FOREWORD **INTRODUCITON**

ε-CAPROLACTONE

CAS N°: 502-44-3

SIDS Initial Assessment Report

For

SIAM 19

Berlin, Germany, 19-22 October 2004

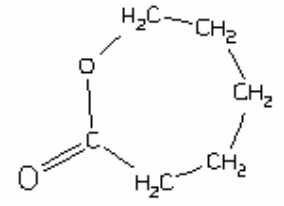
1. Chemical Name:	ε-Caprolactone
2. CAS Number:	502-44-3
3. Sponsor Country:	Belgium Dr. T. Lakhanisky Ministry of Social Affairs, Public Health and Environment Scientific Institute of Public Health – Division Toxicology Rue J. Wytsman 16, B-1050 Brussels Tel. + 32 2 642 5104, fax. + 32 2 642 5224
4. Shared Partnership with:	e-Caprolactone consortium
5. Roles/Responsibilities of the Partners:	
• Name of industry sponsor /consortium	Members of the e-caprolactone consortium are BASF Corporation, Daicel Chemicals, Solvay SA (leader) and The DOW Chemical Company. Contact point: A.G. Berends Solvay S.A., CC/Health, Safety and Environment Rue de Ransbeek 310, B-1120 Brussels Tel. + 32 2 2643398, fax. + 32 2 2642990
• Process used	Industry did the literature search, collected all references, did additional non-vertebrate tests and prepared the dossier.
6. Sponsorship History	
• How was the chemical or category brought into the SIDS Program?	The substance is an ICCA HPV chemical. The sponsor country was contacted by the industry. The substance has never been part of another international assessment program.
7. Review Process Prior to the SIAM:	Not applicable.
8. Quality check process:	The Human Health part and related issues of the dossier were reviewed by experts at the Scientific Institute of Public Health; Division of Toxicology. The Environment related issues were reviewed by members of the National Health Council in collaboration with the Federal Public Service Health, Food Chain Safety and Environment; DG Environment; Risk Management.

9. Date of Submission: 23 July 2004

10. Date of last Update: 13 June 2005

11. Comments:

SIDS INITIAL ASSESSMENT PROFILE

CAS No.	502-44-3
Chemical Name	ε-Caprolactone
Structural Formula	

SUMMARY CONCLUSIONS OF THE SIAR

Human Health

After absorption of ε-caprolactone, the substance will be hydrolyzed rapidly in stomach and blood resulting in the formation of 6-hydroxyhexanoic acid. This hydrolysis product is water soluble and expected to be distributed throughout the body and excreted rapidly, principally through the urine.

ε-Caprolactone exhibits low acute toxicity by all potentially relevant routes of exposure. The acute oral LD_{50} for rats was 4290 mg/kg, while the acute dermal LD_{50} in rabbits was 6400 mg/kg body weight. The primary symptoms following a single high exposure are skin erythema (dermal) as well as apathy and effects on motor coordination and respiration (oral). ε-Caprolactone is considered not-irritating to skin and irritating to eyes.

In a 9-day inhalation study in which ε-caprolactone was administered at a concentration of 45 ppm (213 mg/m^3), no treatment-related effects were found. Therefore the 45 ppm level can be considered a NOAEL. A 90-day inhalation study with ε-caprolactone at concentrations of 15 ppm (71 mg/m^3) and 45 ppm (213 mg/m^3) resulted in perinasal and periocular encrustation and eyelid swelling in the males of the 45 ppm group. As no other treatment related effects were found, this level is considered the lowest observed adverse effect level (LOAEL). The 15 ppm level is considered the NOAEL. ε-Caprolactone given by drinking water to rats at levels of 500, 2000 and 5000 ppm in a 14-day study did not result in any treatment-related clinical signs of toxicity, clinical pathology findings, organ weight changes, necropsy observations or histopathological findings. ε-Caprolactone affected food and water consumption (low palatability) as well as body weight gain at the level of 5000 ppm only. The NOAEL was 2000 ppm, which is equivalent with a dose 152 and 184 mg/kg bw for males and females, respectively.

Bacterial and mammalian *in vitro* mutagenicity tests gave in general negative results. *In vivo*, ε-caprolactone was negative in the mouse micronucleus assay.

No studies are available with regard to reproduction and developmental toxicity of ε-caprolactone. However, a well conducted 90-day inhalation repeated dose study showed no macroscopic and histopathological changes on reproductive organs and also other systemic effects were not found during this study. The lack of systemic effects can be explained by the rapid hydrolysis in stomach and blood, resulting in the formation of 6-hydroxyhexanoic acid. Analogues of 6-hydroxyhexanoic acid show no evidence of reproductive or developmental toxicity. For this reason there is no indication for a reprotoxic concern. This is supported by the toxicological profile of structurally similar lactones, where also no organ specific toxicity was observed in long term studies (with up to 2-year exposure).

Environment

ε-Caprolactone is a colourless liquid with a melting point of -1.3 °C and a boiling point of 237 °C. The substance is miscible with water in all proportions and the calculated log Kow (octanol-water partition

coefficient) is 0.68. The vapour pressure of ε-caprolactone is 0.81 Pa.

Based on the Mackay model (level III) ε-caprolactone is expected to partition almost exclusively to the aquatic compartment (> 99.9 %). In water ε-caprolactone is hydrolysed to 6-hydroxyhexanoic acid. At 20 degrees Celsius the half-life at pH values of 4, 7 and 9 was 16, 53 and 2.2 days, respectively. 6-Hydroxyhexanoic acid (CAS 1191-25-9) is not listed on the European Inventory of Existing Commercial Substances (EINECS). Ecotoxicity data of this hydrolysis product were not found. However, based on structural comparison the ecotoxicological properties of this substance are expected to be similar to hexanoic acid and adipic acid. ε-Caprolactone is readily biodegradable according to an OECD 301 B guideline study. It is anticipated that ε-caprolactone will not bioaccumulate based on its low octanol-water partition coefficient and rapid degradation in the environment.

Aquatic ecotoxicity tests, which were done according to GLP and standard guidelines, are available for 4 different species encompassing the 3 trophic levels and microorganisms. A 72 hour toxicity test with algae (*Scenedesmus subspicatus*) revealed an EC_{50} and NOEC value of 1217 and 256 mg/l, respectively (based on biomass). Based on the specific growth rate (μ), the EC_{50} (72 h) was calculated to be 2616 mg/l. Water fleas (*Daphnia magna*) appeared to be more sensitive than algae. An acute test with an exposure period of 48 hours resulted in EC_{50} and NOEC values of 204 and 124 mg/l, respectively. For guppy (*Poecilia reticulata*) a steep concentration-response relatonship was observed. A toxicity test with a duration of 96 hours with this fish species revealed an LC_{50} and NOEC value of 295 and 250 mg/l, respectively. However for bacteria (*Pseudomonas putida*) a large difference between the EC_{50} (1260 mg/l) and NOEC (32 mg/l) was found. During this test the bacteria were exposed for 16 hours. Neither chronic aquatic toxicity tests nor terrestrial toxicity tests are available for ε-caprolactone.

Exposure

In 2003, the estimated world-wide production of ε-caprolactone was 40,000 – 60,000 tonnes. In recent years the world-wide production was growing slowly (<5 % per year). The production occurs at four sites located in the USA (two sites), Japan and the United Kingdom.

About 50 % of the quantity produced is used on site for the production of polymers (polycaprolactones). The remaining 50 % is sold to customers (downstream users). The total number of downstream users is less than 1000. ε-Caprolactone is used by downstream users to modify resins and polymers in order to enhance the performance of the end-products. The majority is used for the modification of acrylic resins and polyesters, but it is also used for modification of epoxy resins and polyurethanes. A small quantity of ε-caprolactone (< 1 %) is used as reactive diluent and as a solvent (e.g. for vinyl resins).

During production and processing, inhalation of vapours and direct skin contact are potentially relevant exposure scenarios. However, inhalation exposures to vapours of ε-caprolactone at ambient temperature are likely to be limited due to its low volatility. Based on the information available to the consortium members, ε-caprolactone is not used in consumer products.

Releases into the environment may occur during production and processing of ε-caprolactone. A release of ε-caprolactone to the environment could potentially also occur via use and disposal of polycaprolactone because polycaprolactone may be (bio)degraded to ε-caprolactone, 6-hydroxyhexanoic acid and oligomers.

RECOMMENDATION AND RATIONALE FOR THE RECOMMENDATION AND NATURE OF FURTHER WORK RECOMMENDED

Human Health: The chemical possesses properties indicating a hazard for human health (eye irritation). This hazard does not warrant further work as it relates to reversible effects. This should nevertheless be noted by chemical safety professionals and users.

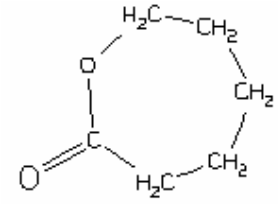
Environment: The chemical is currently of low priority for further work because of its low hazard potential.

SIDS Initial Assessment Report

1 IDENTITY

1.1 Identification of the Substance

CAS Number: 502-44-3
IUPAC Name: Hexano-6-lactone
Molecular Formula: $C_6H_{10}O_2$
Structural Formula:



Molecular Weight: 114
Synonyms: 1,6-Hexanolide; 2-oxepanone; 6-hexanolactone; 6-hydroxyhexanoic acid lactone; caprolactone; ε-caprolactone; epsilon-caprolactone; hexan-6-olide; hexanoic acid, epsilon-lactone

In this dossier the synonym ε-caprolactone will be used as this is the name that is commonly used.

1.2 Purity/Impurities/Additives

The purity of ε-caprolactone is at least 99.5 %, although higher purity grades (e.g. 99.9 %) are also marketed. ε-Caprolactone does not contain additives.

1.3 Physico-Chemical properties

Table 1 Summary of physico-chemical properties

Property	Value	Reference
Physical state	Colourless liquid	
Melting point	-1.3°C	Weast (1986)
Boiling point	237 °C	White *et al*. (2004)
Relative density	1.07 g/cm³ at 20°C	Weast (1986)
Vapour pressure	0.81 Pa at 25°C	Tremain (2004)
Water solubility	Miscible in all proportions	White *et al*. (2004)
Partition coefficient n-octanol/water (log value)	0.68	Calculated value, EPA (2000a)
Henry's law constant	3.62E-05 atm.m^3/mol	Calculated value, EPA (2000b)

2 GENERAL INFORMATION ON EXPOSURE

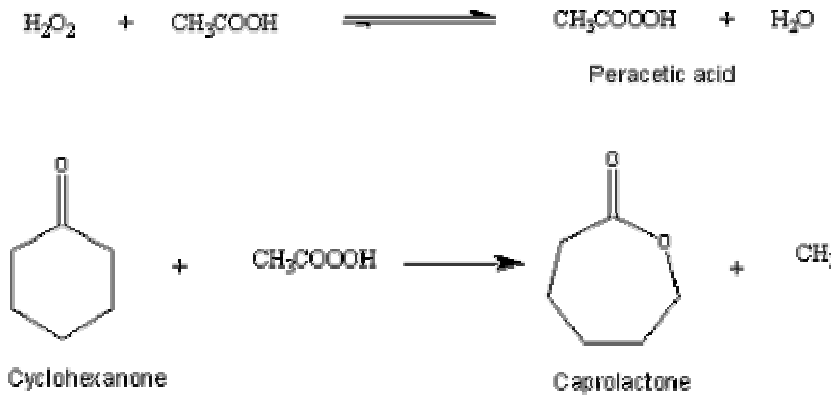
About 50 % of the quantity produced is used on site for the production of polymers (polycaprolactones). The remaining 50 % is sold to customers (downstream users) as an

intermediate for use in the manufacture of resins and polymers. The information on production and use, presented in chapter 2, is based on internal information from the consortium members. Published data could not be found.

2.1 Production Volumes and Use Pattern

In 2003, the estimated world-wide production of ε-caprolactone was 40,000 – 60,000 tonnes. In recent years the world-wide production was growing slowly (<5 % per year). The production occurs at four sites located in the USA (two sites), Japan and the United Kingdom.

