FOREWORD INTRODUCTION

DIISOPROPYLBENZENE

CAS N°: 25321-09-9

SIDS Initial Assessment Report

For SIAM 15

Boston, USA, 22-25 October, 2002

1. Chemical Name: Diisopropylbenzene

2. CAS Number: 25321-09-9

3. Sponsor Country: Japan
National SIDS Contact Point in Sponsor Country:
Ms. Mizuho Hayakawa, Ministry of Foreign Affairs, Japan

4. Shared Partnership with:

5. Roles/Responsibilities of the Partners:

- Name of industry sponsor /consortium
- Process used

6. Sponsorship History

- 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: August 13, 2002

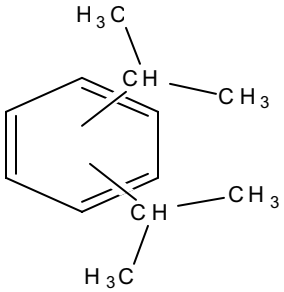
10. Date of last Update: September 2003

11. Comments: The original draft documents were prepared by the Japanese government. Tests:

No testing ( )
Testing (x) log Pow, Water solubility, Hydrolysis, Biodegradation and Bioconcentration, Acute toxicity to fish, daphnia and algae, Chronic toxicity to daphnia, Repeated dose toxicity, Reproduction/developmental toxicity, Ames test and Chromosomal aberration test

Literature search was performed using the Toxline and Medline, and review articles were looked for in IUCLID, RTECS, IRIS, IARC, EHC, and Toxicological Profile.

SIDS INITIAL ASSESSMENT PROFILE

CAS No.	25321-09-9
Chemical Name	Diisopropylbenzene
Structural Formula	

SUMMARY CONCLUSIONS OF THE SIAR

The mixture of *m*- and *p*-diisopropylbenzene (60:40/*m*-:*p*-) was assessed.

Human Health

There is no data available regarding toxicokinetics and metabolism.

No data are available for the acute toxicity of diisopropylbenzene with the composition of 60:40/*m*-:*p*-. The oral LD_{50} value of each constitutive isomer, *m*-diisopropylbenzene and *p*-diisopropylbenzene, was shown to be greater than 3,000 mg/kg b.w. in rodents. Acute toxicity studies of diisopropylbenzene with an unknown isomer ratio are available. The oral and dermal acute toxicity of diisopropylbenzene are negligible: the oral LD_{50} in rats is 5,850 mg/kg b.w., and the dermal LD_{50} in rabbits is 14,400 mg/kg b.w. In an acute inhalation study, no deaths occurred at doses below 5,300 mg/m^3 after 4h exposure in rats and after 2h exposure in mice. The weight of evidence shows that the acute toxicity of diisopropylbenzene with the composition of 60:40/*m*-:*p*- can be considered to be low.

There is no reliable information on eye and skin irritation and sensitization.

In accordance with the Japanese guideline, equivalent to OECD TG 407, SD rats received diisopropylbenzene with the composition of 60:40/*m*-:*p*- by gavage at doses of 0, 6, 30, 150 and 750 mg/kg b.w./day for 28 days. Mydriasis was observed in 2 of 6 males and 2 of 6 females at 150 mg/kg b.w./day and in 10 of 12 males and all of 12 females at 750 mg/kg b.w./day. This sign was observed from about 2 to 6 hours after administration mainly during the latter half of the dosing period. On histopathological examination, centrilobular hypertrophy of hepatocytes was observed in 1 of 6 males in the 150 mg/kg b.w./day group and 1 of 6 males and 4 of 6 females in the 750 mg/kg b.w./day group. Based on mydriasis and histopathological changes in the liver at 150 mg/kg b.w./day, the NOAEL of this chemical was considered to be 30 mg/kg b.w./day.

In a reverse gene mutation assay [OECD TG 471, 472], diisopropylbenzene with the composition of 60:40/*m*-:*p*- was not mutagenic in *Salmonella typhimurium* TA100, TA1535,

TA98, TA1537 and *Escherichia coli* WP2 *urv*A with and without an exogenous metabolic activation. In a chromosomal aberration test [OECD TG 473] with diisopropylbenzene with the composition of 60:40/*m*-:*p*-, structural chromosomal aberrations and polyploidy were not induced with and without an exogenous metabolic activation in cultured Chinese hamster lung (CHL/IU) cells.

There are no data available on carcinogenicity.

In a reproductive/developmental toxicity screening test [OECD TG 421], diisopropylbenzene with the composition of 60:40/*m*-:*p*- was administered to SD rats by gavage at doses of 0, 6, 30, 150 and 750 mg/kg b.w./day from day 14 before mating to day 14 after mating in males and to day 3 of lactation in females. This treatment produced no effect on reproductive performance and no adverse effect on offspring. In view of these findings the NOAEL for reproductive/developmental toxicity is considered to be 750 mg/kg b.w./day.

Environment

The melting points of *m*- and *p*- diisopropylbenzene are –63 and –17.1 degree C respectively. The boiling point of *m*- and *p*- diisopropylbenzene is 203 degree C. The vapour pressures of *m*- and *p*- diisopropylbenzene are 0.524 hPa and 0.328 hPa respectively at 25 degree C. Water solubility is (m-) 72.0 ug/L, (p-) 40.5ug/L at 25 degree C. Henry's law constants of *m*- and *p*- diisopropylbenzene are 1.17 and 1.30 atm m^3/moL, respectively. The partition coefficients of *m*- and *p*- diisopropylbenzene are 5.13 and 5.23 respectively.

The fugacity model (Mackay level III) suggests that if diisopropylbenzene is released to air or soil, it is unlikely to distribute into other compartments and that if it is released to water it has a tendency to go into the sediment compartment.

m- and *p*-Diisopropylbenzene are not inherently biodegradable and both of their bioconcentration potentials seems to be high (BCF= 503 – 3210, mixture). In the air, *m*- and *p*- diisopropylbenzene are expected to be photodegraded (*m*-: $T_{1/2}$=8.3 hours, *p*-: $T_{1/2}$=13 hours) by OH radicals. Hydrolysis is not expected to occur.

The acute toxicity of diisoprophylbenzene has been tested in three aquatic species belonging to three trophic levels. For algae (OECD TG 201, *Selenastrum capricornutum,* closed system) a 72hEbC50 of 1.6 mg/L and a 72hErC50 of 2.7 mg/L were determined. For daphnids (OECD TG 202 part 1, *Daphnia magna,* semistatic) a 48hEC50 of 0.39 mg/L, and for fish (OECD TG 203, *Oryzias latipes*) a LC50 of 0.71 mg/L were reported.

Regarding chronic toxicity, for algae (OECD TG 201 growth inhibition, *Selenastrum capricornutum*) a 72 h NOErC of 0.69 mg/L, a 72 h NOEbC of 0.31 mg/L, and for daphnids (TG 211 reproduction, *Daphnia magna*) a 21 d NOEC of 0.063 mg/L were reported.

Most of the toxicity results were above the water solubility limit of the substance (*m*-; 0.072 mg/L and *p*- ; 0.0405 mg/L). As the NOEC of the chronic toxicity study with daphnids is close to the solubility limit of the substance, it can be considered that this result reflects the actual toxicity of the substance.

Exposure

Diisopropylbenzenes are produced as by-products of cumene synthesis in closed systems. The production volume was ca. 30,000 tonnes/year by two companies in Japan. These chemicals are blended into gasoline, diesel and other hydrocarbon fuels, and also used for diisopropylbenzeneperoxide synthesis.

Diisopropylbenzenes are volatile liquids and consumer and worker exposure through inhalation and dermal contact is possible.

RECOMMENDATION

Human Health: The chemical is currently of low priority for further work based on a low hazard potential.

Environment: The chemical is a candidate for further work.

RATIONALE FOR THE RECOMMENDATION AND NATURE OF FURTHER WORK RECOMMENDED

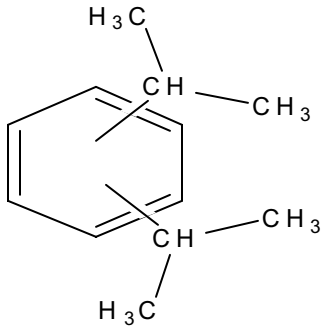
Diisopropylbenzene is not inherently biodegradable and is expected to have a high bioaccumulation potential and aquatic toxicity. An environmental exposure assessment and if necessary a risk assessment is recommended.

SIDS Initial Assessment Report

1 IDENTITY

1.1 Identification of the Substance

CAS Number: 25321-09-9
Name: Diisopropylbenzene
Molecular Formula: C12H18
Structural Formula:



Molecular Weight:
Synonyms: Benzene, bis(1-methylethyl)-
Diisopropylbenzene, all isomers
Diisopropylbenzene (mixture)

1.2 Purity/Impurities/Additives

Substance type: organic

Physical status: liquid

Purity: mixture of m- and p-configuration (about 60 % w/w of m - diisopropylbenzene, about 40 % w/w of p - diisopropylbenzene)

1.3 Physico-Chemical properties

Both of m - and p - diisopropylbenzene are colorless liquids, which are insoluble in water (m - : 72.0 µg/L at 25 °C, p - : 40.5 µg/L at 25 °C). Other physical-chemical properties are shown in Table 1. There are no big differences between the m- and p- isomer regarding the physical-chemical properties.

Table 1 Summary of physico-chemical properties

	Protocol	Results (m -)	Results (p -)
Melting Point	Unknown	- 63 °C	- 17.1 °C
Boiling Point	Unknown	203 °C	203 °C
Density	Unknown	0.8549 g/cm^3 at 25 °C (mixture)	
Vapor Pressure	Unknown	52.4 Pa at 25 °C	32.8 Pa at 25 °C
Partition Coefficient (Log Pow)	OECD TG 107	5.13	5.23
Water Solubility	OECD TG 105	72.0 µg/L at 25°C	40.5 µg/L at 25°C
Henry's Law constant	Unknown	1.17 atm m^3/mol	1.30 atm m^3/mol

2 GENERAL INFORMATION ON EXPOSURE

2.1 Production Volumes and Use Pattern

The production volume was ca.30,000 tons/year in Japan. This chemical (Cas No.25321-09-9) is not a high production chemical in the EU countries.

Packing operation is carefully conducted in order to avoid leaking and overflow. After the packing operation, diisopropylbenzene which is not packed is transferred to a tank lorry by nitrogen pressure. In the case of leaking, a bucket will be used to receive diisopropylbenzene which leaked. Waste of diisopropylbenzene is incinerated in a factory incinerator; it is not treated in an activated sludge treatment plant.

Diisopropylbenzene is blended into gasoline, diesel and other hydrocarbon fuels. It is also used for the synthesis of diisopropylbenzeneperoxide.

2.2 Environmental Exposure and Fate

Diisopropylbenzene is not inherently biodegradable (OECD TG302C: 2 % by BOD after 14 days (Data of Existing Chemicals based on the CSCL Japan, 1992)). The bioconcentration potential in carp is moderately high (OECD TG305: 503 - 1680 for m - diisopropylbenzene at a test concentration of 20 µg/L and 546 - 3210 at a test concentration of 2 µg/L for 56 days: 530 - 2300 for p - diisopropylbenzene at a test concentration of 20 µg/L and 512 - 2960 at a test concentration of 2 µg/L for 56 days (Data of Existing Chemicals based on the CSCL Japan, 1992). Hydrolysis is not expected to occur (OECD TG117: stable at pH 4, 7 and 9 at 50 °C for five days). In the air, this chemical is expected to be photodegradated (half-life time is 8.3 hours for m-diisopropylbenzene and 13 hours for p-diisopropylbenzene) by OH radicals. The estimated rate constants are 1.55×10^{-11} cm^3 $molecule^{-1}$ sec^{-1} and 1.01×10^{-11} cm^3 $molecule^{-1}$ sec^{-1} respectively.

The potential environmental distribution of diisopropylbenzene obtained from a generic level III fugacity model under three emission scenarios is shown in Table 2.2-1 and 2.2-2. The results show that if diisopropylbenzene is released to one of the compartments of air and soil, it is unlikely to distribute into other compartments and that if it is released to water it has a tendency to go into the sediment compartment.

As shown in Table 2.2-1 and Table 2.2-2, the tendency for enviromental distribution of the m- and p- isomers is almost the same.

Table 2.2-1: Environmental distribution of m -diisopropylbenzene using a generic level III fugacity model under three emission scenarios

Compartment	Release: 100 % to air	Release: 100 % to water	Release: 100 % to soil
Air	99.1 %	1.0 %	1.9 %
Water	0.0 %	22.6 %	0.0 %
Soil	0.9 %	0.0 %	98.1 %
Sediment	0.0 %	76.4 %	0.0 %

Data used

Melting point: - 63 °C, Vapour pressure: 52.4 Pa, Water solbility: 0.072 mg/L, LogPow: 5.13, Half-life time in air: 8.3 hours, Half-life times in water, soil and sediment are assumed 240,000, 240,000 and 720,000 hours, because this chemical is not inherently biodegradable.

Table 2.2-2: Environmental distribution of p -diisopropylbenzene using a generic level III fugacity model under three emission scenarios

Compartment	Release: 100 % to air	Release: 100 % to water	Release: 100 % to soil
Air	99.0 %	1.2 %	2.4 %
Water	0.0 %	19.6 %	0.0 %
Soil	1.0 %	0.0 %	97.6 %
Sediment	0.0 %	79.2 %	0.0 %

Data used

Melting point: - 17.1 °C, Vapour pressure: 32.8 Pa, Water solbility: 0.0405 mg/L, LogPow: 5.23, Half-life time in air: 12.7 hours, Half-life times in water, soil and sediment are assumed 240,000, 240,000 and 720,000 hours, because this chemical is not inherently biodegradable.

