3, and 6 hours postdosing. The in vitro hydrolysis of DEHA by liver homogenates was also investigated.

Remark Radioactivity was eliminated primarily in the urine and expired air with most of the elimination between 10-24 hours after dosing. Tissue distribution consisted of liver, kidneys, blood, muscle, and fat as well as stomach and GI tract. Quantities of radioactivity in blood-rich tissues decreased by 50% between 12 and 24 hours postdosing. Total radioactivity was 10% in tissues at 24 hours, and declined to 0.5% by 96 hours postdosing. The major metabolite appeared to be AA which appeared in the urine and intestine by 3 hours. AA was also the major hydrolysis product from the liver and intestine, but the pancreas hydolyzed DEHA primarily to MEHA.

Test substance [Carbonyl-14C]DEHA was synthesized and estimated to be 99% pure by thin layer chromatography.

Reference Takahashi, T., Tanaka, A., and Yamaha, T. (1981). Elimination, Distribution and Metabolism of Di(2-ethylhexyl)Adipate (DEHA) in Rats, Toxicol. 22:223-233.

Type Metabolism

Remark Male Wistar rats were given oral doses of 665 or 1500 mg/kg DEHA in corn oil for 5 days. Urine was collected each morning and analyzed for metabolites. In other studies, hepatocytes were incubated with 0.1, 0.5, or 1.0 mM DEHA for 24 hours and the metabolites determined.

Remark No DEHA was recovered in the urine after 24 hours. On Day 1, adipic acid (AA) was the main metabolite regardless of the dose level. Ethylhexanoic acid (EHA)-glucuronide and the diacid (diEHA) were also found. By Day 3, the amount of AA was slightly lower than on Day 1 and more EHA-glucuronide was found particularly in the high-dose group. The total amount of radioactivity excreted into the urine remained the same, however.

In vitro metabolism also indicated AA as the major metabolite with EHA and EHA-glucuronide as the next component.

Test substance [1-6 14C]DEHA was supplied by ICI. No analysis given.

Reference Cornu, M.C., Keith, Y., Elcombe, C.R., and Lhugeunot, J.C.(1988). In Vivo and In Vitro Metabolism of Di-(2-ethylhexyl) Adipate a Peroxisome Proliferator, in the Rat, Arch. Toxicol., Suppl. 12:265-268.

Type Chemobiokinetics general studies

Remark Six male human volunteers were treated with 46 mg of 2H-DEHA through an indwelling iv catheter. Blood samples were taken prior to dosing, and 0.5, 1, 2, 3, 4, 5, 6, 8, and 12 hours after dosing. Additional samples were taken after 24 and 31 hours. Urine was collected over 4-hour intervals for the first 12 hours, 12-hour intervals until 48 hours, and 24-hour intervals until 96 hours post dosing.

Remark Unconjugated EHA was detected in the plasma, but there was no evidence of absorption of the parent DEHA. EHA could not be detected after 31 hours. In the urine, a conjugated form of EHA was detected as the primary metabolite accounting for 99% of the total deuterium measured, and about 12% of the administered dose. The feces contained only a small amount of the administered dose.

Test substance Deuterated di(2-ethylhexyl) adipate (purity > 99%)

Reference Loftus, N.J., Laird, W.J.D., Steel, G.T., Wilks, M.F., and Woollen, B.H. (1993). Metabolism and Pharmacokinetics of Deuterium-Labelled Di-2-(ethylhexyl) Adipate (DEHA) in Humans, Fd. Chem. Toxic. 31: 609-614.

Type Exposure assessment

Remark In the first phase, six human volunteers (four male and two female) were provided a cheese sandwich wrapped in DEHA plasticized cling wrap. The amount of DEHA contained in each sandwich was determined to be 5.2 mg. Urine samples were collected over a 24 hour period prior to exposure and 24 hours after exposure to determine the level of EHA excreted.

In the second phase, urine from 112 human volunteers (65 men; 9 boys; 36 women; and 2 girls) from 5 locations in the UK were collected to measure the amount of EHA and extrapolate to the exposure of DEHA.

Remark The data from phase 1 indicated that urine from a 24 hour collection would be required to obtain sufficient information on DEHA exposure. The data from phase 2 indicated that exposure to DEHA varied from < 1 mg/day to ca 11 mg/day with the median value of 2.7 mg/person/day obtained for this population.

Test substance Di(2-ethylhexyl) adipate in cling wrap

Reference Loftus, N.J., Woollen, B.H., Steel, G.T., Wilks, M.F., and Castle, L.(1994). An Assessment of the Dietary Uptake of Di-2-(ethylhexyl) Adipate (DEHA) in a Limited Population Study, Fd. Chem. Toxicol. 32: 1-5.

Type Estrogenicity, *in vitro*

Remark Estrogen receptors isolated from the cytosol of liver from rainbow trout was used for receptor binding assays. Receptors were incubated with ^{3}H-17β-estradiol and concentrations of up to 1 mM DEHA. The bound and unbound fraction of estradiol was determined.

Estrogen activation was evaluated using 2 human breast cell lines (ZR-75 and MCF-7). Proliferation of ZR-75 cells cultured with 10^{-5} M DEHA was determined after 10 days and compared to the activation by 17β-estradiol. Likewise, the ability of 10^{-5} M DEHA to activate transcription of MCF-7 cells was evaluated using a pTKLUC reporter gene.

Remark DEHA bound to the fish liver estrogen receptor at concentrations of 10^{-7} to 5 x 10^{-5} M (0-40% binding). Affinity constants could not determined because of low water solubility.

DEHA did not activate the estrogen receptor in the ZR-75 of MCF-7 assay systems.

Test substance Di(2-ethylhexyl) adipate

Reference Jobling, S., Reynolds, T., White, R., Parker, M.G., and Sumpter, J.P. (1995). A Variety of Environmentally Persistent Chemicals, Including Some Phthalate Plasticizers, are Weakly Estrogenic, *Environ. Health Perspect.* 103:582-587.

Type Estrogenicity, *in vivo*

Remark Ten female Sprague-Dawley rats per group were ovariectomized at 7 weeks of age. Animals were treated orally by intubation with 30 μg/kg/day ethynylestradiol, 30 mg/kg/day genistein, or 1000 mg/kg/day DEHA for 3 days. Body weight and uterine wet weight was measured at necropsy 24 hours after the last exposure. Vaginal smears were prepared on the day of necropsy for evaluation of estrous.

Remark Animals treated with ethynylestradiol showed signs of estrous (8/10 animals), while all other animals were in diestrous. The uterine weights for animals treated with ethynylestradiol and genistein were significantly increased above controls. No increase in uterine weight was observed following treatment with DEHA.

Test substance Di(2-ethylhexyl) adipate (> 98% purity)

Reference Japan Plasticizer Industry Association (1998). Evaluation Of Adipic Acid Esters On Estrogenicity by *in vivo* Uterotrophy in Ovariectomized Rats, Mitsubushi Chemical Safety Institute Ltd., unpublished report 8L306.