ε-Caprolactone is manufactured using a process which utilises a high strength oxidising agent to produce a high purity peracetic acid. Peracetic acid is used to oxidise cyclohexanone by a Bayer-Villager reaction. The unreacted cyclohexanone is separated by distillation and recycled to the oxidation stage. Acetic acid is also recycled to the oxidation stage. The reactions are shown in the following equations:



The total number of downstream users is less than 1000. ε-Caprolactone is used by downstream users to modify resins and polymers in order to enhance the performance of the end-products. It is capable of addition reactions with a range of functional groups such as OH, COOH and NH_2. The majority is used for the modification of acrylic resins and polyesters, but it is also used for modification of epoxy resins and polyurethanes. A small quantity of ε-caprolactone (< 1 %) is used as reactive diluent and as a solvent (e.g. for vinyl resins). Based on the information available to the consortium members, ε-caprolactone is not used in consumer products.

e-Caprolactone is listed as a monomer in Section B of Commission Directive 2002/72/EC relating to the plastic materials and articles intended to come into contact with foodstuffs. To continue the use of this monomer, a dossier was submitted in 2004 to the European Food Safety Authority (EFSA) for re-evaluation of the substance.

2.2 Environmental Exposure and Fate

A fugacity level III calculation, using a four compartment (air, water, soil and sediment) model has been conducted using the Mackay model (De Groot, 2004). A 100 % release to the water compartment was assumed. Based on the results of the calculation, ε-caprolactone is expected to partition almost exclusively to the aquatic compartment (> 99.9 %) with the remainder to sediment (0.042 %), soil (0.012) and air (0.00014 %).

2.2.1 Sources of Environmental Exposure

Releases into the environment may potentially occur during production and processing of ε-caprolactone. A release of ε-caprolactone to the environment could potentially also occur via use and disposal of polycaprolactone because polycaprolactone may be (bio)degraded to ε-caprolactone, 6-hydroxyhexanoic acid and oligomers, substances which are ultimately biodegradable (Hakkarainen and Albertsson, 2002).

2.2.2 Photodegradation

The rate of photodegradation of ε-caprolactone has been estimated with the AOP (v.1.90) component of EPIWIN suite, developed by EPA's Office of Pollution Toxics and Syracuse Research Corporation (EPA, 2000c). There is no absorption of solar radiation by ε-caprolactone in the troposphere. ε-Caprolactone undergoes photochemical degradation by hydroxyl radicals. The half-life was estimated to be 1.7 days based on a mean hydroxyl radical concentration of 1.5×10^6 cm^{-3} over a 12-hour day.

2.2.3 Stability in Water

In a study conducted according to OECD Guideline 111, the hydrolysis as a function of pH was determined at pH 1.2, 4, 7 and 9 at two different temperatures (Dolich, 2003). Analysis was performed with NMR. The following results were obtained:

- pH 1.2: $t_{½, 37°C}$ = 0.4 h (0.02 d)
- pH 4: $t_{½, 37°C}$ = 100 h (4.2 d); $t_{½, 20°C}$ = 376 h (16 d)
- pH 7: $t_{½, 37°C}$ = 258 h (11 d); $t_{½, 20°C}$ = 1261 h (53 d)
- pH 9: $t_{½, 37°C}$ = 8.2 h (0.3 d); $t_{½, 20°C}$ = 52 h (2.2 d)

No other hydrolysis products than 6-hydroxyhexanoic acid were observed. 6-Hydroxyhexanoic acid (CAS 1191-25-9) is not listed on the European Inventory of Existing Commercial Substances (EINECS). Toxicity data on this substance were not found. However, based on structural comparison the toxicological properties of this substance are expected to be similar to hexanoic acid and adipic acid.

2.2.4 Biodegradation

ε-Caprolactone is readily biodegradable according to an OECD 301 B guideline study with > 60 % biodegradation within 14 days (Keetelaar-Jansen and Thus, 1993) based on the evolution of CO_2. Domestic activated sludge at a concentration of 30 mg dry weight/l was used as inoculum. The test concentrations of ε-caprolactone were 10 and 20 mg/l. After 28 days, degradation was 100 % and 58 %, respectively (the reason for this difference in degradation rate is not known). The activity of the inoculum displayed adequate activity based on the 60 % degradation of sodium acetate within 28 days. ε-Caprolactone was not toxic for the inoculum.

2.2.5 Bioaccumulation

It is anticipated that ε-caprolactone will not bioaccumulate based on its low octanol-water partition coefficient and rapid degradation in the environment. Based on modelling the bioconcentration factor was estimated to be 3.16 (EPA, 2000d).

2.3 Human Exposure

2.3.1 Occupational Exposure

During production and processing, inhalation of vapours and direct skin contact are potentially relevant exposure scenarios. However, inhalation exposures to vapours of ε-caprolactone at ambient temperature are likely to be limited due to its low volatility. Although accidental worker exposures to the skin (with subsequent oral uptake) and eyes could occur, these exposures are likely to be limited by the use of personal protective equipment recommended by the manufacturers. This is consistent with the fact that accidental exposures during the production and processing of ε-caprolactone have not been found in the medical literature. Occupational exposure limits have not been established by authorities.

2.3.2 Consumer Exposure

Based on the information available to the consortium members, ε-caprolactone is not used in consumer products. Based on the household products database of the US National Library of Medicine, ε-caprolactone is not an ingredient of consumer products (NIH, 2003). The substance is listed in the SPIN database (SPIN, 2005). In Norway (not for Denmark nor Finland; Sweden no data given due to confidentiality issue), consumer preparations would be available since 2002. However, with the most important application being 'construction materials' and 'paints, lacquers and varnishes' one might wonder if the entry is not due to the use of polycaprolactone i.e. the polymerisation product of the studied substance.

3 HUMAN HEALTH HAZARDS

3.1 Effects on Human Health

3.1.1 Toxicokinetics, Metabolism and Distribution

ε-Caprolactone is rapidly hydrolysed in the stomach because the half-life at a pH of 1.2 and a temperature of 37 °C is 0.4 hours (see section 2.2.3). Hydrolysis of e-caprolactone results in the formation of 6-hydroxyhexanoic acid. Specific toxicological studies with 6-hydroxyhexanoic acid could not be found in the literature but information on analogues can be found in the Annex.

ε-Caprolactone is not only hydrolysed at low pH but is also hydrolysed in the blood. Billecke et al. (2000) reported that human serum paraoxonase (PON1) isozymes Q and R are able to hydrolyse a large group of different lactones including ε-caprolactone. γ-Butyrolactone (CAS No. 96-48-0) has a similar structure as ε-caprolactone but has only 4 instead of 6 carbon atoms. γ-Butyrolactone was rapidly hydrolysed by an enzyme found in the blood and liver and the half-life of the conversion was less than 1 minute (NTP, 1996). For this reason e-caprolactone is expected to be hydrolysed rapidly in the blood.

3.1.2 Acute Toxicity

Inhalation

Six adult male rats were exposed for 8 hours to a saturated vapour (actual concentration is not reported) generated at room temperature by passing air through a fritted glass disc immersed in 50 ml ε-caprolactone (Smyth *et al.*, 1953). The rats were subsequently observed for a total of 14 days. No mortality was observed. The only notable response was slight skin irritation. No details of this

study are available and therefore the reliability of this study could not be established. A reliable guideline study is not available.

Dermal

ε-Caprolactone was administered to the clipped skin of male New Zealand White rabbits (Smyth *et al.*, 1953). The rabbits were exposed (occlusive method) to the test substance for 24 hours to 5,000 or 10,000 mg/kg bw. No control group was used. The LD_{50} for the undiluted compound was 5,990 ml/kg bw with confidence intervals of 4,270 – 8,420 ml/kg bw. Autopsies revealed congested or hemorrhagic lungs, and extremely congested livers. Skin erythema was observed, but no further details are available. Although the study was not conducted under current test guidelines, it is considered the critical study for acute dermal toxicity because (a) the test methodology is not significantly different from that used today, (b) the study is reasonably well documented, and (c) the study provides an adequate balance between data needs and animal welfare concerns.

Oral

A guideline study of Snoeij and Buse-Pot (1992) reported an LD_{50} of > 2000 mg/kg bw. Five male and five female Wistar rats were dosed with an ε-caprolactone solution at 2,000 mg/kg bw with 1.25 % tragancanth in distilled water as vehicle. None of the male rats died within the 14-day observation period, but 2 of the 5 females were found dead on day 2. Clinical signs observed were decreased locomotor activity, abnormal gait and posture, loss of righting reflex, changes in body and limb tone, decreased respiratory rate, respiratory difficulties, apathy and changes in startle response. Signs were noted within 30 minutes of dosing and had disappeared by 3 days. Observations at necropsy, of the animals that died during the study and those surviving until the end of the observation period, did not reveal any macroscopic abnormalities.

Carworth-Wistar rats were dosed by gavage with 10% aqueous solutions of ε-caprolactone at doses of 2,000; 4,000 and 8,000 mg/kg bw (Smyth *et al.*, 1953). The LD_{50} was 4,290 mg/kg bw with confidence intervals of 3,070-5,980 mg/kg bw. Symptoms apparent within 4 hours of dosing included narcosis, prostration, ruffed coats and sluggishness. Autopsies of those dying revealed congestion of the lungs, mottling of livers, paleness of kidneys and gastrointestinal tract irritation and burning. Although this study was not performed under GLP (Good Laboratory Practice) or current test guidelines, it is considered the critical study for acute oral toxicity because (a) the methodology used was comparable to current guidelines, (b) results are consistent with those of a guideline study at a limit dose, (c) the study is reasonably well documented, and (d) the study provides a balance between data needs and animal welfare concerns.

Other Routes of Exposure

A reliable i.p. study was conducted with ε-caprolactone to set dose levels for the mouse micronucleus assay (Ramadevi and Ritter, 1997). Groups of five male and five female ICR mice were dosed by intraperitoneal injection at a constant volume of 20 ml/kg body weight at dosage levels at 800, 1000, 1200, 1400, 2000 and 3000 mg/kg bw. Mortality occurred within three hours of dose administration in 5/5 males and 5/5 females at 1400, 2000 and 3000 mg/kg bw. Mortality occurred within two days of dose administration in 2/5 males and 2/5 females at 1200 mg/kg bw. Clinical signs noted after dosing included lethargy in male and female mice at 800, 1000 and 1200 mg/kg bw and gasping and convulsions in male and female mice at 1400, 2000 and 3000 mg/kg bw. The LD_{50} was 1255 mg/kg bw. These results are consistent with those of a less reliable study (original reference not available) that reported an LD_{50} of 1300 mg/kg bw in male Swiss Webster mice dosed by intraperitoneal injection (Simmon *et al.*, 1979).

Conclusion

ε-Caprolactone exhibits low acute toxicity by all potentially relevant routes of exposure. In an acute inhalation study, no effects were found at the highest attainable concentration (saturated vapour). The dermal (rabbit) and oral (rat) LD_{50} values were 6,400 mg/kg bw and 4,290 mg/kg bw, respectively. The intraperitoneal (mouse) LD_{50}, which does not represent a relevant route of exposure, was 1255 mg/kg bw.

3.1.3 Irritation

Skin Irritation

In a guideline, study three male rabbits were dermally exposed to 0.5 g undiluted ε-caprolactone (Janssen, 1991a). Neither erythema nor oedema was observed at 0.5, 24, 48 and 72 hours after removal of the patch. These results are consistent with those of a pre-guideline study (Smyth *et al*., 1953) in which 0.01 ml of undiluted test compound was applied uncovered to the clipped skin of five rabbits. Another study (Marhold, 1986) reported ε-caprolactone was moderately irritating when applied neat (0.5 g) to the skin of rabbits. As the original reference is unavailable, the latter study was judged to be of lower reliability.

Eye Irritation

In a guideline study, the left eyes of four male rabbits were exposed to 0.1 ml undiluted ε-caprolactone, while the right eyes served as untreated controls (Janssen, 1991b). The eyes were not rinsed. ε-Caprolactone was considered to cause significant but reversible eye irritation. These results are consistent with those of a pre-guideline study with a lower reliability (Smyth *et al*. 1953) that reported ε-caprolactone to be highly irritating when instilled in the eyes of five rabbits either neat (0.005 ml) or as a 15 % dilution in propylene glycol (0.5 ml).

Conclusion

ε-Caprolactone is not irritating to the skin but is irritating to the eyes.

3.1.4 Sensitisation

No sensitisation studies are available or described in the literature.