2.2.1 Occupational Exposure

Diisopropylbenzene is a volatile liquid and exposure through inhalation and dermal contact is possible. In Japan, m- and p-diisopropylbenzene are produced as byproducts of cumene synthesis.

Diisopropylbenzene is formulated by mixing these isomers in a tank. Workers perform quality control sampling from the tank twice a year, drum filling 1-2 times/month, and lorry filling 3 times/month. The duration of these operations are 1, 10, and 3 minutes, respectively. The quality control sampling and lorry filling are outdoor operation and the drumming machine has a local exhaust ventilation system. The exposure concentrations estimated with the EASE model for sampling and lorry filling was 10 -50 ppm, assuming non-dispersive direct handling, 0.5-3 ppm for drum filling, assuming non-dispersive handling with local exhaust ventilation. The workers wear protective clothing, glasses, boots, and gloves during these operations to prevent dermal exposure. The yearly average EHEinh for the worker who did all these jobs was 0.032 mg/kg/day, and the EHEinh of the working day for drumming worker, the highest exposure, was 0.06 mg/kg/day. The workers did not use respirators during these works.

In Japan, no occupational exposure standard has been assigned to this chemical.

3 HUMAN HEALTH HAZARDS

The mixture of m- and p-diisopropylbenzene (60:40/m-:p-) was assessed.

3.1 Effects on Human Health

3.1.1 Toxicokinetics, Metabolism and Distribution

Toxicokinetics and metabolism- There is no data available.

3.1.2 Acute Toxicity

Studies in Animals

No data are available for the acute toxicity of diisopropylbenzene with the composition of 60:40/m-:p-. Acute toxicity data on the constitutive isomer, m- and p-diisopropylbenzene, and diisopropylbenzene with unknown isomer ratio via the oral, inhalation and dermal route using rats, mice and rabbits are shown in Table 3.1.2. However, none of them were conducted according to the current guidelines and no detailed information, including the ratio of the constitutive isomers and the observed effects is available. As for the constitutive isomers (see section h. Information on related chemicals), the oral LD50 value of the meta-isomer was reported to be 7,400 mg/kg b.w. in rats and 3,100 mg/kg b.w. in mice, and that of the para-isomer 3,400 mg/kg b.w. in mice [Izmerov et al.: 1982]. The oral and dermal LD50 of a mixture with an unknown isomer ratio was reported to be 5,850 mg/kg b.w. in rats and 14,400 mg/kg b.w. in rabbits, respectively. In an acute inhalation study, no deaths occurred at doses below 5,300 mg/m^3 after 4h exposure in rats and after 2h exposure in mice.

Table 3.1.2: Acute toxicity of diisopropylbenzene

Route	Sample	Animal	Type	Values	References
Oral	Isomer (m-Configuration)	Rat	LD50	7,400 mg/kg	Izmerov et al. (1982)
Oral	Isomer (m-Configuration)	Mouse	LD50	3,100 mg/kg	Izmerov et al. (1982)
Oral	Isomer (p-Configuration)	Mouse	LD50	3,400 mg/kg	Izmerov et al. (1982)
Oral	Mixture*	Rat	LCLO	6.5 ml/kg b.w. (5,850 mg/kg b.w.)	Charles et al. (1974)
Inhalation	Mixture*	Rat	LCLO	5,300 mg/m3/4h	Izmerov et al. (1982)
Inhalation	Mixture*	Mouse	LCLO	5,300 .mg/m3/2h	Izmerov et al. (1982)
Dermal	Mixture*	Rabbit	LD50	16 .ml/kg b.w. (14,400 mg/kg b.w.)	Charles et al. (1974)

LCLO: lethal concentration, low

*: mixture with unknown isomer ratio.

The weight of evidence shows that the acute toxicity of diisopropylbenzene with the composition of 60:40/m-:p- can be considered to be low.

Conclusion

The oral LD50 value of each constitutive isomer, m- and p-diisopropylbenzene, was shown to be greater than 3,000 mg/kg b.w. in rodents. Acute toxicity studies with diisopropylbenzene of unknown isomer ratio are available. The oral and dermal acute toxicity of diisopropylbenzene are negligible: oral LD50 in rats 5,850 mg/kg b.w., and dermal LD50 in rabbits 14,400 mg/kg b.w. In an acute inhalation study, no deaths occurred at doses below 5,300 mg/m^3 after 4h exposure in rats and after 2h exposure in mice. The reliability of these studies was uncertain. However, the weight of evidence shows that the acute toxicity of diisopropylbenzene with the composition of 60:40/m-:p- can be considered to be low.

Studies in Humans

There is no available data on humans.

3.1.3 Irritation

Studies in Animals

Data are available regarding irritation of diisopropylbenzene with unknown composition.
The substance is reported to be moderately irritating to the skin at 100 mg/24hr and mildly to eyes at 500 mg/24hr in rabbits but the reliability of the study is uncertain because no further details are available [Marhold: 1986].

3.1.4 Sensitisation

There is no available information on sensitization.

3.1.5 Repeated Dose Toxicity

Studies in Animals

Oral

Two oral studies were available for repeated dose toxicity. One is 28-day repeated dose toxicity test [MHW, Japan: 1998], which was identified as a key study because this study was well conducted according to a Japanese test guideline equivalent to OECD test guideline 407 under GLP. The other is an OECD reproduction/developmental toxicity screening test [TG 421] [MHLW, Japan: 2001]. This study was considered insufficient as a repeated dose toxicity study because of the lack of some examinations including hematological and blood biochemical examination and histopathological examination on liver and kidneys. Details of 28-day study are as follows.

Crj: CD (SD) rats received diisopropylbenzene (meta: 57.0 %, para: 41.0 %) by gavage at 0 (vehicle: corn oil), 6, 30, 150 and 750 mg/kg b.w./day for 28 days [MHW, Japan: 1998]. Twelve rats per sex per dose (6 for scheduled sacrifice and 6 for recovery maintenance) were used. Mydriasis was observed from day 14 of administration in 2 of 6 females at 150 mg/kg b.w./day and from day 8 of administration in 2 of 6 males at 150 mg/kg b.w./day and in 10 of 12 males and all of 12 females in the 750 mg/kg b.w./day group. This sign appeared from about 2 and half hours to 3 and half hours after administration and disappeared within 2 and half hours after the onset. At necropsy, increases in absolute and relative liver weight were observed in both sexes and increases in absolute and relative kidney weight were observed in males of the 750 mg/kg b.w./day group. On histopathological examination, centrilobular hypertrophy of hepatocytes was observed in 1 of 6 males in the 150 mg/kg b.w./day group and 1 of 6 males and 4 of 6 females in the 750 mg/kg

b.w./day group. This change was not observed in recovery animals. Additionally, eosinophilic bodies of the proximal tubular epithelium were observed in 1 of 6 males each in the 6, 30 and 150 mg/kg b.w./day groups and all of 6 males in the 750 mg/kg b.w./day group. This kidney change was considered due to accumulation of the complex with alpha-2u globulin, a male rat specific protein, and not considered relevant to humans. Based on mydriasis and hepatic change, the NOAEL for repeated dose toxicity was concluded to be 30 mg/kg b.w./day for both sexes.

Conclusion

In an oral 28-day study using rats, mydriasis and histopathological changes in the liver were observed. The NOAEL for repeated dose toxicity was considered to be 30 mg/kg b.w./day.

Studies in Humans

There is no information available on humans.

3.1.6 Mutagenicity

In vitro Studies

One bacterial mutation test and one chromosomal aberration test in vitro were performed [MHW, Japan: 1998]. Those studies were identified as the key studies because they were well conducted according to a current protocol. Details of the studies were as follows.

The bacterial mutation test was conducted according to the Japanese guideline for screening mutagenicity testing of chemicals and OECD guidelines No. 471 and 472 [MHW, Japan: 1998]. Diisopropylbenzene (57.0 % meta and 41.0 % para) was added to Salmonella typhimurium TA100, TA1535, TA98, TA1537 and Escherichia coli WP2 uvrA.

This chemical failed to induce mutations up to the maximum doses of 5,000 ug/plate (WP2 uvrA) and 50.0 ug/plate (other strains) without S9 mix, and 625 ug/plate (WP2 uvrA) and 200 ug/plate (other strains) with S9 mix. Growth inhibition was observed at 12.5 ug/plate and more (TA100, TA98), 6.25 ug/plate and more (TA1535, TA1537) and 5,000 ug/plate (WP2 uvrA) without S9 mix, and at 100 ug/plate and more (TA100, TA1535, TA98, TA1537) and 625 ug/plate (WP2 uvrA) with S9 mix.

A chromosomal aberration test according to the Japanese guideline for screening mutagenicity testing of chemicals and OECD guideline No. 473 was conducted in cultured Chinese hamster lung (CHL/IU) cells [MHW, Japan: 1998]. Diisopropylbenzene constituted of 57.0 % meta- and 41.0 % para-isomer was used. The maximum concentrations were set up at 0.06 mg/ml without S9 mix in the continuous treatment, or 0.12 mg/ml with and without S9 mix in the short-term treatment, based on the results of the growth inhibition test.

Structural chromosomal aberrations and polyploidy were not induced under the conditions of this experiment. Cytotoxicity was observed at 0.03 mg/ml and more after continuous treatment and at 0.015 mg/ml and more after short-term treatment without S9 mix.

In vivo Studies

No studies are found on genotoxicity in vivo.

Conclusion

Diisopropylbenzene was not genotoxic with and without an exogenous metabolic activation system in bacterial tests as well as a chromosomal aberration test in vitro.

3.1.7 Carcinogenicity

There is no data available.

3.1.8 Toxicity for Reproduction/Development

Studies in Animals

Results are available from a reproductive/developmental toxicity screening test according to OECD TG 421 [MHLW, Japan: 2001]. This study was identified as a key study because it was well conducted and reported. Details of the study are as follows.

Diisopropylbenzene (meta: 58.5 %, para: 39.7 %) was administered to Crj: CD (SD) rats by gavage at doses of 0 (vehicle; corn oil), 6, 30, 150 and 750 mg/kg b.w./day from day 14 before mating to day 14 after mating in males and to day 3 of lactation in females [MHLW, Japan: 2001].

For parent animals, there were no effects on the observation of sperm, estrus cycles and histopathology of the testes, epididymides and ovaries. No effects were also observed on reproductive performance such as mating, fertility, delivery and lactation. For offspring, there were no dose-related changes in the number of pups born, stillbirths, live pups born and live pups on day 4 of lactation, the sex ratio, the live birth index, the viability index, the general condition, the body weight and the autopsy findings. Therefore, the NOAEL for reproductive/developmental toxicity was considered to be 750 mg/kg b.w./day.

Studies in Humans

There is no information available on humans.

Conclusion

There is no evidence that this chemical has reproductive/developmental toxicity in rats. The NOAEL for the reproductive/developmental toxicity was considered to be 750 mg/kg b.w./day.

Other

There is some information available on three isomers of diisopropylbenzene as follows.

o-Diisopropylbenzene (Cas No. 577-55-9)

One death occurred in ten rats orally dosed at 5.0 g/kg b.w. [Gerarde: 1960]. There are no other information available on this chemical.

m-Diisopropylbenzene (Cas No. 99-62-7)

No death occurred in ten rats orally dosed at 5.0 g/kg b.w. [Gerarde: 1960]. The oral LD50 values were reported to be 7,400 mg/kg b.w. in rats and 3,100 mg/kg b.w. in mice [Izmerov et al.: 1982]. Oral administration at 45.5 mg/kg b.w. to rats for 26 weeks caused muscle contraction or spasticity, alteration of classical conditioning and changes in erythrocyte count [Sologubov & Bogdanova: 1971]. After inhalation exposure at 1,000 mg/m3 for 22 weeks, liver functions were impaired in rats, mice and rabbits [Pavlova: 1971]. In rats exposed at 200 mg/m3 and 1,000 mg/m3 for 5 months, the sexual cycle was disturbed, the capacity for conception was decreased and the average number of offspring decreased as well as their weight [Elisuiskaia: 1970a]. Inhalation exposure of female mice and rats at 100-300 mg/m3 for 30 days decreased the fertility and caused miscarriages

and inhibition of growth in progeny [Elisuiskaia: 1970b]. However, the reliability of these Russian reports from the 1970s on the repeated dose and reproductive toxicity was uncertain because no details could be obtained.

p-Diisopropylbenzene (Cas No. 100-18-5)

One death occurred in ten rats orally dosed at 5.0 g/kg b.w. [Gerarde: 1960]. The oral LD50 values were reported to be 3,400 mg/kg b.w. in mice [Izmerov et al.: 1982]. No other information are available on this chemical.

3.2 Initial Assessment for Human Health

The mixture of m- and p-diisopropylbenzene (60:40/m-:p-) was assessed.

There is no data available regarding toxicokinetics and metabolism.

No data are available for the acute toxicity of diisopropylbenzene with the composition of 60:40/m-:p-. The oral LD50 value of each constitutive isomer, m-diisopropylbenzene and p-diisopropylbenzene, was shown to be greater than 3,000 mg/kg b.w. in rodents. Acute toxicity studies of diisopropylbenzene with an unknown isomer ratio are available. The oral and dermal acute toxicity of diisopropylbenzene are negligible: the oral LD50 in rats is 5,850 mg/kg b.w. and the dermal LD50 in rabbits is 14,400 mg/kg b.w. In an acute inhalation study, no deaths occurred at doses below 5,300 mg/m3 after 4h exposure in rats and after 2h exposure in mice. The weight of evidence shows that the acute toxicity of diisopropylbenzene with the composition of 60:40/m-p- can be considered to be low.

There is no reliable information on eye and skin irritation and sensitization.