3.1.5 Repeated Dose Toxicity

There are three animal studies available on repeated dose toxicity of ε-caprolactone.

Inhalation

In a well-documented 9-day inhalation study, groups of 20 Sprague-Dawley rats (10/sex) were exposed to either 0 or 45 ppm (213 mg/m^3) ε-caprolactone (Norris and Kintigh, 1991). The animals were exposed 6 hours per day, for 9 exposures during a 2-week period. The concentrations of ε-caprolactone were analyzed six times during each of the nine, six-hour exposure periods. The control chamber atmosphere was analyzed once each exposure period. The mean ε-caprolactone concentration was 44 ppm; no ε-caprolactone was detected in the control chamber. There were no mortalities during the study. Animals exposed to ε-caprolactone did not display changes in body weight, body weight gain, organ weight, ophthalmic observations, hematologic values, necropsy observations, or histopathologic evaluation. Because perinasal encrustation (only effect noted) was

observed in both males and females of the control and exposed groups, this effect was not considered test substance related. The NOAEL was judged to be 45 ppm (213 mg/m^3).

Norris and Kintigh (1992) conducted a well-documented 90-day inhalation study in which groups of 20 rats (10/sex) were exposed to ε-caprolactone at target concentrations of 0, 15, or 45 ppm (0, 71 or 213 mg/m^3) for 6 hours/day, 5 days/week over 13 weeks. Ten additional males and females were added to the 0 and 45 ppm groups to evaluate the effects after a 4-week recovery period. The concentrations of ε-caprolactone vapor were analyzed six times during each daily 6-hour exposure period. The control chamber was analyzed once each exposure period. GC-analysis of the chamber atmosphere resulted in mean concentrations of 14.2 and 42.4 ppm; no ε-caprolactone was detected in the control chamber. There were no mortalities during the study. There were no differences between controls and treated rats with respect to total weights, weight change, food consumption, haematology, clinical chemistry, gross pathology or histopathology. The only ε-caprolactone exposure-related lesions at the 14-week sacrifice were perinasal and periocular encrustation and eyelid swelling in males of the 45 ppm group. This level is considered the lowest observed adverse effect level (LOAEL) for the 90 day study. The 15 ppm level (71 mg/m^3) is considered the NOAEL.

Oral

Groups of 20 Sprague-Dawley rats (10/sex) were exposed to ε-caprolactone via drinking water at doses of 0, 500, 2000 and 5000 ppm for 14 days (Hermansky *et al.*, 1991). The study was well documented. Mean ε-caprolactone intakes (calculated using nominal drinking water concentrations and water consumption rates) were 45, 152 and 347 mg/kg bw/day for males and 53, 184 and 384 mg/kg bw/day for females for the 500, 2000 and 5000 ppm groups, respectively. Treatment with ε-caprolactone did not result in any treatment-related clinical signs of toxicity, clinical pathology findings, organ weight changes, necropsy observations or histopathological findings. The only effect in clinical chemistry that could possibly be related to treatment was an increased urea nitrogen observed in the males of the 5000 ppm group. As there was no dose-effect-relationship and there were no histological lesions observed in the kidneys, this effect was not considered toxicologically relevant. Doses of 2000 and 5000 ppm produced effects on food and water consumption, which were attributed to aversion to the drinking water solutions. Body weight gain in the 5000 ppm group was reduced throughout the study and based on the whole exposure period the reduction was 24 %. In the 2000 ppm group body weight gain was reduced during day 0-4 of the study only (32 %), but based on the whole exposure period (day 0-14) there was no reduction (4 %, not statistically significant) in body weight gain. Therefore, the NOAEL was considered to be 2000 ppm (152 and 184 mg/kg bw for males and females, respectively).

Conclusion

ε-Caprolactone administered as vapour to rats for a 9-day period at a concentration of 45 ppm did not cause any adverse effects. The only effects found in a 90-day inhalation study were perinasal and periocular encrustation and eyelid swelling in the males of the 45 ppm group. No effects were found at the 15 ppm group. ε-Caprolactone given by drinking water to rats for 14 days affected food and water consumption (low palatability) as well as body weight gain at the level of 5000 ppm. The NOAEL was 2000 ppm, which is equivalent with a dose 152 and 184 mg/kg bw for males and females, respectively.

3.1.6 Mutagenicity

In vitro Studies

In vitro studies on possible gene mutation activity of ε-caprolactone are presented in Table 2. The first four studies are well documented studies. Other findings are from literature references and for these studies the documentation was insufficient for assessment.

A mammalian cell gene mutation assay, with and without a metabolic activation system, was conducted (San and Clarke, 1997). Chinese Hamster Ovary (CHO) cells were used and the mutagenic potential was based on mutations of the HGPRT locus. In the non-activated system, cultures were treated with concentrations of 1000-5000 µg/ml. The S9-activated cell cultures were treated with concentrations of 250-5000 µg/ml. No positive responses (i.e., treated cultures with mutant frequencies > 40 mutants per 10^6 clonable cells) were observed. Toxicity was not observed in the non-activated cultures but was observed at doses ≥ 2100 µg/ml with S9 activation.

Another well-documented gene mutation assay with CHO cells was conducted in which ε-caprolactone was tested at concentrations of 0.0625, 0.125, 0.25, 0.5, 1.0 % v/v (without S9 activation) and at 0.00625, 0.0125, 0.025, 0.05 and 0.1 % v/v (with S9 activation). Positive and negative controls were included in the test. ε-Caprolactone produced three statistically significant increases in the frequency of mutations without metabolic activation, but without a dose-related effect. No significant effect on mutant frequency was obtained with S9 metabolic activation (Slesinski *et al*., 1981).

Table 2 In Vitro Genetic Toxicity Assays of ε-caprolactone

Test system	Test organism, Strain	Dose (product)	Metabolic activation	Result	Reference
Mammalian cell gene mutation assay	CHO/ HGPRT	250-5000 µg/ml 1000-5000 µg/ml	With and without	Neg	San and Clarke, 1997
Mammalian cell gene mutation assay	CHO/ HGPRT	0.00625-0.1% 0.0625-1.0%	With without	Neg Ambiguous	Slesinski *et al*., 1981
Sister chromatid exchange assay	CHO cells	0.00625-0.1% 0.0625-1.0%	With and without	Neg	Slesinski *et al*., 1981
Unscheduled DNA synthesis	Hepatocyte suspension	0.0001-0.1%	Not applicable	Ambiguous	Slesinski *et al*., 1981
Mouse lymphoma assay	L5178Y cell	No data	Without	Neg	Clive *et al*., 1983 *
Ames	TA98, 100, 1535, 1537	10, 100, 500 or 1000 µg/plate	With	Neg	McCann *et al*., 1975 *
Ames	TA 1535 and TA1538	0, 1 and 100 µl/plate	With and without	Neg	Rosenkranz and Poirier, 1979 *
Ames	TA98, 100, 1535, 1536, 1537, 1538	Up to 250 µg/plate	With and without	Neg	Simmon, 1979a *
Intraperitoneal host-mediated assay	Mice implanted with S. typhimurium and S. cerevisiae	1st group 432 mg/kg bw, 2nd group 1300 mg/kg bw	Without	Neg	Simmon *et al.*, 1979 *
Chromosome aberration test	Chinese Hamster cells	$1x10^{-5}$, $1x10^{-4}$, $1x10^{-3}$	No data	Neg	Abe and Sasaki, 1977 *
Chromosome aberration test	Chinese Hamster cells	Max. conc. was 0.5 mg/ml	Without	Neg	Ishidate M *et al.*, 1977 *
Sister chromatid exchanges	Hamster cells	$1x10^{-5}$, $1x10^{-4}$, $1x10^{-3}$	No data	Neg	Abe and Sasaki, 1977 *
Cell transformation	Hamster embryo cells	0, 0.1, 1.0, 10 and 100 µg/ml	No data	Neg	Pienta RJ *et al.*, 1977 *
Mitotic recombination	S. cerevisiae	Conc. up to 5 %	Without	Neg	Simmon, 1979b *

* Study had a low reliability (CoR = 4) because the documentation was insufficient for assessment.

A sister chromatid exchange assay with CHO cells was conducted (Slesinski *et al*., 1981). ε-Caprolactone was tested at concentrations of 0.0625, 0.125, 0.25, 0.5, 1.0 % v/v (without S9 activation) and at 0.00625, 0.0125, 0.025, 0.05 and 0.1 % v/v (with S9 activation). Positive and negative controls were included in the test. Only a moderate degree of cytotoxicity was obtained with the top concentration of ε-caprolactone. No statistically significant increase in the frequency of sister chromatid exchange was obtained at any concentration tested with or without the presence of a metabolic activation system.

The possible induction of Unscheduled DNA Synthesis by ε-caprolactone was investigated in rat liver cells (Slesinski *et al*., 1981). Concentrations of 0, 0.0001, 0.001, 0.003, 0.01, 0.03 and 0.1 % (v/v) ε-caprolactone were used. Positive controls were included. Three concentrations tested for potential activity produced a statistically significant increase in the amount of tritiated-thymidine incorporation. Also, all six concentrations produced numerical increases in the amount of UDS in comparison to the solvent control. However, there was no distinct dose-related increase in the

amount of UDS, characteristic of strong mutagenic agents. In this assay only a moderate degree of cytotoxicity was obtained with the top concentration of ε-caprolactone.

Many other *in vitro* studies are mentioned in Table 2 but the documentation of these studies was insufficient for assessment. However, all responses were negative.

In vivo Studies

In a well-documented micronucleus assay male and female mice were dosed by i.p. injection with 250, 500 or 1000 mg ε-caprolactone/kg body weight (Ramadevi and Ritter, 1997). No mortality was observed in either male or female mice. However, male and female mice showed lethargy at all dose levels and at 1000 mg/kg body weight prostration of males and females was observed. Bone marrow cells, collected at 24, 48 and 72 hours after treatment, were examined microscopically for micronucleated polychromatic erythrocytes. No significant increase in micronucleated polychromatic erythrocytes in the exposed groups relative to the control group was observed in male or female mice ($p>0.05$, Kastenbaum-Bowman). ε-Caprolactone was concluded to be negative in the mouse micronucleus assay.

Conclusion

Bacterial and mammalian *in vitro* mutagenicity tests gave in general negative results. *In vivo*, ε-caprolactone was negative in the mouse micronucleus assay.

3.1.7 Carcinogenicity

Limited information is available from a skin painting study (Mellon Institute of Industrial Research, 1961) in mice. Mice were painted daily with undiluted ε-caprolactone for 24 month during which no tumors were observed.

3.1.8 Toxicity to Development/Reproduction

No studies are available with regard to reproduction and developmental toxicity of ε-caprolactone. However, a well conducted 90-day inhalation study showed no macroscopic and histopathological effects on reproductive organs and also other systemic effects were not found during this study. The lack of systemic effects can be explained by the rapid hydrolysis in stomach and blood, resulting in the formation of 6-hydroxyhexanoic acid (see section 3.1.1).

Specific toxicological studies could not be located for 6-hydroxyhexanoic acid or other hydroxyhexanoic acids. However, the toxicological properties of 6-hydroxyhexanoic acid can be predicted based on the chemical structure. Information is available for analogues of 6-hydroxyhexanoic acid (see Annex). In conclusion:

- 1-Hexanol was not teratogenic to rats,
- For 1,6-hexanediol there is no indication of toxic effects on reproductive function or developmental toxicity,
- Adipic acid was not teratogenic and there is no reason to expect specific reproductive toxicity and
- Aliphatic carboxylic acids show no significant evidence of either reproductive or developmental toxicity.

γ-Butyrolactone (CAS No. 96-48-0) has a similar structure as ε-caprolactone but has only 4 instead of 6 carbon atoms. This chemical was evaluated in 14-day, 13-week and 2-year toxicology and carcinogenesis studies and no organ-specific toxicity was observed (NTP, 1996). Furthermore

γ-butyrolactone was rapidly hydrolysed by an enzyme found in the blood and liver and the half-life of the conversion was less than 1 minute.

δ-Valerolactone (CAS No. 542-28-9) also has a similar structure as ε-caprolactone but has 5 instead of 6 carbon atoms. Based on a "Commission Decision of 23 January 2002 amending Commission Decision 1999/217/EC as regards the register of flavouring substances used in or on foodstuffs" this substance is allowed in the European Union as a flavouring substance.

Conclusion

No studies are available with regard to reproduction and developmental toxicity of ε-caprolactone. However, a well conducted 90-day inhalation repeated dose study showed no macroscopic and histopathological changes on reproductive organs and also other systemic effects were not found during this study. The lack of systemic effects can be explained by the rapid hydrolysis in stomach and blood, resulting in the formation of 6-hydroxyhexanoic acid. Analogues of 6-hydroxyhexanoic acid show no evidence of reproductive or developmental toxicity. For this reason there is no indication for a reprotoxic concern. This is supported by the toxicological profile of structurally similar lactones, where also no organ specific toxicity was observed in long term studies (with up to 2-year exposure).

3.2 Initial Assessment for Human Health

After absorption of ε-caprolactone, the substance will be hydrolyzed rapidly in stomach and blood resulting in the formation of 6-hydroxyhexanoic acid. This hydrolysis product is water soluble and expected to be distributed throughout the body and excreted rapidly, principally through the urine.

ε-Caprolactone exhibits low acute toxicity by all potentially relevant routes of exposure. The acute oral LD_{50} for rats was 4290 mg/kg, while the acute dermal LD_{50} in rabbits was 6400 mg/kg body weight. The primary symptoms, following a single high dose, are skin erythema (dermal) as well as apathy and effects on motor coordination and respiration (oral). ε-Caprolactone is considered not-irritating to skin and irritating to eyes.

In a 9-day inhalation study in which ε-caprolactone was administered at a concentration of 45 ppm (213 mg/m^3), no treatment-related effects were found. Therefore the 45 ppm level can be considered a NOAEL. A 90-day inhalation study with ε-caprolactone at concentrations of 15 ppm (71 mg/m^3) and 45 ppm (213 mg/m^3) resulted in perinasal and periocular encrustation and eyelid swelling in the males of the 45 ppm group. As no other treatment related effects were found, this level is considered the lowest observed adverse effect level (LOAEL). The 15 ppm level is considered the NOAEL. ε-Caprolactone given by drinking water to rats at levels of 500, 2000 and 5000 ppm in a 14-day study did not result in any treatment-related clinical signs of toxicity, clinical pathology findings, organ weight changes, necropsy observations or histopathological findings. ε-Caprolactone affected food and water consumption (low palatability) as well as body weight gain at the level of 5000 ppm only. The NOAEL was 2000 ppm, which is equivalent with a dose 152 and 184 mg/kg bw for males and females, respectively.

Bacterial and mammalian *in vitro* mutagenicity tests gave in general negative results. *In vivo*, ε-caprolactone was negative in the mouse micronucleus assay.

No studies are available with regard to reproduction and developmental toxicity of ε-caprolactone. However, a well conducted 90-day inhalation repeated dose study showed no macroscopic and histopathological changes on reproductive organs and also other systemic effects were not found during this study. The lack of systemic effects can be explained by the rapid hydrolysis in stomach and blood, resulting in the formation of 6-hydroxyhexanoic acid. Analogues of 6-hydroxyhexanoic acid show no evidence of reproductive or developmental toxicity. For this reason there is no

indication for a reprotoxic concern. This is supported by the toxicological profile of structurally similar lactones, where also no organ specific toxicity was observed in long term studies (with up to 2-year exposure).

4 HAZARDS TO THE ENVIRONMENT

4.1 Aquatic Effects

The results of available aquatic ecotoxicity tests, conducted according to GLP and standard guidelines, have been summarised in Table 3.

Table 3 Ecotoxicity of ε-caprolactone

Species	L(E)C_{50} (mg/l)	NOEC (mg/l)	Reference
Scenedesmus subspicatus (alga)	1217	256	Werner, 2003
Daphnia magna (invertebrate)	204	124	Hisgen, 2003
Poecilia reticulata (fish)	295	250	Groeneveld *et al*., 1992 Tolboom, 2004
Pseudomonas putida (bacterium)	1260	32	Jansen and van den Berg, 1992

Acute Toxicity Test Results

In order to estimate the toxicity of ε-caprolactone to aquatic plants, a growth inhibition test to the alga *Scenedesmus subspicatus* was performed according to GLP and standard test guidelines (Werner, 2003). Nominal ε-caprolactone concentrations in the growth medium ranged from 102 to 4000 mg/l. The analytical results yielded 80 % or higher recoveries. The EC_{50} (72 h) for the growth inhibition based on biomass was calculated to be 1217 mg/l. Based on the specific growth rate (μ), the EC_{50} (72 h) was calculated to be 2616 mg/l. The NOEC (72 h) was 256 mg/l based on biomass.

The effects of ε-caprolactone on the water flea *Daphnia magna* have been studied by Hisgen (2003) according to GLP and standard test guidelines. Daphnids were exposed for 48 hours to nominal test concentrations of 0, 62.5, 125, 250, 500 and 1000 mg/l. The ε-caprolactone concentrations were measured at the start and at the end of the test with GC analysis. The measured concentration of ε-caprolactone was > 95 % of nominal. The EC_{50} (48h) and NOEC (48h) of ε-caprolactone were 204 and 125 mg/l, respectively. No control immobility was observed.

A static acute toxicity study with the guppy (*Poecilia reticulata*) and ε-caprolactone has been conducted according under GLP and OECD test guidelines (Groeneveld *et al*., 1992; Tolboom, 2004). Fish were exposed for 96 hours to nominal ε-caprolactone concentrations of 0; 31; 62; 125; 250; 500 and 1000 mg/l and observations were made after 24, 48, 72 and 96 hours. Samples of the test solution were taken on several days during the study and analysed with HPLC. The measured concentration of ε-caprolactone was > 90 % of nominal. The LC_{50} (96h) and NOEC (96h) for ε-caprolactone were 295 and 250 mg/l, respectively. No control mortality was observed.

Another static acute toxicity study with the fathead minnow (*Pimephales promelas*) was performed (Waggy and Payne, 1974). No analytical measurements were performed. The LC_{50} (96h) was 320 mg/l. This study was performed before official guidelines and GLP were in place, however the study is reasonably documented. The LC_{50} value of the fathead minnow study (320 mg/l) agreed well with the LC_{50} value of the guppy study (295 mg/l).

Hydrolysis of ε-caprolactone results in the formation of 6-hydroxyhexanoic acid. Data on the ecotoxicity of this substance were not found. However, ecotoxicity data are available for the analogue adipic acid (OECD, 2004). The acute EC50 values for fish (*Danio rerio*) and water flea (*Daphnia magna*) were > 1000 and 86 mg/l, respectively. In an algae growth inhibition test with *Desmodesmus subspicatus* the 96 h-E_bC_{50} of adipic acid was 27 mg/l and the 72 h-E_bC_{50} was 31 mg/l.

Chronic Toxicity Test Results

No data on chronic toxicity are available.

Toxicity to Microorganisms

To determine the toxicity to microorganisms, a test with the bacteria *Pseudomonas putida* was conducted under GLP and ISO guidelines (Jansen and van den Berg, 1992). In this test *Pseudomonas putida* was exposed for 16 hours to nominal ε-caprolactone concentrations of 0, 16, 31, 63, 125, 250, 500 and 1000 mg/l. Samples of the stock solution were taken and the total organic carbon content was determined. For the endpoints calculated concentrations were used. Significant inhibition of cell multiplication occurred at measured concentrations of 63 mg/l and higher. The EC_{50} and NOEC were 1260 mg/l and 32 mg/l, respectively.

4.2 Terrestrial Effects

No data on terrestrial effects are available.

4.3 Other Environmental Effects

No other environmental effects are expected.

4.4 Initial Assessment for the Environment

Based on the Mackay model (level III) ε-caprolactone is expected to partition almost exclusively to the aquatic compartment (> 99.9 %). In water ε-caprolactone is hydrolysed to 6-hydroxyhexanoic acid. At 20 degrees Celsius the half-life at pH values of 4, 7 and 9 was 16, 53 and 2.2 days, respectively. 6-Hydroxyhexanoic acid (CAS 1191-25-9) is not listed on the European Inventory of Existing Commercial Substances (EINECS). Ecotoxicity data of this hydrolysis product were not found. However, based on structural comparison the ecotoxicological properties of this substance are expected to be similar to hexanoic acid and adipic acid. ε-Caprolactone is readily biodegradable according to an OECD 301 B guideline study. It is anticipated that ε-caprolactone will not bioaccumulate based on its low octanol-water partition coefficient and rapid degradation in the environment.

Aquatic ecotoxicity tests, which were done according to GLP and standard guidelines, are available for 4 different species encompassing the 3 trophic levels and microorganisms. A 72 hour toxicity test with algae (*Scenedesmus subspicatus*) revealed an EC_{50} and NOEC value of 1217 and 256 mg/l, respectively (based on biomass). Based on the specific growth rate (μ), the EC_{50} (72h) was calculated to be 2616 mg/l. Water fleas (*Daphnia magna*) appeared to be more sensitive than algae. An acute test with an exposure period of 48 hours resulted in EC_{50} and NOEC values of 204 and 124 mg/l, respectively. For guppy (*Poecilia reticulata*) a steep concentration-response relationship was observed. A toxicity test with a duration of 96 hours with this fish species revealed an LC_{50} and NOEC value of 295 and 250 mg/l, respectively. However for bacteria (*Pseudomonas putida*) a large difference between the EC_{50} (1260 mg/l) and NOEC (32 mg/l) was found. During this test the

bacteria were exposed for 16 hours. Neither chronic aquatic toxicity tests nor terrestrial toxicity tests are available for ε-caprolactone.

5 RECOMMENDATIONS

Human Health: The chemical possesses properties indicating a hazard for human health (eye irritation). This hazard does not warrant further work as it relates to reversible effects. This should nevertheless be noted by chemical safety professionals and users.

Environment: The chemical is currently of low priority for further work because of its low hazard potential.

6 REFERENCES

Abe S and Sasaki M (1977). Chromosome Aberrations and Sister Chromatid Exchanges in Chinese Hamster Cells Exposed to Various Chemicals. J. Nat. Cancer, 58, 1635-1640.

Billecke S, Draganov D, Counsell R, Stetson P, Watson C, Hsu C and La Du BN (2000). Human serum paraoxonase (PON1) isozymes Q and R hydrolyze lactones and cyclic carbonate esters. Drug Metabolism and Disposition, 28 (11), 1335-1342.

Clive D, McCuen R, Spector JFS, Piper C and Mavournin KH (1983). Specific gene mutations in L5178Y cells in culture. Mutation Research, 115, 225-251.

De Groot WA (2004). Estimation of environmental partitioning of epsilon-caprolactone. Internal Memorandum AdG/HES/im/04.316 of Solvay Pharmaceuticals.

Dolich T (2003). Hydrolysis as a function of pH of E-caprolactone. BASF GKA Analytik, Study No. 03L00213.

EPA (2000a). KOWWIN v1.66.

EPA (2000b). HENRYWIN v1.90.

EPA (2000c), AOPWIN v1.90

EPA (2000d) BCFWIN v2.14

Groeneveld AHC, Van Dijk NRM and Van den Berg G (1992). The acute toxicity of ε-caprolactone to the guppy Poecilia reticulate. Solvay Duphar Int. Doc. No. 56635/19/92.

Hakkarainen M and Albertsson AC (2002). Heterogeneous Biodegradation of Polycaprolactone – Low Molecular Weight Products and Surface Changes. Macromol. Chem. Phys., 203, 1357-1363.

Hermansky SJ, Chun JS and Loughran KA (1991). Caprolactone: Two-Week Drinking Water Study in Sprague-Dawley Rats. Bushy Run Research Center, Report No. 54-40.

Hisgen (2003). Epsilon-Caprolactone - Determination of the acute effect on the swimming ability of the water flea Daphnia magna Straus. BASF Experimental Toxicology and Ecology Project No. 03/0034/50/1.

Ishidate M and Odashima S (1977). Chromosome tests with 134 compounds on Chinese hamster cells in vitro - A screening for chemical carcinogens. Mutation Research, 48, 337-354.

Jansen WAJ and Van den Berg G (1992). The toxicity of ε-caprolactone to the bacteria Pseudomonas putida. Solvay Duphar Int. Doc. No. 56635/47/92.