In accordance with a Japanese guideline, equivalent to OECD TG 407, SD rats received diisopropylbenzene with the composition of 60:40/m-p- by gavage at doses of 0, 6, 30, 150 and 750 mg/kg b.w./day for 28 days. Mydriasis was observed in 2 of 6 males and 2 of 6 females at 150 mg/kg b.w./day and in 10 of 12 males and all of 12 females at 750 mg/kg b.w./day. This sign was observed from about 2 to 6 hours after administration mainly during the latter half of the dosing period. On histopathological examination, centrilobular hypertrophy of hepatocytes was observed in 1 of 6 males in the 150 mg/kg b.w./day group and 1 of 6 males and 4 of 6 females in the 750 mg/kg b.w./day group. Based on mydriasis and histopathological changes in the liver at 150 mg/kg b.w./day, the NOAEL of this chemical was considered to be 30 mg/kg b.w./day.

In a reverse gene mutation assay [OECD TG 471, 472], diisopropylbenzene with the composition of 60:40/m-p- was not mutagenic in Salmonella typhimurium TA100, TA1535, TA98, TA1537 and Escherichia coli WP2 urvA with and without an exogenous metabolic activation. In a chromosomal aberration test [OECD TG 473] with diisopropylbenzene with the composition of 60:40/m-:p-, structural chromosomal aberrations and polyploidy were not induced with and without an exogenous metabolic activation in cultured Chinese hamster lung (CHL/IU) cells.

There are no data available on carcinogenicity.

In a reproductive/developmental toxicity screening test [OECD TG 421], diisopropylbenzene with the composition of 60:40/m-:p- was administered to SD rats by gavage at doses of 0, 6, 30, 150 and 750 mg/kg b.w./day from day 14 before mating to day 14 after mating in males and to day 3 of lactation in females. This treatment produced no effect on reproductive performance and no adverse effect on offspring. In view of these findings the NOAEL for reproductive/developmental toxicity is considered to be 750 mg/kg b.w./day.

4 HAZARDS TO THE ENVIRONMENT

4.1 Aquatic Effects

Diisopropylbenzene has been tested with a number of aquatic species from three trophic levels. Results are summarized in Table 4.1.

Table 4.1: Summary of effects of Diisopropylbenzene on aquatic organisms

Organisms	Test duration	Result (mg/L)	Reference
Aquatic plants, e.g. algae			
Green alga (*Selenastrum capricornutum*)	72 h closed system	Growth rate method ErC50 = 2.7 > W.S. NOErC = 0.69 > W.S. Biomass method EbC50 = 1.6 > W.S. NOEbC = 0.31 > W.S.	MOE, Japan(2000)
Invertebrates			
Daphnid (*Daphnia magna*)	48 h semistatic	Immobilization EC0 = 0.246 > W.S EC50 = 0.392 > W.S	MOE, Japan(2000)
	21 d semistatic	Reproduction EC50 = 0.154 > W.S LOEC = 0.168 > W.S. NOEC = 0.063	MOE, Japan(2000)
Fish			
Medaka (*Oryzias latipes*)	96 h semistatic	LC100 = 1.48 > W.S LC50 = 0.71 > W.S LC0 = 0.25 > W.S	MOE, Japan(2000)

These toxicity data shown in Table 4.1 were obtained from the tests carried out according to OECD TG under GLP and well documented (MOE, Japan, 2000). Analytical measurement was conducted in each tests, the results were calculated based on measured concentration.

Acute Toxicity Test Results

In an algal growth inhibition test (OECD TG 201, *Selenastrum capricornutum*, closed system), the 72 h EC50s were 2.7 mg/L (growth rate method) and 1.6 mg/L (biomass method). The analytical monitoring showed that the concentration of diisopropylbenzene in each test water were reduced after the 72 h test period to 24.6 – 30.9 % of those of initial measured concentration at 0 h although the test was conducted using a dispersant which was a mixture of HCO-40 (60 mg/L) and dimethl formamide(DMF, 20 mg/L). Because the rate of reduction during test period seemed to be consistent among the exposure concentrations, the toxicity values were calculated based on the geometric mean of the measured concentrations at 0 h and 72 h.

In an acute toxicity test with daphnids (OECD TG 202 part 1, *Daphnia magna*, semistatic), the 48 h EC50 was 0.39 mg/L and the EC0 was 0.25 mg/L. In this test a mixture of 6 mg/L HCO-40 and 6 mg/L DMF was used as a dispersant and Elendt M4 medium was used as dilution water. The concentration of the test substance were kept at 57 – 61 % of the nominal concentrations during the test period, so the toxicity was calculated based on the geometric mean of two measured concentrations such as fresh (at 0 h) and old (at 24 h) test water.

One report on the toxicity to a seawater species (*Balanus amphitrite niveus*, Donahur et al., 1972) was available, but the reliability was considered to be low due to lack of information on the test method.

In an acute toxicity test with fish (OECD TG 203, *Oryzias latipes*, semistatic), the 96 h LC50 was 0.71 mg/L and the LC0 was 0.25 mg/L. In this test a mixture of HCO-40 (40 mg/L) and DMF (60 mg/L) was used as a dispersant, and the diisopropylbenzene concentration was kept for a 24 h period above 45 % of the nominal concentration. The test solution was replaced every 24 h. The toxicity values were calculated based on the geometric mean of measured concentrations of fresh and old test solutions. All fish were killed at the highest concentration of 1.48 mg/L. Toxic symptoms, abnormal swimming and/or paralysis were observed at concentrations of 0.43 and 0.82 mg/L.

All acute toxicity results are greater than the water solubility of diisopropylbenzene (m-; 0.072 mg/L and p- ; 0.0405 mg/L).

Chronic Toxicity Test Results

Two chronic toxicity values, for algae (*Selenastrum capricornutum*) and for daphnids (*Daphnia magna*) are available. For algae, 72 h NOECs on growth inhibition of 0.69 mg/L (growth rate method) and 0.31 mg/L (biomass method) were reported (MOE, Japan, 2000). The test conditions were the same as in the acute test mentioned above.

In a *Daphnia magna* reproduction toxicity test (OECD TG 211) with diisopropylbenzene, a 21 d NOEC was 0.063 mg/L and a LOEC was 0.168 mg/L calculated based on measured concentrations (time weighted averages). The cumulative number of offspring produced during 21 days test period at 0.063 mg/L was 145, and that at 0.168 mg/l was 46.7 (a third of the control and the lower concentrations). An EC50 for reproduction would be between 0.1-0.2 mg/L, however the original report presented an EC50 value of 0.154 mg/L, estimated from a few data.

The lowest chronic NOEC is 0.063 mg/L (the LOEC was 0.168 mg/L) which is close to the water solubility of this substance.

4.2 Terrestrial Effects

There is no available information.

4.3 Other Environmental Effects

There is no available information.

4.4 Initial Assessment for the Environment

The melting points of m- and p- diisopropylbenzene are –63 and –17.1 degree C respectively. The boiling point of m- and p- diisopropylbenzene is 203 degree C. The vapour pressures of m- and p- diisopropylbenzene are 0.524 hPa and 0.328 hPa respectively at 25 degree C. Water solubility is (m-) 72.0 ug/L, (p-) 40.5ug/L at 25 degree C. Henry's law constants of m- and p- diisopropylbenzene are 1.17 and 1.30 atm m^3/moL, respectively. The partition coefficients of m- and p- diisopropylbenzene are 5.13 and 5.23 respectively.

The fugacity model (Mackay level III) suggests that if diisopropylbenzene is released to air or soil, it is unlikely to distribute into other compartments and that if it is released to water it has a tendency to go into the sediment compartment.

m- and p-Diisopropylbenzene are not inherently biodegradable and both of their bioconcentration potentials seem to be high (BCF= 503 – 3210, mixture). In the air, m- and p-diisopropylbenzene are expected to be photodegraded (m-: T1/2 = 8.3 hours, p-: T1/2 = 13 hours) by OH radicals. Hydrolysis is not expected to occur.

The acute toxicity of diisoprophylbenzene has been tested in three aquatic species belonging to three trophic levels. For algae (OECD TG 201, *Selenastrum capricornutum*, closed system) a 72h EbC50 of 1.6 mg/L and a 72hErC50 of 2.7 mg/L were determined. For daphnids (OECD TG 202 part 1, *Daphnia magna*, semistatic) a 48hEC50 of 0.39 mg/L, and for fish (OECD TG 203, *Oryzias latipes*) a LC50 of 0.71 mg/L were reported.

Regarding chronic toxicity, for algae (OECD TG 201 growth inhibition, Selenastrum capricornutum) a 72 h NOErC of 0.69 mg/L, a 72 h NOEbC of 0.31 mg/L, and for daphnids (TG 211 reproduction, Daphnia magna) a 21 d NOEC of 0.063 mg/L were reported.

Most of the toxicity results were above the water solubility limit of the substance (m-; 0.072 mg/L and p- ; 0.0405 mg/L). As the NOEC of the chronic toxicity study with daphnids is close to the solubility limit of the substance, it can be considered that this result reflects the actual toxicity of the substance.

5 RECOMMENDATIONS

Human Health: The chemical is currently of low priority for further work based on a low hazard potential.

Environment: The chemical is a candidate for further work.

Diisopropylbenzene is not inherently biodegradable and is expected to have a high bioaccumulation potential and aquatic toxicity. An environmental exposure assessment and if necessary a risk assessment is recommended.

6 REFERENCES

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I U C L I D D A T A S E T

Existing Chemical	:	ID: 25321-09-9
CAS No.	:	25321-09-9
EINECS Name	:	diisopropylbenzene
EINECS No.	:	246-835-6
TSCA Name	:	Benzene, bis(1-methylethyl)-
Molecular Formula	:	C12H18

Producer Related Part

Company	:	National Institute of Health & Sciences
Creation date	:	24.12.0002

Substance Related Part

Company	:	National Institute of Health & Sciences
Creation date	:	24.12.0002
Memo	:	
Printing date	:	08.01.2003
Revision date	:	
Date of last Update	:	24.12.2002
Number of Pages	:	1
Chapter (profile)	:	Chapter: 1, 2, 3, 4, 5, 7
Reliability (profile)	:	Reliability: without reliability, 1, 2, 3, 4
Flags (profile)	:	Flags: without flag, confidential, non confidential, WGK (DE), TA-Luft (DE), Material Safety Dataset, Risk Assessment, Directive 67/548/EEC, SIDS

1.0.1 OECD AND COMPANY INFORMATION

Type : sponsor country
Name : Chemicals Evaluation and Research Institute(CERI)
Partner :
Date :
Street : 1-4-25 Koraku, Bunkyo-ku
Town : 112-0004 Tokyo
Country : Japan
Phone : 03-5804-6135
Telefax : 03-5804-6139
Telex :
Cedex :
Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
Flag : Critical study for SIDS endpoint
26.07.2002

1.0.2 LOCATION OF PRODUCTION SITE

1.0.3 IDENTITY OF RECIPIENTS

1.1 GENERAL SUBSTANCE INFORMATION

Substance type : organic
Physical status : liquid
Purity : % w/w
Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
Reliability : (1) valid without restriction
Flag : Critical study for SIDS endpoint
26.07.2002

1.1.0 DETAILS ON TEMPLATE

1.1.1 SPECTRA

1.2 SYNONYMS

Benzene, bis(1-methylethyl)-
Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
Flag : Critical study for SIDS endpoint

26.07.2002

Diisopropylbenzene (mixture)
Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
26.07.2002

Diisopropylbenzene, all isomers
Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
26.07.2002

1.3 IMPURITIES

1.4 ADDITIVES

1.5 QUANTITY

Production during the last 12 months	:	
Import during the last 12 months	:	
Quantity produced	:	10 000 - 50 000 tonnes in 2000
Remark	:	In Japan, m- and p-diisopropylbenzene are produced as byproducts of cumene synthesis. The production volume was ca.30,000 tonnes/year.
Source	:	Chemicals Evaluation and Research Institute (CERI) Tokyo
Reliability	:	(1) valid without restriction
Flag	:	Critical study for SIDS endpoint

05.11.2002

1.6.1 LABELLING

1.6.2 CLASSIFICATION

1.7 USE PATTERN

Type	:	type
Category	:	Use in closed system
Remark	:	Diisopropylbenzene is blended into gasoline, diesel and other hydrocarbon fuels, and also used for diisopropylbenzeneperoxide synthesis.
Source	:	Chemicals Evaluation and Research Institute (CERI) Tokyo

Flag : Critical study for SIDS endpoint
29.07.2002

Type : use
Category : Fuel additives
Remark : blended into gasoline, diesel, other hydrocarbon fuels, synthesis for diisopropylbenzeneperoxide
Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
Reliability : (1) valid without restriction
Flag : Critical study for SIDS endpoint
05.11.2002

1.7.1 TECHNOLOGY PRODUCTION/USE

1.8 OCCUPATIONAL EXPOSURE LIMIT VALUES

1.9 SOURCE OF EXPOSURE

1.10.1 RECOMMENDATIONS/PRECAUTIONARY MEASURES

1.10.2 EMERGENCY MEASURES

1.11 PACKAGING

1.12 POSSIB. OF RENDERING SUBST. HARMLESS

1.13 STATEMENTS CONCERNING WASTE

1.14.1 WATER POLLUTION

1.14.2 MAJOR ACCIDENT HAZARDS

1.14.3 AIR POLLUTION

1.15 ADDITIONAL REMARKS

1.16 LAST LITERATURE SEARCH

1.17 REVIEWS

1.18 LISTINGS E.G. CHEMICAL INVENTORIES

2.1 MELTING POINT

Value	:	= -63 - -17.1 ° C
Sublimation	:	
Method	:	other: unknown
Year	:	
GLP	:	no data
Test substance	:	no data
Remark	:	MP of m-Diisopropylbenzene is -63 degree C. MP of p-Diisopropylbenzene is -17.1 degree C.
Source	:	m-: LANGE'S HANDBOOK OF CHEMISTRY (13th edition) p-: HAZARDOUS SUBSTANCE DATA BANK (U.S. NATIONAL LIBRARY OF MEDICINE) Chemicals Evaluation and Research Institute (CERI) Tokyo
Conclusion	:	MP of Diisopropylbenzene is -63 - -17.1 degree C.
Reliability	:	(2) valid with restrictions
Flag	:	Critical study for SIDS endpoint