Janssen PJM (1991a). Primary irritation of ε-caprolactone to the skin of male rabbits. Solvay Duphar Int. Doc. No. 56645/69/91.

Janssen PJM (1991b). Primary irritation study of ε-caprolactone to the eye of male rabbits. Solvay Duphar Int. Doc. No. 56645/106/91.

Keetelaar-Jansen WAJ and Thus J (1993). Determination of the biodegradability of ε-caprolactone in a modified Sturm test. Solvay Duphar Int. Doc. No. 56835/2/93.

Marhold J (1986). Avicenum, p. 802 (in RTECS dd 8/3/89).

McCann J, Choi E, Yamasaki E and Ames BN (1975). Detection of carcinogens as mutagens in the Salmonella/microsome test: Assay of 300 chemicals. Proc. Nat. Acad. Sci. USA, 72 (12), 5135-5139.

Mellon Institute of Industrial Research (1961). Studies on Carcinogenesis Completed in 1961. Union Carbide Chemicals Co Report 24-117.

Nelson BK, Brightwell WS, Khan A, Krieg EF and Hoberman AM (1989). Developmental Toxicology Evaluation of 1-Pentanol, 1-Hexanol, and 2-Ethyl-1-Hexanol Administered by Inhalation to Rats. Journal of the American College of Toxicology, 8 (2), 405-410.

NIH (2003). Household Products Database. http://householdproducts.nlm.nih.gov/index.htm

Norris DB and Trudgill PW (1971). The metabolism of cyclohexanol by *Nocardia globerula* CL1. Biochem. J., 121, 363-370.

Norris JC and Kintigh WJ (1991). Caprolactone: Nine-Day Vapor Inhalation Study on Rats. Bushy Run Research Center, Report No. 54-2.

Norris JC and Kintigh WJ (1992). Caprolactone: Ninety-Day Vapor Inhalation Study on Rats. Bushy Run Research Center, Report No. 54-94.

NTP (1996). NTP Summary report on the metabolism, disposition and toxicity of 1,4-butanediol. Toxicity Report Series Numbers 54. NIH Publication 96-3932. http://ehp.niehs.nih.gov/ntp/docs/tox54/tx54cov.pdf

OECD (2004). SIDS Initial Assessment Profile (SIAP) of adipic acid. Available via: http://cs3-hq.oecd.org/scripts/hpv/

Pienta RJ, Poiley JA and Lebherz WB (1977). Morphological transformation of early passage golden syrian hamster embryo cells derived from cryopreserved primary cultures as reliable in vitro bioassays for identifying diverse carcinogens. Int. J. Cancer 19, 642-55.

Ramadevi G and Ritter P (1997). Micronucleus cytogenetic assay in mice. Microbiological Associates Inc., Laboratory Study Number G96CJ23.122.

Rosenkranz S and Poirier AL (1979). Evaluation of the Mutagenicity and DNA-Modifying Activity of Carcinogens and Noncarcinogens in Microbial Systems. J. Natl. Cancer Inst., 62 (4), 873-891.

San HC and Clarke JJ (1997). In Vitro Mammalian Cell Gene Mutation Test with an Independent Repeat Assay. Microbiological Associates Inc., Study No. G96CJ23.782001.

Simmon VF (1979a). In Vitro Mutagenicity Assays of Chemical Carcinogens and Related Compounds with Salmonella typhimurium. J. Natl. Cancer Inst., 62 (4), 893-899.

Simmon VF (1979b). In Vitro Assays for Recombinogenic Activity of Chemical Carcinogens and Related Compounds with Saccharomyces Cerevisiae D3. J. Natl. Cancer Inst., 62 (4), 901-909.

Simmon VF, Rosenkranz HS, Zeiger E and Poirier LA (1979). Mutagenic activity of chemical carcinogens and related compounds in the intraperitoneal host-mediated assay. J. Natl. Cancer Inst., 62 (4), 911-918.

Slesinski RS, Gaunt MW, Guzzie PJ and Hengler WC (1981). In Vitro Mutagenesis Studies: 3-Test Battery. Bushy Run Research Center, Project Report 43-125.

Smyth HF, Carpenter CP, Weil CS and Pozzani UC (1953). Range-finding Tests on E-caprolactone. University of Pittsburgh, Report No. 16-59.

Snoeij NJ and Buse-Pot TE (1992). Acute oral toxicity with ε-caprolactone in the rat. Solvay Duphar Int. Doc. No. 56645/92/91.

SPIN (2005). Substances in Preparations in Nordic Countries. http://195.215.251.229/spin.html

Tanaka H, Obata H, Tokuyama T, Ueno T, Yoshizako F and Nishimura A (1977). Metabolism of cyclohexanol by *Pseudomonas* sp. Hakko-kogaku, 55, 62-67.

Tolboom J (2004). LC50 determination for fish study. Internal Memorandum of Solvay Pharmaceuticals, Weesp, The Netherlands.

Tremain SP (2004). E-Caprolactone: Determination of vapour pressure. SafePharm Laboratories Ltd Project Number 1524/004.

Waggy GT and Payne JR (1974). Environmental Impact Product Analysis Acute Aquatic Toxicity Testing. Research and Development Department, Chemicals and Plastics Union Carbide Corporation, South Charleston, West Virginia, Report No. 910F44.

Weast RC (1986). CRC Handbook of Chemistry and Physics. 66th Edition. Edited by MJ Astle and WH Beyer. CRC Press.

Werner DI (2003). Epsilon-Caprolactone - Determination of the inhibitory effect on the cell multiplication of unicellular green algae. BASF Experimental Toxicology and Ecology Project No. 03/0034/60/1.

White DF and Mullee DM (2004). E-Caprolactone: Determination of boiling point and water solubility. SafePharm Laboratories Ltd Project Number 1524/003.

ANNEX: REPROTOXICITY OF ANALOGUES OF THE HYDROLYSIS PRODUCT OF E-CAPROLACTONE

The hydrolysis of ε-caprolactone results in the formation of 6-hydroxyhexanoic acid. Please find below some basic information about this substance.

Name: 6-hydroxyhexanoic acid

Synonyms: hexanoic acid, 6-hydroxy

6-hydroxycaproic acid

ε-hydroxycaproic acid

CAS number: 1191-25-9

EINECS number: not available

ELINCS number: not available

Molecular formula: $C_6H_{12}O_3$

A literature search has been done concerning the toxicity of 6-hydroxyhexanoic acid. Relevant toxicological studies were not found. However, the literature search revealed that the micro-organisms *Nocardia globerula* CL1 and *Pseudomonas* spp. are able to oxidize 6-hydroxyhexanoic acid to adipic acid (Norris et al., 1971; Tanaka et al., 1977).

Other hydroxyhexanoic acids

In addition to 6-hydroxyhexanoic acid other hydroxyhexanoic acids do exist and their regulatory status is given below.

Substance	CAS number	EINECS no.	ELINCS no.
2-hydroxyhexanoic acid	6064-63-7	227-991-4	not available
3-hydroxyhexanoic acid	10191-24-9	not available	not available
4-hydroxyhexanoic acid	not available	not available	not available
5-hydroxyhexanoic acid	44843-89-2	not available	not available

Toxicological information does not seem to be available for these substances based on a literature search.

1-Hexanol

Nelson et al. (1989) reported a large study evaluating the developmental toxicity of industrial alcohols (including 1-hexanol):

> Groups of approximately 15 Sprague-Dawley rats were exposed for 7 h/day on gestation days 1-19 at the highest concentration they could generate as a vapor. The study indicated that inhalation of pentanol, 1-hexanol, or 2-ethyl-1-hexanol can produce limited maternal toxicity, but was none was teratogenic to rats.

1,6-Hexanediol

A substance which is similar to 6-hydroxyhexanoic acid is 1,6-hexanediol (CAS number 629-11-8). An OECD SIDS dossier has been prepared for this substance using the name hexamethylene glycol. The SIAR reported the following conclusion:

> In a valid OECD 421 study no indication of toxic effects on reproductive function or developmental toxicity were observed.

Hexanoic acid

Although a limited amount of data is available on the hydroxyhexanoic acids, more information is available for the substance hexanoic acid (CAS number 142-62-1 and EINECS number 205-55-7). A literature search revealed a significant amount of references. In several cases the substance was mentioned in publications on QSARs (Quantitative Structure-Activity Relationships).

Based on USA FDA Requirements [1 CFR 172.515 (4/1/91)] hexanoic acid is a food additive permitted for direct addition to food for human consumption, as long as

1) the quantity added to food does not exceed the amount reasonably required to accomplish its intended physical, nutritive, or other technical effect in food, and

2) when intended for use in or on food it is of appropriate food grade and is prepared and handled as a food ingredient.

In the European Union hexanoic acid is listed on Annex III of Commission Directive 2002/72/EC of 6 August 2002 relating to plastic materials and articles intended to come in contact with foodstuffs. Therefore hexanoic acid may be used as an additive in the manufacture of plastic materials and articles and a Specific Migration Limit (SML) does not apply.

Adipic acid

Another substance which is similar to 6-hydroxyhexanoic acid is adipic acid or hexanedioxic acid ($C_6H_{10}O_4$). The CAS number and EINECS number of adipic acid are 124-04-9 and 204-673-3, respectively. Adipic acid is formed when 6-hydroxyhexanoic acid is oxidized to the corresponding dicarboxylic acid, which is adipic acid.

Recently an OECD SIDS dossier has been prepared for adipic acid. Based on the SIAP:

> Adipic acid was not embryo- or fetotoxic and not teratogenic up the highest tested doses of 288, 263, and 250 mg/kg bw/day via oral administration to rats, mice, and rabbits, respectively. Based on the available data there is no reason to expect specific reproductive toxicity of adipic acid.

Also adipic acid is listed on Annex III of Commission Directive 2002/72/EC of 6 August 2002 relating to plastic materials and articles intended to come in contact with foodstuffs. Therefore adipic acid may be used as an additive in the manufacture of plastic materials and articles and a Specific Migration Limit (SML) does not apply.

Additional information on aliphatic carboxylic acids

In the context of the EPA HPV Challenge Program a dossier has been prepared on C6-C10 aliphatic aldehydes and carboxylic acids. This dossier is available on internet:

http://www.epa.gov/chemrtk/alipalde/c13033.pdf

The following text can be found on page 24:

Based on the lack of histopathology of reproductive organs in repeat dose studies, the lack of significant reproductive or developmental effects in the absence of maternal toxicity in two reproductive/developmental screening studies, and the lack of developmental or fetotoxicity in studies with structurally related carboxylic acids, it is concluded that members of this chemical category show no significant evidence of either reproductive or developmental toxicity.

Conclusions

Specific toxicological studies does not seem to be available for 6-hydroxyhexanoic acid or other hydroxyhexanoic acids. However, the toxicological properties of 6-hydroxyhexanoic acid can be predicted based on the chemical structure.

Information is available for analogues of 6-hydroxyhexanoic acid. In conclusion:

- Hexanol was not teratogenic to rats,
- For 1,6-hexanediol there is no indication of toxic effects on reproductive function or developmental toxicity,
- Adipic acid was not teratogenic and there is no reason to expect specific reproductive toxicity and
- Aliphatic carboxylic acids show no significant evidence of either reproductive or developmental toxicity.

Furthermore the analogues hexanoic acid and adipic acid are listed on Annex III of Commission Directive 2002/72/EC of 6 August 2002 and therefore they may be used as an additive in the manufacture of plastic materials and articles intended to come in contact with foodstuffs. It should also be realised that both substances are endogenous and therefore these substances are incorporated in metabolic cycles. Hexanoic acid is a food additive in the USA.

ε-Caprolactone is hydrolysed to 6-hydroxyhexanoic acid. Based on a comparison with analogues of this substance, it is concluded that 6-hydroxyhexanoic acid shows no evidence of reproductive or developmental toxicity.