01.11.2002

2.2 BOILING POINT

Value	:	= 203 ° C at 1013 hPa
Decomposition	:	
Method	:	other: unknown
Year	:	
GLP	:	no data
Test substance	:	no data
Source	:	LANGE'S HANDBOOK OF CHEMISTRY (13th edition) Chemicals Evaluation and Research Institute (CERI) Tokyo
Conclusion	:	Boiling point of m- and p- Diisopropylbenzene is each 203 degree C.
Reliability	:	(2) valid with restrictions
Flag	:	Critical study for SIDS endpoint

01.11.2002

2.3 DENSITY

Type	:	density
Value	:	= .8549 g/cm3 at 25° C
Method	:	other: unknown
Year	:	2000
GLP	:	no data
Test substance	:	
Source	:	Wako Pure Chemical Industries, Ltd. Chemicals Evaluation and Research Institute (CERI) Tokyo

Test substance	:	Wako Pure Chemical Industries, Ltd. Purity ; mixture of m- and p-configuration (58.4 % w/w of m- Diisopropylbenzene, 41.4 % w/w of p - Diisopropylbenzene)
Conclusion	:	Density of Diisopropylbenzene (mixture) is 0.8549 g/cm3.
Reliability	:	(2) valid with restrictions
Flag	:	Critical study for SIDS endpoint

01.11.2002

2.3.1 GRANULOMETRY

2.4 VAPOUR PRESSURE

Value	:	= 1.3 hPa at 25° C
Decomposition	:	
Method		
Year	:	
GLP	:	no data
Test substance	:	no data
Remark	:	The data of the Hazardous Substance Data Bank (U.S. National library of Medicine) was 1 mmHg (m-Diisopropylbenzene at 34.7 degree C, p-Diisopropylbenzene at 40.0 degree C), it was converted into hPa unit.
Source	:	SRC PhysProp Database Chemicals Evaluation and Research Institute (CERI) Tokyo
Conclusion		Vapour pressure of m- Diisopropylbenzene is 1.3 hPa at 34.7 degree C, and vapour pressure of p- Diisopropylbenzene is 1.3 hPa at 40.0 degree C.
Reliability	:	(2) valid with restrictions
Flag	:	Critical study for SIDS endpoint

01.11.2002

Value	:	= .328 hPa at 25° C
Decomposition	:	
Method		
Year	:	
GLP	:	no data
Test substance	:	
Remark	:	The data of the SRC PHYPROP DATABASE was 0.246mmHg (at 25 degree C), it was converted into hPa unit. The vapour pressure of p-isopropylbenzene is 0.328hPa.
Source	:	SRC PHYPROP DATABASE Chemicals Evaluation and Research Institute (CERI) Tokyo
Reliability	:	(1) valid without restriction
Flag	:	Critical study for SIDS endpoint

31.10.2002 (2)

Value : = .524 hPa at 25° C
Decomposition :
Method
Year :
GLP : no data
Test substance :
Remark : The data of the SRC PHYPROP DATABASE was 0.246mmHg (at 25
degree C), it was converted into hPa unit. The vapour pressure of p-isopropylbenzene is 0.328hPa.
Source : SRC PHYPROP DATABASE
Chemicals Evaluation and Research Institute (CERI) Tokyo
Reliability : (1) valid without restriction
Flag : Critical study for SIDS endpoint
31.10.2002 (2)

2.5 PARTITION COEFFICIENT

Log pow : = 5.13 - 5.23 at 25° C
Method OECD Guide-line 107 "Partition Coefficient (n-octanol/water), Flask-shaking Method"
Year : 1983
GLP : no
Test substance :
Source : Chemicals Evaluation and Research Institute, Japan
Chemicals Evaluation and Research Institute (CERI) Tokyo
Test substance : Wako Pure Chemical Industries, Ltd.
Purity 97.2% (mixture)
Conclusion : LogPow of m - Diisopropylbenzene is 5.13, and LogPow of p-Diisopropylbenzene is 5.23.
Reliability : (1) valid without restriction
Flag : Critical study for SIDS endpoint
31.10.2002

2.6.1 WATER SOLUBILITY

Value : = .0405 - .072 mg/l at 25 ° C
Qualitative : insoluble (< 0.1 mg/L)
Pka : at 25 ° C
PH : at and ° C
Method : OECD Guide-line 105 "Water Solubility"
Year : 1983
GLP : no
Test substance : no data
Source : Chemicals Evaluation and Research Institute, Japan

Chemicals Evaluation and Research Institute (CERI) Tokyo

Conclusion : Water solubility of m - Diisopropylbenzene is 0.072 mg/L at 25 degree C.
Water solubility of p - Diisopropylbenzene is 0.0405 mg/L at 25 degree C.

Reliability : (1) valid without restriction

Flag : Critical study for SIDS endpoint

25.07.2002

2.6.2 SURFACE TENSION

2.7 FLASH POINT

2.8 AUTO FLAMMABILITY

2.9 FLAMMABILITY

2.10 EXPLOSIVE PROPERTIES

2.11 OXIDIZING PROPERTIES

2.12 ADDITIONAL REMARKS

3.1.1 PHOTODEGRADATION

Type	:	air
Light source	:	
Light spect.	:	nm
Rel. intensity	:	based on Intensity of Sunlight
Indirect photolysis		
Sensitizer	:	OH
Conc. of sens.	:	1500000 molecule/cm3
Rate constant	:	= .0000000000101158 cm3/(molecule*sec)
Degradation	:	= 50 % after 12.7 hour(s)
Deg. Product	:	
Method	:	other (calculated)
Year	:	
GLP	:	
Test substance	:	
Remark	:	The reaction rate constant with hydroxyl radicals was estimated by SRC AOPWIN. Photodegradation of m- Diisopropylbenzene is as follows ; The half-life time (8.268 hours) was calculated based on the calculated rate constant (15.524E-12 cm3/mol-sec) and hydroxyl radicals concentration in atmosphere of 1.5E6 OH/cm3. Photodegradation of p- Diisopropylbenzene is as follows ; The half-life time (12.688 hours) was calculated based on the calculated rate constant (10.1158E-12 cm3/mol-sec) and hydroxyl radicals concentration in atmosphere of 1.5E6 OH/cm3.
Source	:	Chemicals Evaluation and Research Institute (CERI) Tokyo
Conclusion	:	Half-life time of m- and p- Diisopropylbenzene is 8.268 - 12.688 hours.
Flag	:	Critical study for SIDS endpoint

25.07.2002

3.1.2 STABILITY IN WATER

Type	:	abiotic
t1/2 pH4	:	at degree C
t1/2 pH7	:	at degree C
t1/2 pH9	:	at degree C
Degradation	:	< 10 % after 5 day at pH and 50 degree C
Deg. Product	:	no
Method	:	OECD Guide-line 111 "Hydrolysis as a Function of pH"
Year	:	2000
GLP	:	no

Test substance	:	
Source	:	Chemicals Evaluation and Research Institute, Japan Chemicals Evaluation and Research Institute (CERI) Tokyo
Test substance	:	Wako Pure Chemical Industries, Ltd. Purity ; mixture of m- and p-configuration (58.4 % w/w of m- Diisopropylbenzene, 41.4 % w/w of p - Diisopropylbenzene)
Conclusion	:	This chemical is stable at pH 4, 7 and 9 at 50 degree C for five days.
Reliability	:	(1) valid without restriction
Flag	:	Critical study for SIDS endpoint

31.10.2002

3.1.3 STABILITY IN SOIL

3.2 MONITORING DATA

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

Type	:	fugacity model level III
Media	:	
Air (level I)	:	
Water (level I)	:	
Soil (level I)	:	
Biota (level II / III)	:	
Soil (level II / III)	:	
Method	:	
Year	:	2002
Method	:	Parameters used in calculation of distribution by Mackay level III fugacity model are as follows; Regarding to m - Diisopropylbenzene; Melting point: - 63 degree C Vapour pressure: 52.4 Pa Water solbility: 0.072 mg/L LogPow: 5.13 As this chemical is not inherently biodegradable, we assume the following half-life times; Half-life time in air: 8.268 hours Half-life times in water: 240000 hours Half-life times in soil: 240000 hours Half-life times in sediment: 720000 hours Regarding to p - Diisopropylbenzene; Melting point: - 17.1 degree C Vapour pressure: 32.8 Pa

Water solbility: 0.0405 mg/L
LogPow: 5.23
As this chemical is not inherently biodegradable, we assume the following half-life times;
Half-life time in air: 12.688 hours
Half-life times in water: 240000 hours
Half-life times in soil: 240000 hours
Half-life times in sediment: 720000 hours

Result : m- Diisopropylbenzene is as follows;

Compartment	Release: 100 % to air	100 % to water	100 % to soil
Air	99.1 %	1.0 %	1.9 %
Water	0.0 %	22.6 %	0.0 %
Soil	0.9 %	0.0 %	98.1 %
Sediment	0.0 %	76.4 %	0.0 %

p- Diisopropylbenzene is as follows;

Compartment	Release: 100 % to air	100 % to water	100 % to soil
Air	99.0 %	1.2 %	2.4 %
Water	0.0 %	19.6 %	0.0 %
Soil	1.0 %	0.0 %	97.6 %
Sediment	0.0 %	79.2 %	0.0 %

Source : Chemicals Evaluation and Research Institute (CERI) Tokyo
Conclusion : If this chemical is released to one of the compartments of air and soil, it has a tendency to remain in the original compartment.
If this chemical is released to water, it has a tendency to go into the sediment compartment.
Reliability : (1) valid without restriction
Flag : Critical study for SIDS endpoint
31.10.2002

3.3.2 DISTRIBUTION

3.4 MODE OF DEGRADATION IN ACTUAL USE

3.5 BIODEGRADATION

Type : aerobic

Inoculum	:	activated sludge
Concentration	:	30mg/l related to Test substance related to
Contact time	:	14 day
Degradation	:	= 0 % after 14 day
Result	:	under test conditions no biodegradation observed
Control substance	:	Aniline
Kinetic	:	% %
Deg. Product	:	no
Method	:	OECD Guide-line 302 C "Inherent Biodegradability: Modified MITI Test (II)"
Year	:	1982
GLP	:	no
Test substance	:	other TS
Remark	:	The concentration of activated sludge is 100mg/L.
Result	:	The biodegradations of this chemical were as follows; 0 % by BOD after 14 days 0 % by GC after 14 days.
Source	:	Chemicals Evaluation and Research Institute, Japan Chemicals Evaluation and Research Institute (CERI) Tokyo
Test substance	:	Wako Pure Chemical Industries, Ltd. Purity ; mixture of m- and p-configuration (60 % w/w of m - Diisopropylbenzene, 40 % w/w of p - Diisopropylbenzene)
Conclusion	:	This chemical is not inherently biodegradable.
Reliability	:	(1) valid without restriction
Flag	:	Critical study for SIDS endpoint

31.10.2002

3.6 BOD5, COD OR BOD5/COD RATIO

3.7 BIOACCUMULATION

Species	:	Cyprinus carpio (Fish, fresh water)
Exposure period	:	56 day at 25 degree C
Concentration	:	20µg/l
BCF	:	= 513 - 3120
Elimination	:	no
Method	:	OECD Guide-line 305 E "Bioaccumulation: Flow-through Fish Test"
Year	:	1983
GLP	:	no
Test substance	:	other TS
Result	:	The bioconcentration of m- Diisopropylbenzene is as follows; 503 - 1680 at 20 ug/L of test concentration 546 - 3210 at 2 ug/L of test concentration.

The bioconcentration of p- Diisopropylbenzene is as follows;
530 - 2300 at 20 ug/L of test concentration
512 - 2960 at 2 ug/L of test concentration.

Source : Chemicals Evaluation and Research Institute, Japan
Chemicals Evaluation and Research Institute (CERI) Tokyo

Test substance : Wako Pure Chemical Industries, Ltd.
Purity ; mixture of m- and p-configuration
(61.7 % w/w of m- Diisopropylbenzene,
35.5 % w/w of p - Diisopropylbenzene)

Conclusion : The bioconcentration potential is moderately high.

Reliability : (1) valid without restriction

Flag : Critical study for SIDS endpoint

25.07.2002

3.8 ADDITIONAL REMARKS

4.1 ACUTE/PROLONGED TOXICITY TO FISH

Type	:	semistatic
Species	:	Oryzias latipes (Fish, fresh water)
Exposure period	:	96 hour(s)
Unit	:	mg/l
Analytical monitoring	:	yes
LC0	:	= .25
LC50	:	= .707
LC100	:	= 1.48
Method	:	OECD Guide-line 203 "Fish, Acute Toxicity Test"
Year	:	2000
GLP	:	yes
Test substance	:	other TS: Tokyo Kasei Kogyo Co., Ltd., Lot. No.; FGF01, Purity: o- 1.8%, m- 58.2%, p- 39.7%
Method	:	-Test Organisms: a) Supplier: Test organisms were obtained from Sankyo Suisan (Private Fish Farm,Tokyo Japan). b) Size (length and weight): 1.88cm (1.64 - 2.11 cm, n = 10) in length; 0.099 g (0.074 - 0.122 g, n = 10) in weight c) Age: Not described d) Any pretreatment: Test organisms were acclimated for at least 17 days before testing. Any groups showing > 5 % mortality for 7 days before testing were not used for testing. During acclimination, test fishes were fed with TETRAMINE equivalent to 2% of weight. These test organisms were not fed for 24 hours before the test. -Test substance: Diisopropylbenzene This test substance is very sparingly soluble in water and tends to volatilize for low boiling point. a) Empirical Formula: C12H18 b) Molecular Weight: 162.27g/mol c) Purity: o- 1.8%, m- 58.2%, p- 39.7% -Test Conditions: a) Dilution Water Source: Tap water (Yokohama city, Kanagawa, Japan) dechlorinated using activated carbon. After that Residual Chlorine was removed from the water by aeration. b) Dilution Water Chemistry: pH: = 7.6 Total hardness (as CaCO3): = 60 mg/L c) Exposure Vessel Type: 5 L test solution in a 5 L glass tank with a teflon cover sheet to prevent loss of test substance d) Nominal Concentrations: control, solvent control,0.200, 0.360, 0.630, 1.10 and 2.00 mg/L e) Stock Solution: 100mg test substance was dissolved in

pure water of 100mL with vehicles (DMF 3g + HCO60 2g).
f) Vehicle/Solvent and Concentrations: Dimethyl formamide and HCO-40 were used for solvent in test water except the control. The concentration of Dimethyl formamide and HCO-40 in the test solutions was 60mg/L and 40mg/L at the maximum, respectively.
g) Number of Replicates: 1
h) Fish per Replicates: 10
i) Renewal Rate of Test Water: Every 24 hours
j) Water Temperature: 24+/-1ºC
k) Light Condition: 16:8 hours, light-darkness cycle
l) Feeding: None
m) Aeration : Not described

-Analytical Procedure: Test substance in each test solution were measured at the start and 24th hour (before exchange of test solution), using HPLC with detection limit of 0.002mg/L.

-Statistical Method:
a) Data Analysis: Probit method, binomial or Moving average for LC50
b) Method of Calculating Mean Measured Concentrations (i.e. arithmetic mean, geometric mean, etc.): Geometric mean

Result : - Measured Concentrations : The test concentrations were measured at the start and 24th hour (before exchange of test solution). For some of them, the deviation from the nominal concentration were not less than +/- 20% of nominal concentration.

Nominal Conc. mg/L	Measured Conc., mg/L			Percent of Nominal	
	0 Hour	24 Hours	Mean*	0 Hour	24 Hours
	Fresh	Old	mg/L	Fresh	Old
Control	<0.002	<0.002	---	---	---
Solvent Control	<0.002	<0.002	---	---	---
0.200	0.168	0.111	0.137	84	56
0.360	0.298	0.209	0.250	83	58
0.630	0.526	0.353	0.431	83	56
1.10	0.942	0.706	0.816	86	69
2.00	1.73	1.26	1.48	87	63

*: Mean measured concentration (Geometric Mean)
Fresh: Start of renewal period
Old: End of renewal period

- Water chemistry (pH and DO) and temperature in test:

Water chemistry and temperature were measured for old and renewal solution with control and each concentration at the start of test and every 24 hours.
pH: 7.2 - 7.8
DO: 5.8 - 8.4 mg/L
Water Temperature: 24+/-1ºC

-Effect Data(mortality):
LC50 (96hr) = 0.707 mg/L (95% C. I.: 0.539-0.935) (mc)
LC0 (96hr) = 0.250 mg/L (mc)
LC100 (96hr) = 1.48 mg/L (mc)
mc: based on measured concentration

- Cumulative Mortality: None of test organisms were killed during exposure period at solvent control, 0.200 and 0.360 mg/L, however all test organisms were killed at 2.00 mg/L on and after 72 hours. Although at the end of test in the control 1 test fish was killed, the mortality did not exceed 10%. Therefore the test was valid OECD test guideline. The lowest concentration that the test organisms were killed was 0.630 mg/L after 72 hours.

Nominal Conc. mg/L	Mean* mg/L	Cumulative Number of Dead (Percent Mortality) 24hr	48hr	72hr	96hr
Control	---	0 (0)	0 (0)	0 (0)	1(10)
Solvent Control	---	0 (0)	0 (0)	0 (0)	0(0)
0.200	0.137	0 (0)	0 (0)	0 (0)	0(0)
0.360	0.250	0 (0)	0 (0)	0 (0)	0(0)
0.630	0.431	0 (0)	0 (0)	1 (10)	2(20)
1.10	0.816	0 (0)	2 (20)	2 (20)	5(50)
2.00	1.48	3 (30)	9 (90)	10 (100)	10(100)

*: Mean measured concentration (Geometric Mean)

-Other Effect: Toxicological symptom was first observed at 0.630mg/L (24 hour).

Nominal Conc. mg/L	Mean* mg/L	Symptoms 24hr	48hr	72hr	96hr
Control	----	n	n	n	n
Solvent Control	----	n	n	n	n
0.200	0.137	n	n	n	n
0.360	0.250	n	n	n	n
0.630	0.431	as-2	as-5	as-5	as-8
1.10	0.816	as-10	as-8	as-1	as-8

				ap-7	ap-4	
2.00		1.48	as-7	ap-1	---	---

*: Mean measured concentration (geometric mean)
n: No abnormalities are detected
as: abnormal swimming
ap: paralyzation
---: All fishes were dead at this observation time.

- Calculation of toxic values: It was the mean measured concentrations. The reason is that some of the error ranges of measured concentration were not less than +/-20% of nominal concentration.

Source : MOE,Japan(2000)
National Institute of Environmental Studies, Environment Agency Tsukuba-Ibaraki

Reliability : (2) valid with restrictions
Remarks: This test was conducted using dispersant, and the estsimated values were higher than water solubility, however an analytical monitoring was dane.

Flag : Critical study for SIDS endpoint

20.12.2002 (8)

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

Type : semistatic
Species : Daphnia magna (Crustacea)
Exposure period : 48 hour(s)
Unit : mg/l
Analytical monitoring : yes
EC0 : = .246
EC50 : = .392
Method : OECD Guide-line 202, part 1 "Daphnia sp., Acute Immobilisation Test"
Year : 2000
GLP : yes
Test substance : other TS: Tokyo Kasei Kogyo Co., Ltd., Lot. No.; FGF01, Purity: o- 1.8%, m- 58.2%, p- 39.7%
Method : - Test Organisms:
a) Age: < 24 hours old
b) Supplier/Source: Test organisms were obtained from National Institute for Environmental Studies (JAPAN) and had been reproduced in the testing laboratory for four years and six months.
c) Any pretreatment: Parental daphnids were acclimated for at least 20 days on test condition before testing, the mortality of

the parental daphnids for two weeks before testing was 0%. During acclimination, any male daphnid was not borne and test daphnids were fed with Chlorella vulgaris, 0.2 mg carbon/day/individual.

-Test substance: Diisopropylbenzene
This test substance is very sparingly soluble in water and tends to volatilize for low boiling point.
a) Empirical Formula: C12H18
b) Molecular Weight: 162.27g/mol
c) Purity: o- 1.8%, m- 58.2%, p- 39.7%

-Test Conditions:
a) Dilution Water Source: Elendt M4 medium recommended by OECD guideline No.211 was used as dilution water.
b) Exposure Vessel Type: 100mL test solution in a 100mL Glass Beaker covered with teflon sheet
c) Nominal Concentrations: control, solvent control, 0.200, 0.360, 0.630, 1.10 and 2.00 mg/L
d) Stock Solution: Diisopropylbenzene of 100mg/L was prepared for stock solution according to the following method. 50mg test substance was dissolved in 150mg Dimethyl formamide with 150mg HCO-40. The solution was diluted with pure water in 500mL.
e) Vehicle/Solvent and Concentrations: Dimethyl formamide and HCO-40 were used for solvent. The concentration of Dimethyl formamide and HCO-40 in the test solutions was 6mg/L and 6mg/L, respectively, in each concentration.
f) Number of Replicates: 4
g) Individuals per Replicates: 5
h) Renewal Rate of Test Water: Every 24 hours
i) Water Temperature: 20+/-1ºC
j) Light Condition: 16:8 hours, light-darkness cycle
k) Feeding: None- Analytical Procedure: measured at the start and 24th hour, using HPLC with detection limit of 0.002mg/L

- Statistical Method:
a) Data Analysis: Probit method, binomial or Moving average for LC50
b) Method of Calculating Mean Measured Concentrations: Geometric mean

Result : - Measured Concentrations : The test concentrations were measured at the start and 24th hour(before exchange of test solution). For some of them, the deviation from the nominal were not less than +/- 20%.

Nominal Conc.	Measured Conc., mg/L			Percent of Nominal	
mg/L	0 Hour	24 Hour	Mean*	0 Hour	24 Hour

	Fresh	Old	mg/L	Fresh	Old
Control	<0.002	<0.002	---	---	---
Solvent Control	<0.002	<0.002	---	---	
0.200	0.159	0.113	0.134	80	57
0.360	0.279	0.217	0.246	78	60
0.630	0.469	0.374	0.419	74	59
1.10	0.863	0.658	0.754	78	60
2.00	1.52	1.21	1.36	76	61

Fresh: freshly prepared test solution
Old: test solution after 24 hours exposure
*: Mean measured concentration during 24 hours (Geometricmean)

- Water chemistry (pH and DO) and temperature in test:
Water chemistry and temperature were measured for control and each concentration at the start and 24th hour.
pH: 8.2 - 8.6
DO: 8.4 - 8.8 mg/L
Water Temperature: 19.9 - 20.0ºC

-Effect Data:
EC50 (24hr) = 0.725 mg/L (mc) (95% C.I.=0.624-0.843 mg/L)
EC0 (24hr) = 0.246 mg/L (mc)
EC50 (48hr) = 0.392 mg/L (mc) (95% C. I. =0.246-0.754 mg/L)
EC0 (48hr) = 0.246 mg/L (mc)
mc: based on measured concentration

- Mortality or Immobility: No test organisms were immobilized at control, solvent control, 0.200 and 0.360mg/L. One individual was immobilized at 0.630mg/L after 24 hours. All individuals were immobilized at 1.10mg/L after 48th hour and 2.00mg/L on and after 24th hour.

Nominal Conc. mg/L	Mean* mg/L	Cumulative Number of Immobilized Daphnids (Percent Mortality or Immobility) 24Hour	48Hour
Control	----	0 (0)	0 (0)
Solvent Control	----	0 (0)	0 (0)
0.200	0.134	0 (0)	0 (0)
0.360	0.246	0 (0)	0 (0)
0.630	0.419	1 (5)	12 (60)
1.10	0.754	10 (50)	20 (100)
2.00	1.36	20 (100)	20 (100)

*: Mean measured concentration (Geometric Mean)

- Calculation of toxicity values: To calculate toxicity, the mean measured concentrations was used.

Source : MOE,Japan(2000)

Reliability : (2) valid with restrictions
- Remarks: This test was conducted using solvent (mixture of a dispersant of HCO40 and DMF) and the estimated values were higher than water solubility, however an analytical monitoring was done. In this test, it could not separate the true toxicity of the test substance from the physical effect of non-dissolving substance

Flag : Critical study for SIDS endpoint
20.12.2002 (8)

Type : static
Species : other: Balanus amphitrite niveus
Exposure period : 1 hour(s)
Unit :
Analytical monitoring :
Method :
Year : 1977
GLP :
Test substance :
Method : -Test Organisms:Nauplii released from freshly collectedbarnacles Balanus amphitrite niveus were used.

-Test substance: Diisopropylbenzene
a) Empirical Formula: C12H18
b) Molecular Weight: 162.27g/mol

-Test Conditions: The sea water used in the experiments was collected offshore from Port Aransas, and dilluted to 30 per mil. Experiments carried out room temperature, approximately 22ºC.

Source : National Institute of Environmental Studies, Environment Agency Tsukuba-Ibaraki
National Institute of Environmental Studies, Environment Agency Tsukuba-Ibaraki

Reliability : (4) not assignable
20.12.2002 (3)

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

Species	:	Selenastrum capricornutum (Algae)
Endpoint	:	Growth rate and biomass
Exposure period	:	72 hour(s)
Unit	:	mg/l
Analytical monitoring	:	yes
NOEC	:	= . 0.69(growth rate), =0.31(biomass)
EC50	:	= 2.7(growth rate), =1.6(biomass)
Method	:	OECD Guide-line 201 "Algae, Growth Inhibition Test"
Year	:	2000
GLP	:	yes
Test substance	:	other TS: Tokyo Kasei Kogyo Co., Ltd., Lot. No.; FGF01, Purity: o- 1.8%, m- 58.2%, p- 39.7%
Method	:	-Test Organisms a) Supplier/Source: Obtained from American Type Culture Collection and reproduced in aseptic culture for 6 months. b) Method of Cultivation: Sterile c) Stain Number: ATCC22662 -Test substance: Diisopropylbenzene This test substance is very sparingly soluble in water and tends to volatilize for low boiling point. a) Empirical Formula: C12H18 b) Molecular Weight: 162.27g/mol c) Purity: o- 1.8%, m- 58.2%, p- 39.7% - Test Conditions: a) Medium: OECD medium b) Exposure Vessel Type: 100 mL Medium in an 500mL Erlenmeyer Flask with glass cap(closed system) c) Nominal Concentrations: control, solvent control, 0.200, 0.430, 0.930, 2.00, 4.31, 9.28 20 mg/L d) Stock Solution: 100mg test substance was dissolved in 100mg DMF with 300mg HCO-40. The solution was diluted with OECD medium in 1000mL. e) Vehicle/Solvent and Concentrations: Dimethyl formamide(DMF) and HCO-40 were used for solvent. DMF 20.0mg/L and HCO-40 60.0mg/L were contained in each test solution and solvent control. f) Number of Replicates: 3 g) Initial Cell Number: 10,000 cells/mL h) Water Temperature: 23+/-2ºC i) Light Condition: 3,200 - 4,800 lux, continuously j) Shaking: 100 rpm- Analytical Procedure: Test concentrations were measured at the start and the 72nd hour, using HPLC with detection limit 0.002mg/L.