S I D S

Dossier

Existing Chemical	:	ID: 502-44-3
CAS No.	:	502-44-3
EINECS Name	:	hexan-6-olide
EC No.	:	207-938-1
TSCA Name	:	2-Oxepanone
Molecular Formula	:	C6H10O2

Producer related part

Company	:	Solvay Interox S.A.
Creation date	:	30.05.1994

Substance related part

Company	:	Solvay Interox S.A.
Creation date	:	30.05.1994
Status	:	
Memo	:	JPE
Printing date	:	13.06.2005
Revision date	:	
Date of last update	:	13.06.2005
Number of pages	:	
Chapter (profile)	:	
Reliability (profile)	:	
Flags (profile)	:	

1.0.1 APPLICANT AND COMPANY INFORMATION

Type : lead organisation
Name : Solvay S.A.
Contact person : A.G. Berends
Date :
Street : Rue de Ransbeek 310
Town : 1120 Brussel
Country : Belgium
Phone : + 32 2 264 3398
Telefax : + 32 2 264 2990
Telex :
Cedex :
Email : albert.berends@solvay.com
Homepage : http://www.solvay.com

02.09.2003

Type : cooperating company
Name : BASF
Contact person : R. Parod
Date :
Street : 1609 Biddle Avenue
Town : 48192 Wyandotte, Michigan
Country : United States
Phone : + 1 734 324 6212
Telefax : + 1 734 324 5226
Telex :
Cedex :
Email : parodr@basf-corp.com
Homepage : http://www.basf.com

08.09.2003

Type : cooperating company
Name : Daicel Chemical Industries, Ltd.
Contact person : N. Ikeda
Date :
Street : 3-2-5, Kasumigaseki Chiyoda-ku
Town : 100-6077 Tokyo
Country : Japan
Phone : + 81 3 3507 3199
Telefax : + 81 3 3507 3191
Telex :
Cedex :
Email : no_ikeda@daicel.co.jp
Homepage : http://www.daicel.com

08.09.2003

Type : cooperating company
Name : The Dow Chemical Company
Contact person : W.M. Clous
Date :
Street : Bachtobelstrasse 3
Town : CH-8810 Horgen
Country : Switzerland

Phone : +41-1-7282708
Telefax : +41-1-7282096
Telex :
Cedex :
Email : wmclous@dow.com
Homepage : http://www.dow.com

11.11.2003

1.0.2 LOCATION OF PRODUCTION SITE, IMPORTER OR FORMULATOR

Remark : There are 4 production sites of epsilon-caprolactone:
- BASF, Freeport, Texas, USA.
- Daicel, Japan.
- Dow, Taft, Louisiana, USA.
- Solvay, Warrington, United Kingdom.

The consortium members are not aware of other production sites.

04.09.2003

1.0.3 IDENTITY OF RECIPIENTS

1.0.4 DETAILS ON CATEGORY/TEMPLATE

1.1.0 SUBSTANCE IDENTIFICATION

IUPAC Name : Hexano-6-lactone
Smiles Code : C1(=O)CCCCCO1
Molecular formula : C6H10O2
Molecular weight : 114
Petrol class : other: Not applicable

10.09.2003

1.1.1 GENERAL SUBSTANCE INFORMATION

Purity type : typical for marketed substance
Substance type : organic
Physical status : liquid
Purity : > 99.5 % w/w
Colour : colourless
Odour : characteristic but difficult to describe

Remark : The purity is at least 99.5 % but epsilon-caprolactone with a higher purity (e.g. 99.9 %) is marketed also.

05.04.2002

1.1.2 SPECTRA

1.2 SYNONYMS AND TRADENAMES

1,6-hexanolide

06.01.2004

2-oxepanone

30.05.1994

6-hexanolactone

30.05.1994

6-hydroxyhexanoic acid lactone

30.05.1994

caprolactone

31.05.1994

e-caprolactone

06.01.2004

epsilon-caprolactone

05.04.2002

hexan-6-olide

05.04.2002

hexanoic acid, epsilon-lactone

30.05.1994

1.3 IMPURITIES

Purity : typical for marketed substance
CAS-No : 7732-18-5
EC-No : 231-791-2
EINECS-Name : water
Molecular formula :
Value : < .005 % w/w

08.09.2003

Purity : typical for marketed substance
CAS-No : 124-04-9
EC-No : 204-673-3
EINECS-Name : adipic acid
Molecular formula :
Value : < .05 % w/w

08.09.2003

Purity : typical for marketed substance
CAS-No : 1191-25-9
EC-No :
EINECS-Name : 6-hydroxyhexanoic acid
Molecular formula :
Value : < .05 % w/w

08.09.2003

Purity : typical for marketed substance
CAS-No : 108-94-1
EC-No : 203-631-1
EINECS-Name : cyclohexanone
Molecular formula :
Value : < .05 % w/w

08.09.2003

Purity : typical for marketed substance
CAS-No : 109-52-4
EC-No : 203-677-2
EINECS-Name : valeric acid
Molecular formula :
Value : < .05 % w/w

08.09.2003

Purity : typical for marketed substance
CAS-No : 64-19-7
EC-No : 200-580-7
EINECS-Name : acetic acid
Molecular formula :
Value : < .05 % w/w

08.09.2003

1.4 ADDITIVES

Remark : Additives are not used for epsilon-caprolactone.
05.04.2002

1.5 TOTAL QUANTITY

Remark : The total quantity which was produced by the consortium members in 2003 was 40,000 - 60,000 tonnes.

29.06.2004

1.6.1 LABELLING

Labelling : provisionally by manufacturer/importer

Specific limits : no
Symbols : Xi, , ,
Nota : , ,
R-Phrases : (36) Irritating to eyes
S-Phrases : (26) In case of contact with eyes, rinse immediately with plenty of water and seek medical advice

04.08.2003

1.6.2 CLASSIFICATION

Classified : provisionally by manufacturer/importer
Class of danger : irritating
R-Phrases : (36) Irritating to eyes
Specific limits : no
05.04.2002

1.6.3 PACKAGING

1.7 USE PATTERN

Type of use : type
Category : Use resulting in inclusion into or onto matrix

31.05.1994

Type of use : industrial
Category : Chemical industry: used in synthesis

31.05.1994

Type of use : industrial
Category : Paints, lacquers and varnishes industry

31.05.1994

Type of use : industrial
Category : Polymers industry

31.05.1994

Type of use : use
Category : Intermediates

08.09.2003

Type of use : use
Category : Solvents

08.09.2003

Remark : About 50 % of the produced quantity is used on site for the production of polymers (polycaprolactones). The remaining 50 % is sold to customers

(downstream users).

The total number of downstream users is less than 1000. E-caprolactone is used to modify resins and polymers in order to enhance the performance of the end-products. It is capable of addition reactions with a range of functional groups such as OH, COOH and NH2. The majority is used for the modification of acrylic resins and polyesters, but it is also used for modification of epoxy resins and polyurethanes. A small quantity of e-caprolactone (< 1 %) is used as reactive diluent and as a solvent (e.g. for vinyl resins).

Based on the information available to the consortium members, e-caprolactone is not used in consumer products.

The information on use, presented in this section, is based on internal information from the consortium members. Published data could not be found.

29.06.2004

1.7.1 DETAILED USE PATTERN

1.7.2 METHODS OF MANUFACTURE

Origin of substance : Synthesis
Type : Production

Remark : Epsilon-caprolactone is manufactured using a process which utilises a high strength oxidising agent to produce a high purity peracetic acid. Peracetic acid is used to oxidise cyclohexanone by a Bayer-Villager reaction. The unreacted cyclohexanone is separated by distillation and recycled to the oxidation stage. Acetic acid is also recycled to the oxidation stage.

08.09.2003

1.8 REGULATORY MEASURES

1.8.1 OCCUPATIONAL EXPOSURE LIMIT VALUES

Remark : No occupational exposure limits have been set.
11.11.2003

1.8.2 ACCEPTABLE RESIDUES LEVELS

1.8.3 WATER POLLUTION

1.8.4 MAJOR ACCIDENT HAZARDS

1.8.5 AIR POLLUTION

1.8.6 LISTINGS E.G. CHEMICAL INVENTORIES

1.9.1 DEGRADATION/TRANSFORMATION PRODUCTS

1.9.2 COMPONENTS

1.10 SOURCE OF EXPOSURE

1.11 ADDITIONAL REMARKS

Memo : Submission to EFSA

Remark : e-Caprolactone is listed as a monomer in Section B of Commission Directive 2002/72/EC relating to the plastic materials and articles intended to come into contact with foodstuffs. To continue the use of this monomer, a dossier was submitted in 2004 to the European Food Safety Authority (EFSA) for re-evaluation of the substance. The dossier was submitted on behalf of the (4) consortium members (see section 1.0.1).

The EFSA opinion is available on internet:
http://www.efsa.eu.int/science/afc/afc_opinions/675_en.html

10.02.2005

1.12 LAST LITERATURE SEARCH

Type of search : Internal and External
Chapters covered : 3, 4, 5
Date of search : 31.03.2002

Remark : A literature search has been done in 1994 by the industry to prepare the IUCLID in the context of 'Council Regulation (EEC) No. 793/93 on the Evaluation and Control of the Risks of Existing Substances'. This IUCLID has been published by the European Chemicals Bureau.

An additional literature search has been done in March 2002 by Solvay. It covered the period 1994-2002. The following databases were used: AQUIRE, BIODEG, BIOLOG, CCRIS, CHRIS, DART/ETIC, DATALOG, EMIC, ENVIROFATE, GENETOX, GIABS, HSDB SUBSET, IRIS, MEDLINE, NIOSHTIC SUBSET , PHYTOTOX, RTECS, TERRETOX, TSCATS, TOXCENTER and TOXLINE.

04.09.2003

1.13 REVIEWS

2.1 MELTING POINT

Value	:	= -1.3 °C
Sublimation	:	
Method	:	
Year	:	1986
GLP	:	no
Test substance	:	no data
Reliability	:	(2) valid with restrictions Data from reliable handbook

29.04.2004 (47)

2.2 BOILING POINT

Value	:	= 237 °C at 1007.7 hPa
Decomposition	:	no
Method	:	OECD Guide-line 103 "Boiling Point/boiling Range"
Year	:	2004
GLP	:	yes
Test substance	:	as prescribed by 1.1 - 1.4
Method	:	The determination was carried out by differential scanning calorimetry (DSC).
Reliability	:	(1) valid without restriction

12.07.2004 (49)

Value	:	= 108 °C at 10 hPa
Decomposition	:	
Method	:	
Year	:	1986
GLP	:	no
Test substance	:	no data
Reliability	:	(2) valid with restrictions Data from reliable handbook

29.04.2004 (47)

2.3 DENSITY

Type	:	density
Value	:	= 1.07 g/cm³ at 20 °C
Method	:	
Year	:	1986
GLP	:	no
Test substance	:	no data
Reliability	:	(2) valid with restrictions Data from reliable handbook

29.04.2004 (47)

2.3.1 GRANULOMETRY

2.4 VAPOUR PRESSURE

Value	:	= .0081 hPa at 25 °C
Decomposition	:	no
Method	:	OECD Guide-line 104 "Vapour Pressure Curve"
Year	:	2004
GLP	:	yes
Test substance	:	as prescribed by 1.1 - 1.4
Remark	:	To determine the vapour pressure the isoteniscope system was used. Pressuring readings were done between 216 and 239 degrees Celsius.
Reliability	:	(1) valid without restriction

10.02.2005 (44)

Value	:	= .18 hPa at 25 °C
Decomposition	:	
Method	:	
Year	:	1989
GLP	:	no
Test substance	:	no data
Reliability	:	(3) invalid

13.06.2005 (5)

2.5 PARTITION COEFFICIENT

Partition coefficient	:	octanol-water
Log pow	:	= .68 at 20 °C
pH value	:	
Method	:	other (calculated)
Year	:	
GLP	:	no
Test substance	:	
Method	:	The Log Pow was estimated using the EPI (estimation program interface) Suite, developed by EPA's Office of Pollution Toxics and Syracuse Research Corporation.
Reliability	:	(2) valid with restrictions

29.04.2004 (11)

2.6.1 SOLUBILITY IN DIFFERENT MEDIA

Solubility in	:	Water
Value	:	at °C
pH value	:	= 5.7
concentration	:	94.9 other: % w/w at 20 °C
Temperature effects	:	
Examine different pol.	:	
pKa	:	at 25 °C
Description	:	of very high solubility
Stable	:	yes
Deg. product	:	not measured
Method	:	OECD Guide-line 105
Year	:	2004
GLP	:	yes

Test substance : as prescribed by 1.1 - 1.4

Remark : Miscible in all proportions at 19.5 - 20.5 degrees Celsius, based on the flask method.
Reliability : (1) valid without restriction
12.07.2004 (49)

2.6.2 SURFACE TENSION

2.7 FLASH POINT

Value : = 127 °C
Type : open cup
Method :
Year :
GLP : no
Test substance : no data

Reliability : (4) not assignable
13.07.2004 (42)

2.8 AUTO FLAMMABILITY

Value : = 204 °C at
Method :
Year :
GLP : no
Test substance : no data

Reliability : (4) not assignable
29.06.2004 (42)

2.9 FLAMMABILITY

2.10 EXPLOSIVE PROPERTIES

Result : not explosive
Method :
Year :
GLP : no
Test substance : no data

Reliability : (4) not assignable
29.06.2004 (42)

2.11 OXIDIZING PROPERTIES

Result : no oxidizing properties
Method :
Year :

GLP : no
Test substance : no data

Reliability : (4) not assignable
29.06.2004 (42)

2.12 DISSOCIATION CONSTANT

Remark : Based on the structural formula, e-caprolactone does not dissociate in water.