- Statistical Method:
a) Data Analysis: The EC50 values and associated 95%confidence limits were determined by least squares linear regression analysis of the logarithm of m easured test concentration against percent growth inhibition relative to the solvent control. The NOEC values were determined by analysis of variance (ANOVA), Dunnet test, subsequent to Bartlett test for homogeneity of variances.
b) Method of Calculating Mean Measured Concentrations (i.e. arithmetic mean, geometric mean, etc.): Geometric mean of measured concentrations at 0 hr and at 72 hr were used for calculation.

Remark : NOEC was determined based on growth inhibition.
Result : - Measured Concentrations : The tested concentrations were measured at the start and the 72nd hour. The concentrations of 72 hours after were low. Those percent of nominal were 17 - 23%. The test substance seemed to be incorporated into cell slightly.

Nominal conc. mg/L	Measured Conc., mg/L 0 Hour	72 Hour	Percent of nominal 0 Hour	72 Hour	Mean*
Control	<0.002	<0.002	---	---	---
Solvent Control	<0.002	<0.002	---	---	---
0.200	0.138	0.034	69	17	0.068
0.430	0.291	0.076	68	18	0.15
0.930	0.615	0.158	66	17	0.31
2.00	1.37	0.345	69	17	0.69
4.31	2.98	0.762	69	18	1.51
9.28	6.25	1.90	70	20	3.45
20.0	14.8	4.57	74	23	8.22

* Geometric mean

- Water chemistry (pH) in test:
pH was measured for control and each concentration at the start and end of test.
pH: 8.1 - 10.6

-Effect Data:
Area Method
EbC50(0-72hr) = 1.6 mg/L (mc) (95% C.I.: 1.77-5.44 0.88-2.72 mg/L)
NOEC (0-72hr) = 0.31 mg/L (mc)

Rate Method

ErC50(24-48hr) >= 3.05 mg/L (mc) (95% C.I.: 1.65-5.5 mg/L)
NOErC(24-48hr) = 0.69 mg/L (mc)
ErC50(24-72hr) >= 2.7 mg/L (mc)
NOErC = 0.69 mg/L (mc)
mc: based on mean measured concentration

- Percent Growth Inhibition of Selenastrum capricornutum

Measured Conc. mg/L	Area under the growth curves (Average) Area A (0-72hr)	Inhibition (%)*1 IA (0-72hr)
Control	7,934,000	---
Solvent Control	10,816,000	---
0.068	10,995,000	-1.7
0.15	10,629,000	1.7
0.31	9,770,000	9.7
0.69	9,170,000	15.2*
1.51	6,651,000	38.5**
3.45	1,638,000	84.9**
8.22	397,000	96.3**

Measured Conc. mg/L	Growth rates and percent inhibition(Average) Rate u(24-48hr)	Inhibition(%)*1 Im(24-48hr)	Rate u(24-72hr)	Inhibition(%)*1 Im(24-72hr)
Control	0.0684	---	0.0629	---
Solvent Control	0.0680	---	0.0593	---
0.068	0.0691	-1.6	0.0599	-1.0
0.15	0.0691	-1.6	0.0591	0.3
0.31	0.0661	2.8	0.0582	1.9
0.69	0.0648	4.7	0.0570	3.9
1.51	0.0552	18.8**	0.0526	11.3**
3.45	0.0300	55.9**	0.0227	61.7**
8.22	0.0094	86.2**	0.0057	90.4**

*1 : Values are the percent inhibition relative to the control
*: Indicates a significant difference (alpha=0.05) from the solvent control
**: Indicates a significant difference (alpha=0.01) from the solvent control

- Growth Curves: Log phase during the test period

- Calculation of toxicity values: The calculation of toxicity values was the mean measured concentrations. The reason is that some of the error ranges of measured concentration were not less than +/-20% of nominal concentration.

Source : MOE,Japan(2000)
Reliability : (2) valid with restrictions
Remarks: This test was conducted using solvent(mixture of a dispersant of HCO40 and DMF) and the estimated values were higher than water solubility, however an analytical monitoring was done. In this test, it could not separate the true toxicity of the test substance from the physical effect of non-dissolving substance
Flag : Critical study for SIDS endpoint
20.12.2002 (9)

4.4 TOXICITY TO MICROORGANISMS E.G. BAC TERIA

4.5.1 CHRONIC TOXICITY TO FISH

4.5.2 CHRONIC TOXICITY TO AQUATIC INVERTEBRATES

Species : Daphnia magna (Crustacea)
Endpoint : reproduction rate
Exposure period : 21 day
Unit : mg/l
Analytical monitoring : yes
NOEC : = .063
LCEC : = .168
Method : OECD Guide-line 202, part 2 "Daphnia sp., Reproduction Test"
Year : 2000
GLP : yes
Test substance : other TS
Method : -Test Organisms:
a) Age: < 24 hours old
b) Supplier/Source: Test organisms were obtained from National Institute for Environmental Studies (JAPAN) and reproduced by the testing laboratory for four years and six months.
c) Any pretreatment: Parental daphnids were acclimated for at least 27 days on test conditions before testing, the mortality of the parental daphnids for two weeks before testing was 0% and any male dapnid was not born. During acclimination, test

daphnids were fed with Chlorella vulgaris, 0.2 mg carbon/day/individual.

-Test substance: Diisopropylbenzene This test substance is very sparingly soluble in water and tends to volatilize for low boiling point.
a) Empirical Formula: C12H18
b) Molecular Weight: 162.27g/mol
c) Purity: o- 1.8%, m- 58.2%, p- 39.7%

- Test Conditions:
a) Dilution Water Source: Elendt M4 medium (OECD guideline 211)
b) Exposure Vessel Type: A 80 mL test solution in a 100mL heat-resistance glass jar with glass screw cap
c) Nominal Concentrations: control, solvent control, 0.012, 0.032, 0.085, 0.230 and 0.600 mg/L
d) Stock Solutions: Diisopropylbenzene of 100mg/L was prepared for stock solution according to the following method. 50mg test substance was dissolved in 8000mg Dimethyl formamide(DMF) with 300mg HCO-40. The solution was diluted with pure water in 500mL.
e) Vehicle/Solvent and Concentrations: DMF and HCO-40 were used for solvent. DMF 20.0mg/L and 60.0mg/L HCO-40 were contained in each test solution and solvent control.
f) Number of Replicates: 10
g) Individuals per Replicates: 10
h) Renewal Rate of Test Water: Every 24 hours
i) Water Temperature: 20+/-1ºC
j) Light Condition: 16:8 hours, light-darkness
k) Feeding: 0.15 mg carbon/day/individual (Chlorella vulgaris: Green Algae)

- Analytical Procedure: The test concentrations were measured for both new and old test solution at the start of test and 1st, 7th 8th, 14th and 15th day.

- Statistical Method:
a) Data Analysis: Dunnett multiple comparison for NOEC and LOEC Probit method, binomial or Moving average for EC50
b) Method of Calculating Mean Measured Concentrations (i.e. arithmetic mean, geometric mean, etc.): Time-weighted Mean

Remark : NOEC was determined based on the cumulative number of juveniles produced per adult alive for 21 days.

Result : - Effect: reproduction- Measured Concentrations : The test concentrations were measured for both renewal and old test solution at the start of test and 1st, 7th 8th, 14th and 15th day. For some of them, the deviation from the nominal were not

less than +/-20%.

Nominal Conc. mg/L	Date	Measured Conc., mg/L 0 Fresh	0 Old	1 Fresh	7 Old	8 Fresh	14 Old	15 Fresh	TWM*1 Old mg/L	%of Nominal
Control		<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	---	---
Solvent Control		<0.002	<0.002	<0.002	<0.002	<0.002	<0.002	---	---	
0.012		0.012	0.007	0.010	0.008	0.010	0.007	0.009		75
0.032		0.028	0.019	0.026	0.022	0.029	0.023	0.024		75
0.085		0.075	0.052	0.069	0.054	0.075	0.055	0.063		74
0.230		0.199	0.143	0.193	0.143	0.190	0.146	0.168		73
0.600		0.486	0.380	0.483	0.383	*	*	0.431		72

Fresh: Start of renewal period
Old: End of renewal period
*1: Time-weighted mean of measured concentration during 21 days
*: No measured was made because all dapnids were dead at this time.

-Measured Concentration as a Percentage of Nominal

Nominal Concentration mg/L	Days	Measured Concentration as a Percentage of Nominal 0 Fresh	0 Old	1 Fresh	7 Old	8 Fresh	14 Old	15
0.012		100	58	83	67	83	58	
0.032		88	59	81	69	91	72	
0.085		88	61	81	64	88	65	
0.230		87	62	84	62	83	63	
0.600		81	63	81	64	*	*	

Fresh: Start of renewal period
Old: End of renewal period
*1: Time-weighted mean of measured concentration during 21 days
*: No measured was made because all dapnids were dead at this time.

-
- Water chemistry (pH and DO) and temperature in test:
Water chemistry and temperature were measured for control and each concentration at the start of test and before and after

renewal of the test solutions.
pH: 7.3 - 8.6 DO: 7.7 - 9.1 mg/L
Water Temperature: 20.0 - 20.5ºC

- Total hardness: 220 - 245 mg/L

-Effect Data:
LC50 (21day) = 0.220 (mc) (95 C. I.: 0.156-0.346 mg/L)
EC50 (21day) = 0.154 mg/L (mc)
NOEC (21day) = 0.063g/L (mc)
LOEC (21day) = 0.168 mg/L (mc)
mc: based on measured concentration

- Cumulative Number of Died Parental Daphnids: No test organism was killed at control and solvent control. The lowest concentration that test organisms were dead was at 0.012 mg/L after 7days. All test organisms were dead at 0.600 mg/L after 8days.

Measured Daphnids Conc.	Cumulative Number of Dead Parental (days)(mg/L)									
	1	2	3	4	5	6	7	8	9	10
Control	0	0	0	0	0	0	0	0	0	0
Solvent Control	0	0	0	0	0	0	0	0	0	0
0.009	0	0	0	0	0	0	1	1	1	1
0.024	0	0	0	0	0	0	0	0	0	1
0.063	0	0	0	0	0	0	0	0	0	0
0.168	0	0	0	0	0	1	1	1	1	1
0.431	0	0	1	9	9	9	9	10	10	10

Nominal Conc.	Cumulative Number of Dead Parental Daphnids (days)(mg/L)										
	11	12	13	14	15	16	17	18	19	20	21
Control	0	0	0	0	0	0	0	0	0	0	0
Solvent Control	0	0	0	0	0	0	0	0	0	0	0
0.009	1	1	1	1	2	2	2	2	2	2	2
0.024	1	1	1	1	1	1	1	1	1	1	2
0.063	1	1	1	1	1	1	1	1	1	1	1
0.168	1	1	1	1	1	1	1	1	1	1	1
0.431	10	10	10	10	10	10	10	10	10	10	10

-Effect Data(reproduction):Juveniels were first producted onthe 7th day control and all concentrations.

Measured Conc. (mg/L)	Mean Cumulative Numbers of Juvenieles Produced per Adult (days) 0	6	7	8	9	10	11	12	13
Control	0	0	4.1	9.6	9.6	18.9	45.0	45.1	45.1
Solvent Control	0	0	9.5	10.7	10.7	29.3	45.3	48.2	48.2
0.009	0	0	8.5	11.5	11.5	18.7	47.5	47.5	47.5
0.024	0	0	14.0	14.0	14.0	14.1	56.4	60.9	60.9
0.063	0	0	12.0	12.0	12.0	11.9	49.0	53.7	53.7
0.168	0	0	1.7	3.0	4.0	1.5	17.0	17.0	17.0
0.431	0	0	---	---	---	---	---	---	---

Measured Conc. (mg/L)	Mean Cumulative Numbers of Juvenieles Produced per Adult (days) 14	15	16	17	18	19	20	21
Control	78.6	78.6	78.6	101.6	106.6	106.6	110.0	130.5
Solvent Control	83.2	83.2	83.2	114.8	115.1	115.1	122.7	140.3
0.009	83.4	83.4	83.4	115.6	115.6	115.6	119.8	139.4
0.024	92.4	92.4	92.4	121.9	122.0	122.0	127.8	144.5
0.063	86.1	86.1	89.9	120.9	120.9	120.9	142.7	145.1
0.168	18.7	20.3	26.9	40.1	42.4	42.4	44.6	46.7
0.431	---	---	---	---	---	---	---	---

: The parental Daphnids were dead during a 21-day testing period

-Cumulative numbers of juveniles produced per adult alive for 21 days

Vessel No.	Cont.**	Solv***	Nominal Conc., mg/L (Measured Conc.*1, mg/L) 0.012 (0.009)	0.032 (0.024)	0.085 (0.063)	0.230 (0.168)	0.600 (0.431)
1	152	135	138	D	151	45	D
2	133	152	149	D	145	11	D
3	129	131	152	144	D	67	D
4	110	152	123	149	145	D	D
5	148	135	151	140	149	37	D
6	135	133	128	139	146	28	D
7	120	154	D	137	125	70	D
8	132	135	129	149	151	54	D
9	116	140	145	151	154	37	D

10	130	136	D	147	140	71	D
Mean	130.5	140.3	139.4	144.5	145.1	46.7	0.0
S. D.	13.1	8.8	11.5	5.3	8.6	20.7	
Inhibition rate(%)		0.7	-3.0	-3.4	66.7	0.0	
Significant difference		---	---	---	**	++	

*1: Time-wighted mean measured concentration
**: Indicates a significant difference (alpha = 0.01) from the solvent control.
++: Statistical comparison test could not be performed for this concentration because adult alive after 21 days was none.
D: Were not included for calculation because the parental daphnids were dead during a 21-day testing period.
---: Indicates no significant difference.

- Calculation of toxicity values: The calculation oftoxicity values was the mean measured concentrations. The reason is that some of the error ranges of measured concentration were not less than +/-20% of nominal concentration.