30.07.2003

2.13 VISCOSITY

Value : = 6.67 - mPa s (dynamic) at 20 °C
Result :
Method :
Year :
GLP : no
Test substance : no data

Reliability : (4) not assignable
29.06.2004 (42)

2.14 ADDITIONAL REMARKS

Remark : The Refractive Index at 20 degrees C is 1.4611
04.08.2003 (47)

3.1.1 PHOTODEGRADATION

Type	:	air
Light source	:	
Light spectrum	:	nm
Relative intensity	:	based on intensity of sunlight
DIRECT PHOTOLYSIS		
Halflife t1/2	:	ca. 1.7 day(s)
Degradation	:	% after
Quantum yield	:	
Deg. product	:	
Method	:	other (calculated)
Year	:	
GLP	:	no
Test substance	:	

Result : The AOP component of EPIWIN was used to calculate the rate of photodegradation for epsilon-caprolactone. The half-life was calculated to be 1.7 days. Based on the results of the model, there is no absorption of solar radiation by epsilon-caprolactone in the troposphere. The half-life of 1.7 days is based on a mean hydroxyl radical concentration of 1.5E6 cm3 over a 12-hour day.

Reliability : (2) valid with restrictions

29.04.2004 (8)

3.1.2 STABILITY IN WATER

Type	:	abiotic
t1/2 pH4	:	= 16 day(s) at 20 °C
t1/2 pH7	:	= 53 day(s) at 20 °C
t1/2 pH9	:	= 2.2 day(s) at 20 °C
t1/2 pH 1.2	:	= 0 day(s) at 37 °C
Deg. product	:	yes
Method	:	OECD Guide-line 111 "Hydrolysis as a Function of pH"
Year	:	2003
GLP	:	yes
Test substance	:	other TS

Method : ANALYTICAL METHOD: Samples were taken out of the flasks and 'H-NMR spectra of the samples were recorded. Selected integrals of the NMR signals of Epsilon-caprolactone and of 6-hydroxyhexanoic acid (degradation product) were used for the calculations of the concentrations of these compounds in the test solutions.
CALCULATION METHOD: For the calculation of Kobs and t1/2 first order kinetic is assumed in all experiments.

Result : RESULTS:
No other hydrolysis products than 6-hydroxyhexanoic acid could be observed in all of the conducted tests. Kobs and t1/2 due to the test conditions used are shown in the following table:

pH	T	Kobs (h-1)	t1/2 (h)
1.2 (1.2-1.1)'	37	1.7396	0.4
4.0 (4.0-3.7)'	37	0.0070	100
7.0 (7.0-6.8)'	37	0.0027	258

9.0 (9.0-8.7)'	37	0.0853/0.0844" 8.1/8.2"
4.0 (4.1-3.8)'	20	0.00184 376
7.0 (7.0-7.0)'	20	0.00055 1261
9.0 (9.0-8.7)'	20	0.0134 52

'First values in brackets measured at the start of the test, second values measured at the end of the test.
"At pH 9 and 37 degrees C a second test was performed.

The results show that the hydrolysis at 37 degrees Celsius is 4-5 times higher than the hydrolysis at 20 degrees Celsius.

6-Hydroxyhexanoic acid is not listed on the European Inventory of Existing Commercial Substances (EINECS). However, the CAS number of this substance is 1191-25-9. Toxicity data on this substance were not found. However, based on structural comparison the toxicological properties of this substance are expected to be similar to hexanoic acid and adipic acid.

Test condition : TEST TYPE
- Test medium: demineralized water
- Test system
  * Buffer pH 1.2 (potassium chloride / hydrochlorid acid);
  * Buffer pH 4.0 (potassium dihydrogenphosphate/ orthophosphoric acid);
  * Buffer pH 7.0 (Phosphate mixture);
  * Buffer pH 9.0 (sodium borate/hydrochloric acid)
- Concentration of test substance: 45 - 60 mg epsilon-caprolactone were dissolved in 50 ml of the corresponding buffer solutions.

TEST CONDITIONS: The closed glass flasks were thermostated in a laboratory oven at 37.0 +/- 0.1 degrees C and at 20.0 +/- 0.1 degrees C, respectively.

Test substance : TEST SUBSTANCE
- Supplier: Sigma-Aldrich
- Purity: 99.95 %

Reliability : (1) valid without restriction
GLP Guideline study

10.06.2005 (7)

Type : abiotic
t1/2 pH4 : at °C
t1/2 pH7 : at °C
t1/2 pH9 : at °C
t1/2 pH : > 16 day(s) at °C
Deg. product : no
Method : other: not described
Year : 1991
GLP : yes
Test substance : other TS

Result : RESULTS
Results of the analyses performed on the epsilon-caprolactone solutions were 100, 96.8, 83.2 and 79.2 % of the nominal concentration for days 0, 7, 14 and 16 respectively.

Test condition : TEST TYPE
- Test medium: Milli-Q water
- Concentration of test substance: 1000 micrograms/ml

MEASUREMENTS
- The test solution was analyzed on days 0, 7, 14 and 16 with GC/FID

Test substance : TEST SUBSTANCE

Supplier: Union Carbide Chemicals and Plastics Company Inc.
Purity: > 99.9 %

Reliability : (4) not assignable
Documentation insufficient for assessment

06.01.2004 (2)

3.1.3 STABILITY IN SOIL

3.2.1 MONITORING DATA

3.2.2 FIELD STUDIES

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

Type : volatility
Media : water - air
Air : % (Fugacity Model Level I)
Water : % (Fugacity Model Level I)
Soil : % (Fugacity Model Level I)
Biota : % (Fugacity Model Level II/III)
Soil : % (Fugacity Model Level II/III)
Method : other: QSAR estimation
Year :

Result : Henry's law constant (H): 3.62E-05 atm.m3/mol
= ca. 3.62 Pa.m3/mol

The Henry's law constant was calculated with the group method of Henrywin.

Probability of volatilization is limited.

Reliability : (2) valid with restrictions
10.02.2005 (10)

3.3.2 DISTRIBUTION

Media : other: air - water - soil
Method : Calculation according Mackay, Level III
Year :

Method : The environmental partitioning of e-caprolactone was estimated through Mackay Level III model (Version 2.2), developed by the Environmental Modelling Centre, Trent University, Canada. The following input values in the model were used:

Molecular weight : 114 g/mol
Data temperature : 25°C
Water solubility : 10,000 g/m3
Vapour pressure : 0.8 Pa
Log Kow : 0.68
Melting point : - 1.3°C
Half life in air (hours) : 41 (estimate)

Half life in water (hours) : 1261

All the other degradation rates were considered negligible.

An emission of 1000 kg/h in the water compartment was assumed. No emissions were considered in soil and air compartments.

The model default environmental parameters were chosen to carry out the analysis.

Result : The Mackay Level III model calculated the following distribution of e-caprolactone in the environment

Air	0.0001 %
Water	99.95 %
Soil	0.012 %
Sediment	0.042 %

Reliability : (2) valid with restrictions
12.07.2004 (6)

3.4 MODE OF DEGRADATION IN ACTUAL USE

3.5 BIODEGRADATION

Type : aerobic
Inoculum : activated sludge, domestic, non-adapted
Concentration : 10 mg/l related to Test substance
20 mg/l related to Test substance
Contact time : 28 day(s)
Degradation : (±) % after
Result : readily biodegradable
Control substance : Acetic acid, sodium salt
Kinetic : %
%
Deg. product : not measured
Method : OECD Guide-line 301 B "Ready Biodegradability: Modified Sturm Test (CO2 evolution)"
Year : 1993
GLP : yes
Test substance : other TS

Result : RESULTS

Test subst. conc.	% biodegradability 7 days	14 days	28 days
10 mg/l	43 %	76 %	100 %
20 mg/l	26 %	47 %	58 %

Epsilon-caprolactone can be classified as readily biodegradable according to the results of the study: >60 % biodegradation within 14 days at a concentration of 10 mg/l and 47 % biodegradation within 14 days at a concentration of 20 mg/l. At 10 mg/l the biodegradation was 43 % after 7 days (no lag phase) and 76 % after 14 days which means obviously that the 10-day window was passed. The activity of the inoculum was sufficient: sodium acetate was degraded for more than 60 % within 28 days.

Test condition : INOCULUM

- Source: RWZI Horstermeer, Nederhorst den Berg, The Netherlands
- Initial cell concentration: the concentration of the inoculum was 30 mg dry weight/l.

TEST SYSTEM
- Culturing apparatus: 2 litre glass bottles closed with a plastic screw cap. In each bottle a 15 ml plastic tube with holes was suspended from the screw cap. A vial with 5 ml 1 M KOH was placed in this tube.
- Number of culture flasks per concentration: 2
- Aeration device: Each bottle was aerated with CO2-free air in the dark.
- Measuring equipment: the amount of CO2 absorbed was determined by titration of the residual amount of KOH with 0.5 M HCl with a Titrino 702-SM, Metrohm titrator.
- Blank measurements were included in the test.

TEST CONDITIONS
- Composition of the medium: mineral salt solution according to OECD Guideline 301 B
- Test temperature: 20 degrees C
- pH value: 6.3-7.4
- Aeration of dilution water: yes

Test substance : TEST SUBSTANCE
- Supplier: Solvay Interox S.A.
- Purity: > 99 %

Reliability : (1) valid without restriction
GLP Guideline study

10.06.2005 (20)

Type : aerobic
Inoculum : domestic sewage, non-adapted
Contact time : 20 day(s)
Degradation : > 60 (±) % after 14 day(s)
Result : readily biodegradable
Control substance : Acetic acid, sodium salt
Kinetic : %
%
Deg. product : not measured
Method : other
Year : 1974
GLP : no
Test substance : other TS

Result : RESULTS

% biodegradation of test substance			
5 days	10 days	15 days	20 days
56%	58%	69%	79%

According to the authors of the study, epsilon-caprolactone can be classified as readily biodegradable based on the results of the study: >60 % biodegradation within 14 days. E-Caprolactone was tested in a biodegradation testing program with > 300 other chemicals. One of the chemicals tested also was acetic acid, which showed 96% degradation in 20 days, showing the activity of the inoculum was sufficient.

Test condition : INOCULUM
- Source: Domestic treatment plant. Unacclimated, unadapted
- Initial cell concentration: not indicated.

TEST SYSTEM

- No details given, however measuring Biological Oxygen Demand and relating this to the calculated Theoretical Oxygen Demand (ThOD).

TEST CONDITIONS
- No details given. The ThOD was used to approximate the concentration of the stock solution to be employed for the biochemical oxygen demand (BOD) study. The concentration of the stock solution was selected such that the 2- and 5-ml sample sizes used in the study would provide an oxygen demand ranging from 5 to 12 mg/l. This to ensure that a consumption of oxygen would occur (when biodegradable) that could be measured with suitable degree of accuracy.

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: Not indicated

Reliability : (4) not assignable
Documentation insufficient for assessment

29.06.2004 (45)

3.6 BOD5, COD OR BOD5/COD RATIO

3.7 BIOACCUMULATION

BCF	:	= 3.16
Elimination	:	
Method	:	other: QSAR estimation
Year	:	
GLP	:	no
Test substance	:	

Method : The bioconcentration was estimated using the EPI (estimation program interface) Suite, developed by EPA's Office of Pollution Toxics and Syracuse Research Corporation.