Source : MOE,Japan(2000)
Reliability : (2) valid with restrictions
Remarks: This test was conducted using solvent(mxture of a dispersant of HCO40 and DMF), however an analytical monitoring was dane. In this test, it could not seperate the true toxicity of the test substance from the physical effect of non-disolving substance
Flag : Critical study for SIDS endpoint
20.12.2002 (9)

4.6.1 TOXICITY TO SOIL DWELLING ORGANISMS

4.6.2 TOXICITY TO TERRESTRIAL PLANTS

4.6.3 TOXICITY TO OTHER NON-MAMM. TERRESTRIAL SPECIES

4.7 BIOLOGICAL EFFECTS MONITORING

4.8 BIOTRANSFORMATION AND KINETICS

4.9 ADDITIONAL REMARKS

5.1.1 ACUTE ORAL TOXICITY

Type : LD50
Species : rat
Strain : no data
Sex : no data
Number of animals :
Vehicle : no data
Value : = 6.5 ml/kg bw (5850 mg/kg bw)
Method : other: no data
Year :
GLP : no data
Test substance : no data
Source : Charles, P.C.et al.: 1974
19.11.2002 (1)

Type : LD50
Species : Rat
Strain : no data
Sex : no data
Number of animals :
Vehicle : no data
Value : = 7400 mg/kg bw
Method : other: no data
Year :
GLP : no data
Test substance : other TS: m-diisopropylbenzene
Source : Izmerov, N.F. et al.: 1982
20.11.2002 (4)

Type : LD50
Species : mouse
Strain : no data
Sex : no data
Number of animals :
Vehicle : no data
Value : = 3100 mg/kg bw
Method : other: no data
Year :
GLP : no data
Test substance : other TS: m-diisopropylbenzene
Source : Izmerov, N.F. et al.: 1982
20.11.2002 (4)

Type : LD50
Species : mouse
Strain : no data
Sex : no data

Number of animals :
Vehicle : no data
Value : = 3400 mg/kg bw
Method : other: no data
Year :
GLP : no data
Test substance : other TS: p-diisopropylbenzene
Source : Izmerov, N.F. et al.: 1982
20.11.2002 (4)

5.1.2 ACUTE INHALATION TOXICITY

Type : LCLo (Lethal concentration, low)
Species : rat
Strain : no data
Sex : no data
Number of animals :
Vehicle : no data
Exposure time : 4 hour(s)
Value : = 5300 mg/m³
Method : other: no data
Year :
GLP : no data
Test substance : no data
Source : Izmerov, N.F. et al.: 1982
04.07.2002 (4)

Type : LCLo (Lethal concentration, low)
Species : mouse
Strain : no data
Sex : no data
Number of animals :
Vehicle :
Exposure time : 2 hour(s)
Value : = 5300 mg/m³
Method : other: no data
Year :
GLP : no data
Test substance : no data
Source : Izmerov, N.F. et al.: 1982
04.07.2002 (4)

5.1.3 ACUTE DERMAL TOXICITY

Type : LD50
Species : rabbit
Strain : no data

Sex	:	no data
Number of animals	:	
Vehicle	:	
Value	:	= 16 ml/kg bw (14400 mg/kg bw)
Method	:	other: no data
Year	:	
GLP	:	no data
Test substance	:	no data
Source	:	Charles, P.C.et al.: 1974

05.09.2003 (1)

5.1.4 ACUTE TOXICITY, OTHER ROUTES

5.2.1 SKIN IRRITATION

Species	:	rabbit
Concentration	:	100 other: mg
Exposure	:	no data
Exposure time	:	24 hour(s)
Number of animals	:	
PDII	:	
Result	:	moderately irritating
EC classification	:	
Method	:	other: no data
Year	:	
GLP	:	no data
Test substance	:	no data
Source	:	Marhold, J.: 1986

24.06.2002 (5)

5.2.2 EYE IRRITATION

Species	:	rabbit
Concentration	:	
Dose	:	500 other: mg
Exposure Time	:	24 hour(s)
Comment	:	
Number of animals	:	
Result	:	
EC classification	:	
Method	:	other: no data
Year	:	
GLP	:	no data
Test substance	:	no data
Result	:	mildly irritating
Source	:	Marhold, J. : 1986

04.07.2002 (5)

5.3 SENSITIZATION

5.4 REPEATED DOSE TOXICITY

Species	:	rat
Sex	:	male/female
Strain	:	Crj: CD(SD)
Route of admin.	:	gavage
Exposure period	:	Males and females, 28 days
Frequency of treatment	:	Once daily
Post obs. period	:	14 days
Doses	:	Dosage : 0(vehicle), 6, 30, 150, 750 mg/kg/day
Control group	:	yes, concurrent vehicle
Method	:	other: Guidelines for 28-day Repeat Dose Toxicity Testing of Chemicals (Japan), Equivalent to OECD TG 407
Year	:	1998
GLP	:	yes
Test substance	:	other TS: Source; Wako Pure Chemical Ind., Ltd., Lot No. CAF5267, Purity; 98.0%(m-57.042%, p-40.951%)
Result	:	NOAEL: 30 mg/kg/day for both males and females

1. Clinical signs
No death was observed in any group, and there was no change in clinical signs throughout the observation period in the control group and the 6 and 30mg/kg groups.
In the 150 and 750 mg/kg groups, mydriasis was observed. It was observed on and after Day 8 of administration in males in the 150mg/kg group and in m ales and females in the 750mg/kg group and on and after Day 14 of administration in females in the 150mg/kg group. In total, it was observed in 2 males and females each in the 150mg/kg group and 10 males and all females in the 750mg/kg group. It appeared from about 2 and a half hours to 3 and a half hours after administration and disappeared within 2 and a half hours after the onset. Males and females in the 750mg/kg group showed no change during the recovery period.

2. Body weight
Throughout the administration period or the recovery period, the groups treated with diisopropylbenzene showed the change almost similar to the control group.

3. Food consumption
Throughout the administration period or the recovery period,

the groups treated with diisopropylbenzene showed the change almost similar to the control group.

4. Urinalysis
In the examination in Week 4 of administration, increased urine volume was observed in females in the 750mg/kg group, and the similar tendency was observed in females in the 150mg/kg group, but it was the change within physiological variation.
In the examination in Week 2 of recovery, no change was observed in males and females in the 750mg/kg group.

5. Hematology
In the examinations at the end of the administration period and the recovery period, there was no change in the groups treated with diisopropylbenzene.

6. Blood biochemistry
In the examination at the end of the administration priod, increased potassium and decreased chloride were observed in males in the 750mg/kg group, and increases in total protein, total cholesterol and phospholipid and a decrease in chloride were observed in females of the same group. Additionally, increased chloride and increased glucose were observed in males in the 6mg/kg group and females in the 150mg/kg group, respectively. The changes were not considered to be due to the administration of the test compound because there was no dose response relationship.
In the examination at the end of the recovery period, there were increased gamma-GTP and decreased acetylcholinesterase
in males in the 750mg/kg group and decreased total bilirubin in females of the same group. They were considered the changes not related to the administration of the test compound because they were minimal charges and were not observed at the end of the administration period.

7. Autopsy
In the examination at the end of the administration period, grayish white patches in the lung were observed in one female in the control group. In the examination at the end of the recovery period, no change was observed in both the control group and the 750mg/kg group.

8. Organ weight
In the examination at the end of the administration period, increases in absolute and relative liver weights were observed in males and females in the 750mg/kg group. And increases in absolute and relative kidney weights and a decrease in absolute spleen weight were observed in males of

the same group. In the examination at the end of the recovery period, an increase in relative liver weight was observed in females in the 750mg/kg group, but the degree of change was reduced compared with that at the end of the administration period. Additionally, an increase in relative lung weight was observed in males in the 750mg/kg group, but it was considered to be the change not related to the administration of the test compound because it was not observed at the end of the administration period.

Organ weights of rats treated orally in the 28-day repeat dose toxicity dose

Dose level (mg/kg)	0	6	30	150	750
Male					
Absolute organ weight					
Liver	10.89±1.24	10.29±0.67	10.91±1.72	11.06±1.55	13.15±0.65*
Spleen	0.73±0.02	0.64±0.11	0.72±0.09	0.69±0.11	0.62±0.08*
Kidneys	2.52±0.25	2.47±0.15	2.64±0.15	2.49±0.17	2.81±0.10*
Relative organ weight					
Liver	3.01±0.18	3.02±0.18	2.99±0.22	3.13±0.29	3.64±0.14**
Spleen	0.70±0.06	0.72±0.04	0.73±0.04	0.71±0.05	0.78±0.05*
Female					
Absolute organ weight					
Liver	6.75±0.65	6.77±0.80	6.79±0.55	7.20±0.37	8.16±0.69**
Relative organ weight					
Liver	3.12±0.16	3.14±0.22	3.05±0.10	3.23±0.17	3.71±0.23**

*: significant difference from control, $p<0.05$
**: significant difference from control, $p<0.01$
Values are mean±S.D.

9. Histopathology
At autopsy at the end of the administration period, centrilobular hypertrophy of hepatocytes was observed in one male in the 150mg/kg group and one male and 4 females in the 750mg/kg group, and there was a significant difference between the control and the 750mg/kg group. In the kidneys, eosinophilic bodies of the proximal tubular epithelium were observed in one male each in the 6, 30 and 150 mg/kg groups and all males in the 750mg/kg group, and there was a significant difference between the control group and the 750mg/kg group. This kidney change was considered to be due to
accumulation of the complex with alpha2u-globulin, a male rat specific protein. Additionally, accumulation of foam cells was observed at the site macroscopically showing grayish white patches in the lung in one male in the control group. No change was observed in the heart, spleen and adrenal gland examined.

In the animals autopsied at the end of the recovery period, eosinophilic bodies of the proximal tubular epithelium in the kidney were observed in one male in the 750mg/kg group. But the similar change was observed in one male in the control group, and there was no difference between the control and the 750mg/kg group. Additionally, granular casts and an eosinophilic change of the proximal tubular epithelium were also observed in one male showing eosinophilic bodies in the proximal tubular epithelium in the 750mg/kg group. But they were the changes related to regeneration of the renal tubule, and recovery was observed at the end of the administration period. Furthermore, there was no change in the liver showing some changes at the end of the administration period.

Histpatological findings of rats treated orally in the 28-day repeat dose toxicity test

Dose level (mg/kg)	0	6	30	150	750
Male					
No. of animal	6	6	6	6	6
Liver					
Hypertrophy, hepatocyte, centribular					
-	6		6	5	5
+	0		0	1	1
++	0		0	0	0
+++	0		0	0	0
Kidney					
Eosinophilic body, proximal tubule					
-	6	5	5	5	0
+	0	1	1	1	5
++	0	0	0	0	1
+++	0	0	0	0	0**
Female					
No. of animal	6	6	6	6	6
Liver					
Hypertrophy, hepatocyte, centribular					
-	6		6	6	2
+	0		0	0	4
++	0		0	0	0
+++	0		0	0	0*

Grade sign: -, none; +, mild; ++, moderate; +++, marked
*: significant difference from control, p<0.05
**: significant difference from control, p<0.01

Source : MHW, Japan (1998)

Test condition : - No. of animals per sex per dose: 0 and 750 mg/kg b.w. group: 12 (6 for scheduled sacrifice group and 6 for recovery maintenance group) 6, 30 and 150 mg/kg b.w. group: 6 (only scheduled sacrifice)

- Vehicle : Corn oil
- Terminal kill : Days 29 or 43
- Organs examined at autopsy:
*Organ weight: brain, heart, lungs, thymus, liver, spleen, kidneys, adrenals,testes, ovary
*Microscopic: the control and the highest dose group: heart, liver, spleen, kidneys, adrenals and any organs which have gross pathological changes.
Other groups: liver and kidneys, which are shown to have histopathological changes at the higher dose. Alpha2u-globulin was not stained.

Conclusion : The kidney change observed in males of all dose groups was considered due to accumulation of the complex with alpha2u-globulin, a male rat specific protein.
The NOAEL was concluded to be 30 mg/kg b.w./day for both males and females because mydriasis observed at 150 mg/kg b.w. was considered dose-related adverse effect.

Reliability : (1) valid without restriction
Well conducted study, carried out by Panapharm Laboratories Co., Ltd.

Flag : Critical study for SIDS endpoint

05.09.2003 (7)

Species : rat
Sex : male/female
Strain : Crj: CD(SD)
Route of admin. : gavage
Exposure period : Males: 50-52 days (from 14 days before mating)
Females: 41-46 days (from 14 days before mating to day 3 of lactation)
Frequency of treatment : Once daily
Post obs. period : 1 day
Doses : Dosage : 0(vehicle), 6, 30, 150, 750 mg/kg/day
Control group : yes, concurrent vehicle
Method : other: OECD TG 421
Year : 2001
GLP : yes
Test substance : other TS: Source; Wako Pure Chemical Ind., Ltd., Lot No. PAK5633, Purity; 98.2%(m-58.5366%, p-39.7107%)
Result : 1) Effects on males
(1) Clinical signs
Neither dead nor moribund animal was observed in any group. In clinical observation, there was no abnormality in the control group and the 6, 30 and 150mg/kg groups. In the 750mg/kg group, transient salivation was observed after administration on and after Day 6 of administration. In the 750mg/kg group, exophthalmos was observed in two animals on and after Day 9 of administration.

(2) Change in body weight
There was no significant difference in the body weight on any day of determination in any treatment group compared with the control group.

(3) Food consumption
In the 6, 30 and 150mg/kg groups, there was no significant difference in the food consumption on any day of determination compared with the control group. In the 750mg/kg group, a significantly low value of food consumption was observed on Day 3 of administration compared with the control group.

(4) Autopsy
In the control group and the 30 and 150mg/kg groups, no abnormality was observed. In the 6mg/kg group, there were atrophied bilateral testes and atrophied bilateral epididymides in one animal and softening of the left testis and the left epididymis in one animal. In the 750mg/kg group, bilateral exophthalmos was observed in two animals.

(5) Organ weight
In any treatment group, there was no significant difference in the body weight on the day of necropsy compared with the control group. In any treatment group, there were no significant differences in the absolute and relative weights in any organ compared with the control group.

(6) Sperm test
In any treatment group, there were no significant differences in the rate of active sperms, the rate of transfer from the standard point, the rate of transfer of the shortest distance, the total transfer rate, the number of crossings of the head, the rate of malformed sperms, the rate of live sperms, the rate of surviving sperms, the number of sperms and the number of sperms per 1g of the tail of left epididymis compared with the control group. In one animal showing atrophy in bilateral testes and atrophy in bilateral epididymides in the 6mg/kg group , since the rate of active sperms was 0%, none of the morphology of sperm, the rate of live sperms and the rate of surviving sperms could be calculated, and the number of sperms and the number of sperms per 1g of the tail of left epididymis were also small. Additionally, in one animal showing softening of the left testis and the left epididymis in the 6mg/kg group, the number of sperms and the number of sperms per 1g of the tail of left epididymis were small. Since none of these changes depended on the dose, however, they were considered incidental changes.

(7) Histopathology
Testis: Atrophy in seminiferous tubules were observed in one animal in the control group, two animals in the 6 mg/kg group and one animal in the 750 mg/kg group. Hyperplasia of Leydig cells was observed in one animal in the control group and two animals in the 6mg/kg group. Vacuolation of Sertoli's cell was observed in one animal in the controlgroup. Since these changes were observed in a few animals, they were considered accidental changes.
Eyeballs: Vacuolation of the lens fiber was observed in two animals in the 750mg/kg group. In one animal showing the above vacuolation of the lens fiber, hyperplasia of the lens epithelium was also observed.
Epididymis (head): There was no abnormality in both the control group and the 750mg/kg group.

2) Effects on females
(1) Clinical signs
Death was observed in one animal (Day 23 of pregnancy) in the control group and one animal (Day 3 of lactation) in the 150mg/kg group.
In clinical observation of dead animals, there was no abnormality in the control group. In the 150mg/kg group, subnormal temperature, decreased spontaneous movement and staggering gait were observed on Day 3 of lactation.
In clinical observation in surviving animals, there was no abnormality in the control group and the 6, 30 and 150 mg/kg groups. In the 750mg/kg group, transient salivation was observed after administration on and after Day 6 of administration. In the 750mg/kg group, additionally, mydriasis was observed in one animal from Day 15 to Day 17 of administration.

(2) Change in body weight
During the pre-mating period, there was no significant difference in the body weight on any day of determination in any treatment group compared with the control group.
During the pregnancy period, there was no significant difference in the body weight on any day of determination in the 6, 30 and 150 mg/kg groups compared with the control group. In the 750mg/kg group, a significantly low value of body weight was observed on Day 14 of gestation compared with the control group.
During lactation period, there was no significant difference in the body weight on any day of determination in any treatment group compared with the control group.

(3) Food consumption
During the pre-mating period, there was no significant difference in the food consumption on any day of

determination in the 6, 30 and 150 mg/kg groups compared with the control group. In the 750mg/kg group, a significant low value of food consumption was observed on Day 3 of administration compared with the control group.
During the pregnancy period, there was no significant difference in the food consumption on any day of determination in the 6, 30 and 750 mg/kg groups compared with the control group. In the 150mg/kg group, a significantly low value of food consumption was observed on Day 21 of pregnancy compared with the control group, but it was not a change depending on the dose.
During the lactation period, there was no significant difference in food consumption in the 6, 30 and 150 mg/kg groups compared with the control group. In the 750mg/kg group, a significantly high value of food consumption was observed on Day 4 of lactation compared with the control group.

(4) Autopsy
In the surviving animals, there was no abnormality in any group.
In dead animals, dark red patches on the glandular gastric mucosa were observed in one animal in the control group. In one animal in the 150 mg/kg group, however, no abnormality was observed.

(5) Organ weight
In the body weight on the day of autopsy, there was no significant difference in any treatment group compared with the control group. In any treatment group, there were no significant differences in the absolute and relative ovary weights compared with the control group.

(6) Histopathology
Ovaries: In both of the control group and the 750mg/kg group, no abnormality was observed.

Source : MHLW, Japan (2001)

Test condition :
- Number of animals/group : Males, 12; Females, 12
- Vehicle : Corn oil
- Terminal kill : Males, days 51-53; Females, day 4 of lactation
- Hematological and blood biochemical examination : not conducted
- organ weight measurement and histopathological examination : only for reproductive organs and organs with abnormal findings at autopsy, alpha2u-globulin was not stained.

Conclusion : The NOAEL could not be estimated because the examined items were insufficient, such as no histopathological examination on liver and kidneys.

Reliability : (1) valid without restriction

Well conducted study, carried out by Nihon Bioresearch Inc..
20.11.2002 (6)

5.5 GENETIC TOXICITY 'IN VITRO'

Type	:	Bacterial reverse mutation assay
System of testing	:	Salmonella typhimurium TA100, TA1535, TA98, TA1537, Escherichia coli WP2 uvrA
Concentration	:	See test condition
Cycotoxic conc.	:	See result
Metabolic activation	:	with and without
Result	:	negative
Method	:	other: Guidelines for Screening Toxicity Testing of Chemicals (Japan) and OECD Guidelines No. 471
Year	:	1998
GLP	:	yes
Test substance	:	other TS: Source; Wako Pure Chemical Ind., Ltd., Lot No. CAF5267, Purity; 98.0%(m-57.042%, p-40.951%)
Remark	:	Dose-finding test was performed at 3 levels in the range of 50 to 5,000 ug/plate. Cytotoxicity was observed at 5,000 ug/plate (WP2 uvrA) and at all doses (other strains) with S9 mix, and at 500 ug/plate and more (WP2 uvrA) and at 150 ug/plate and more (other strains) without S9 mix. From these results, maximum doses establishied were 5,000 ug/plate for WP2 uvrA and 50 ug/plate for other strains with S9 mix, and 625 ug/plate for WP2 uvrA and 200 ug/plate for other strains without S9 mix. Statistical methods: no data
Result	:	This chemical was not mutagenic in Salmonella typhimurium TA100, TA1535, TA98, TA1537 and Escherichia coli WP2 uvrA, with or without an exogenous metabolic activation system. Toxicity was observed at 6.25 ug/plate and more (TA1535, TA1537) and 12.5 ug/plate and more (TA100, TA98), 5000 ug/plate (WP2 uvrA) without an S9 mix and at 100 ug/plate and more (TA100, TA1535, TA98,TA1537) and 625 ug/plate (WP2 uvrA) with an S9 mix.
Source	:	MHW, Japan (1998)
Test condition	:	1)Procedures : Pre-incubation method 2)Solvent : DMSO 3)Positive controls : -S9 mix; 2-(2-Furyl)-3-(5-nitro-2-furyl)acrylamide(TA100, TA98, WP2 uvrA), Sodium azide(TA1535) and 9-Aminoacridine(TA1537) +S9 mix; 2-Aminoanthracene(five strains) 4)Doses : -S9 mix; 0, 0.195, 0.391, 0.781, 1.56, 3.13, 6.25

ug/plate (TA1537); 0, 0.781 - 50.0 ug/plate (TA100, TA1535, TA98 (Test 1)); 0, 0.391 - 12.5 ug/plate(TA1535(Test 2)); 0, 0.781 - 25.0 ug/plate (TA100, TA98(Test 2)); 0, 156 - 5000 ug/plate(WP2 uvrA)
+S9 mix; 0, 6.25 - 200 ug/plate(TA100, TA1535, TA98, TA1537); 0, 19.5 - 625 ug/plate(WP2 uvrA)
5)S9 : Rat liver, induced with phenobarbital and 5,6-benzoflavone
6)Plates/test : 3
7)Number of replicates : 2

Reliability : (1) valid without restriction
Well conducted study, carried out by Hatano Research Institute, Food and Drug Safety Center

Flag : Critical study for SIDS endpoint

20.11.2002 (7)

Type : Chromosomal aberration test
System of testing : Type of cell used: Chinese hamster lung (CHL/IU)cells
Concentration : See remark
Cycotoxic conc. : See remark.
Metabolic activation : with and without
Result : negative
Method : other: Test method: Guidelines for Screening Mutagenicity Testing of Chemicals(Japan)and OECD Guideline No. 473
Year : 1998
GLP : yes
Test substance : other TS: Source; Wako Pure Chemical Ind., Ltd., Lot No. CAF5267, Purity; 98.0%(m-57.042%, p-40.951%)
Remark : The concentrations set up in this test were as follows.
These concentrations were established based on the result of growth inhibition test.
-S9 mix (continuous treatment): 0, 0.0038, 0.0075, 0.015, 0.030*, 0.060* mg/mL
-S9 mix (short-term treatment): 0, 0.0019, 0.0038, 0.0075, 0.015*, 0.030*, 0.060*, 0.12* mg/mL
+S9 mix (short-term treatment): 0, 0.030, 0.060, 0.12 mg/mL

Above asterisks* show that chromosome analysis was not performed because of the severe cytotoxicity.
Short treatment: 6 h
Continuous treatment: 24, 48 h
Statistical methods: Fisher's exact test or Cochran-Armitage's test
Result : This chemical did not induce structual chromosomal aberration or polyploidy under the conditions of this experiment.
Source : MHW, Jaoan (1998)
Test condition : 1)Solvent: Acetone
2)Positive controls:

-S9 mix, Mitomycin C
+S9 mix, Cyclophosphamide
3)S9: Rat liver, induced with phenobarbital and 5,6-benzoflavone
4)Plates/test: 2

Reliability : (1) valid without restriction
Well conducted study, carried out by Hatano Research Institute, Food and Drug Safety Center

Flag : Critical study for SIDS endpoint

19.11.2002 (7)

5.6 GENETIC TOXICITY 'IN VIVO'

5.7 CARCINOGENITY

5.8 TOXICITY TO REPRODUCTION

Type	:	One generation study
Species	:	rat
Sex	:	male/female
Strain	:	Crj: CD(SD)
Route of admin.	:	gavage
Exposure period	:	Males: 50-52 days Females: from 14 days before mating to day 3 of lactation
Frequency of treatment	:	Once daily
Premating exposure period		
Male	:	14 days
Female	:	14 days
Duration of test	:	
Doses	:	Dosage : 0(vehicle), 6, 30, 150, 750 mg/kg/day
Control group	:	yes, concurrent vehicle
Method	:	other: OECD TG 421
Year	:	2001
GLP	:	yes
Test substance	:	other TS: Source; Wako Pure Chemical Ind., Ltd., Lot No. PAK5633, Purity; 98.2%(m-58.5366%, p-39.7107%)
Result	:	NOAEL: Parental; 750 mg/kg for male and female Offspring; 750 mg/kg for male and female

1) Effects on reproduction and development of parent animals
(1) Number of estruses, copulation index and fertility index
The number of estruses during the administration period (14 days) before mating showed no significant difference in any treatment group compared with the control group.

The number of days required for copulation showed no significant difference between any treatment group and the control group.
The pair showing no copulation was observed in one pair each in the 6 and 30 mg/kg groups. However, there was no significant difference in the copulation index between any treatment group and the control group.
There were two females showing no conception in the 6mg/kg group. However, there was no significant difference in the fertility index between any treatment group and the control group.

(2) Pregnancy period, parturition, number of corpora lutea, implantation index and delivery index
The pregnancy period showed no significant difference in any treatment group compared with the control group. In the control group and the 30, 150 and 750mg/kg groups, there was
no abnormality in the parturition in any dam. In one animal in the 6 mg/kg group, since the born offspring died (one stillbirth was confirmed), no neonate could be obtained.
In any treatment group, there were no significant differences in the number of corpora lutea, the number of implantations and the implantation index compared with the control group.
In the control group and the 30, 150 and 750 mg/kg groups, the delivery index was 100%. In the 6mg/kg group, since 1 dam could obtain no neonate, the delivery index was 88.9%, but there was no significant difference from the control group. In the control group and any treatment group, there was no abnormality in the lactation.

2) Effects on offspring
(1) Parturition index and live birth index
In any treatment group, there were no significant differences in the total number of delivered offspring, the number of stillbirth, the number of neonates on Day 0 of lactation, the sex ratio, the parturition index, the delivery index of offspring and the live birth index compared with the control group.

(2) Clinical signs and viability index of offspring
In any treatment group, there were no significant differences in the number of offspring surviving on Day 4 of lactation and the viability index of neonates on Day 4 compared with the control group. In the external observation of neonates, there was no abnormality in any group. In the clinical signs of offspring, there was no abnormality in any group.

(3) Change in body weight of offspring
In any treatment group, there were no significant differences in the body weights on Days 0 and 4 of lactation for males and females compared with the control group.

(4) Autopsy of offspring
In any group, no abnormality was observed.

Source : MHLW, Japan (2001)
Test condition : - Number of animals/group : Males, 12; Females, 12
- Vehicle : Corn oil
- Terminal kill : Males, days 51-53; Females, day 4 of lactation
Conclusion : The reproductive and developmental toxicological NOAEL is considered to be 750 mg/kg/day since there was no effect on male and female parent animals and offspring after administration at a dose of 750 mg/kg.
Reliability : (1) valid without restriction
Well conducted study, carried out by Nihon Bioresearch Inc..
Flag : Critical study for SIDS endpoint
20.11.2002 (6)

5.9 DEVELOPMENTAL TOXICITY/TERATOGENICITY

5.10 OTHER RELEVANT INFORMATION

5.11 EXPERIENCE WITH HUMAN EXPOSURE

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