Reliability : (2) valid with restrictions

29.04.2004 (9)

3.8 ADDITIONAL REMARKS

4.1 ACUTE/PROLONGED TOXICITY TO FISH

Type : static
Species : Poecilia reticulata (Fish, fresh water)
Exposure period : 96 hour(s)
Unit : mg/l
NOEC : = 250
LC50 : = 295
LC100 : = 500
Limit test : no
Analytical monitoring : yes
Method : OECD Guide-line 203 "Fish, Acute Toxicity Test"
Year : 1992
GLP : yes
Test substance : other TS

Method : DEVIATIONS FROM GUIDELINE: No
STATISTICAL METHODS: The LC50 (96h) was calculated using an adapted logistic regression and a probit analysis model (PROBIT of SAS). The profile likelihood method was used to calculate the 95% confidence interval for the LC50. The NOEC was assessed at the highest concentration that did not cause a statistically significant difference with the controls.
ANALYTICAL METHODS: Samples of the test solutions were analysed with HPLC-analysis.

Result : RESULTS EXPOSED AND CONTROLS

Nominal test concentration (mg/l)	Mean measured concentrations (mg/l)	Mortality (%) during test
0	< 1	0
31	31	0
62	60	0
125	119	0
250	240	10
500	496	100
1000	1037	100

The guppies exposed to 1000 mg/l died within 24 hours and all guppies exposed to 500 mg/l died within 72 hours. Before these fishes died they showed uncontrolled movement and hypoactivity. At 250 mg/l one fish died without showing any sign of intoxication. At lower concentrations no effects were observed.

Based on the adapted logistic regression the LC50 was 295 mg/l with a 95% confidence interval of 251 to 392 mg/l. Based on probit analysis the LC50 was 280 mg/l.

Test condition : TEST ORGANISMS
- Source/supplier: RASBORA, Veenendaal, The Netherlands
- Age/size/loading: no data/2.1-3.2 cm/0.9 g/l
- Feeding: each day with troutfeed and waterfleas
- Pretreatment: no
- Feeding during test: no

STOCK AND TEST SOLUTION AND THEIR PREPARATION
- Vehicle/solvent: epsilon-caprolactone was dissolved in medium

REFERENCE SUBSTANCE
- Once a year a test with potassium dichromate is conducted. The EC50 (96h) found in the most recent reference test was 135 mg/l which is valid.

DILUTION WATER
- Source: Synthetic fresh water (ISO-water)
- pH: 7.8
- Hardness: ca. 250 mg/l

TEST SYSTEM
- Nominal concentrations: 0, 31, 62, 125, 250, 500 and 1000 mg/l
- Renewal of test solutions: No
- Exposure vessel type: Aquaria (glass) with a volume of 3 litres
- Number of replicates, fish per replicate: 1/10
- Test temperature: 21.3-22.5 degrees C
- Dissolved oxygen: 7.3-8.4 mg/l
- pH: 7.5-7.8
- Photoperiod: 16h light and 8h dark

TEST PARAMETER: mortality and abnormalities like hyperactivity, hypoactivity, hyperventilation, uncontrolled movement, loss of equilibrium and discolouring.

MONITORING OF TEST SUBSTANCE CONCENTRATION: Yes, at test initiation, at 48 and 96 hours after test initiation and when all fish in an aquarium had died.

Test substance : TESTSUBSTANCE
SOURCE: Solvay Interox S.A.
PURITY: > 99 %

Reliability : (1) valid without restriction
GLP Guideline study

10.06.2005 (12) (43)

Type : static
Species : Pimephales promelas (Fish, fresh water)
Exposure period : 96 hour(s)
Unit : mg/l
LC50 : = 320
Limit test : no
Analytical monitoring : no
Method : other: no details given
Year : 1974
GLP : no
Test substance : other TS

Method : STATISTICAL METHODS: no details given
ANALYTICAL METHODS: No analysis was done

Remark : Study was performed before official guidelines and GLP were in place. The study was performed however according to common standards employed at that time for ecotoxicity testing and is reasonably documented.

Result : RESULTS EXPOSED AND CONTROLS
- LC50 (24 hr) = 370 mg/l
- LC50 (48 hr) = 320 mg/l
- LC50 (96 hr) = 320 mg/l

Test condition : TEST ORGANISMS
- Source/supplier: commercial supplier
- Age/size/loading: adult/2.5-5 cm/10 fish per 18 L
- Feeding: no details given
- Pretreatment: no
- Feeding during test: no details given

STOCK AND TEST SOLUTION AND THEIR PREPARATION

- No details given
REFERENCE SUBSTANCE
- The study was part of a testing program with 217 chemicals (all reported in the same report). However, a reference substance was not used.
DILUTION WATER
- pH: 7.2 - 7.6
- Hardness: ca. 30 - 60 mg/l
TEST SYSTEM
- Renewal of test solutions: No
- Exposure vessel type: Aquaria (glass) with a volume of 18 litres
- Number of replicates, fish per replicate: 1/10
- Test temperature: 71 - 76 degrees Fahrenheit
- Dissolved oxygen: 7.5 - 9.0 mg/l
- pH: 7.2 - 7.6
TEST PARAMETER: mortality

Test substance : TEST SUBSTANCE
- Source: Union Carbide Corporation
- Purity: not indicated

Reliability : (2) valid with restrictions
Comparable to guideline study with acceptable restrictions

10.05.2004 (46)

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

Type : static
Species : Daphnia magna (Crustacea)
Exposure period : 48 hour(s)
Unit : mg/l
EC0 : = 125
EC50 : = 204
EC100 : = 500
Limit Test : no
Analytical monitoring : yes
Method : OECD Guide-line 202
Year : 2003
GLP : yes
Test substance : other TS

Method : DEVIATIONS FROM GUIDELINE: No
STATISTICAL METHODS: For calculation of the EC50 the probit method ($p < 0.05$) was used
ANALYTICAL METHODS: Samples were analysed with GC/FID-analysis

Result : RESULTS EXPOSED AND CONTROL:

Nom. Conc. (mg/l)	No. of mobile daphnids 24 h	48 h
0	20	20
62.5	20	20
125	20	19
250	13	5
500	0	0
1000	0	0

The EC50 (48h) was 204 mg/l with 95 % confidence limits of 172-240 mg/l.
ANALYTICAL RESULTS: The recovery of the samples analysed at t=0 h

ranged from 96.2-106 %, the recovery of the samples at t=48 h ranged from 95.0-99.8 %. As the recovery rate was >80 % no correction of the nominal concentrations was necessary.

Test condition : TEST ORGANISMS
- Source/Supplier: Institut National de Recherche Chimique Appliquee, France
- Age: 2-24 h (starting with the 3rd breed of parent animals)
- Feeding during test: No

STOCK AND TEST SOLUTION AND THEIR PREPARATION
- 500.1 mg epsilon-caprolactone was stirred in 500 ml M4 medium for about 10 minutes at 20 degrees C resulting in a stock solution of 1000 mg/l.

REFERENCE SUBSTANCE
- The EC50 (24h) in the most recent test with potassium dichromate was 1.37 mg/l which is valid.

DILUTION WATER
- Source: Synthetic fresh water (M4 medium)
- Alkalinity: 0.87 mmol/l
- Hardness: 2.42 mmol/l

TEST SYSTEM
- Concentrations: 0, 62.5, 125, 250, 500 and 1000 mg/l
- Renewal of test solution: No
- Exposure vessel type: Test tubes (glass) with flat bottom (nominal volume 20 ml)
- Number of replicates/individuals per replicate: 4/5
- Test temperature: 19.6-20.1 degrees C
- Dissolved oxygen: 7.1-9.1 mg/l
- pH: 7.8-8.1
- Intensity of irradiation: about 1-8 microE/(m2.s) at a wavelength of 400-750 nm
- Photoperiod: 16 hours light, 8 hours dark

TEST PARAMETER: Mobility (swimming ability) of the test animals
MONITORING OF TEST SUBSTANCE CONCENTRATION: Yes, at the begin (after 0 h) samples from vessels without daphnids were analysed. At the end of the test (after 48 h) samples from vessels with daphnids were analysed.

Test substance : TESTSUBSTANCE
SOURCE: Sigma-Aldrich, Germany
PURITY: 99.95 %

Reliability : (1) valid without restriction
GLP guideline study

03.05.2004 (15)

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

Species	:	Scenedesmus subspicatus (Algae)
Endpoint	:	biomass
Exposure period	:	72 hour(s)
Unit	:	mg/l
NOEC	:	= 256
LOEC	:	= 640
EC10	:	= 484
EC50	:	= 1217
EC90	:	> 4000
Limit test	:	no
Analytical monitoring	:	yes
Method	:	OECD Guide-line 201 "Algae, Growth Inhibition Test"

Year : 2003
GLP : yes
Test substance : other TS

Method : DEVIATIONS FROM GUIDELINE: No
STATISTICAL METHODS: The EC values were calculated (linear regression) from the concentration-response relationship.
The LOEC is determined by comparing the calculated biomass or growth rate of the various concentration levels with the control. The Dunnett's One-tailed T test is carried out at a 95 % significance level. The NOEC is the tested concentration immediately below the LOEC.
ANALYTICAL METHODS: Samples were analysed with GC/FID-analysis

Result : RESULTS EXPOSED AND CONTROL:

Nom. Conc. (mg/l)	Inhibition of biomass (%)	Inhibition of growth rate (%)
0	0	0
102	-2.9	1.4
256	-10.3	-1.4
640	19.0	7.5
1600	63.2	31.4
4000	88.4	66.1

ANALYTICAL RESULTS: The analytical results yielded 80 % or higher recoveries; they varied between 91.6 and 105.8 % of the nominal concentrations at test initiation and between 82.0 to 98.6 % at test termination.

Test condition : TEST ORGANISMS
- Source/supplier: SAG (Collection of algal cultures in Gottingen)
- Pretreatment: A pre-culture was incubated for 3 days at 23 +/- 2 degrees C.
- Controls: yes
- Initial cell concentration: 1 x 10E+4

REFERENCE SUBSTANCE
- The EbC50 (72h) of the last control experiment with potassium dichromate was 0.47 mg/l which was valid.

TEST MEDIUM CHEMISTRY
- Test medium: synthetic medium

TEST SYSTEM
- Concentrations: 0, 102, 256, 640, 1600, 4000 mg/l
- Renewal of test solutions: no
- Exposure vessel type: Erlenmeyer flasks (nominal volume 250 ml) plugged with gas permeable siliconsponge caps
- Number of replicates: 3
- Test temperature: 23 +/- 2 degrees C
- pH: 6.3-8.3
- Intensity of irradiation: About 60 - 120 microE/(m2.s) at a wave length of 400-700 nm
- Photoperiod: Continuous illumination

TEST PARAMETER: In vivo chlorophyll-a-fluorescence (pulsed excitation with light flashes having a wavelength of 435 nm)
MEASUREMENTS: Measurement of fluorescence after 0, 24, 48 and 72 hours. Chemical analysis was performed in duplicate in all test solutions at 0 and 72 hours.

Test substance : TESTSUBSTANCE
SOURCE: Sigma-Aldrich, Germany
PURITY: 99.95 %

Reliability : (1) valid without restriction

GLP Guideline study

02.09.2003 (48)

4.4 TOXICITY TO MICROORGANISMS E.G. BACTERIA

Type	:	aquatic
Species	:	Pseudomonas putida (Bacteria)
Exposure period	:	16 hour(s)
Unit	:	mg/l
NOEC	:	= 32
EC50	:	= 1260
Analytical monitoring	:	yes
Method	:	other: ISO/TC 147/SC 5/WG 1 N 133
Year	:	1992
GLP	:	yes
Test substance	:	other TS

Method : DEVIATIONS FROM GUIDELINE: No
STATISTICAL METHODS: The EC values were calculated with linear regression from the concentration-response relationship. The NOEC was determined with William's test, one sided.
ANALYTICAL METHODS: Samples of the stock solutions were taken at test initiation of three tests and the total organic carbon content was determined. The results were used to derive calculated test concentrations.

Result : RESULTS EXPOSED AND CONTROLS:

Calculated test conc. (mg/l)	Inhibition of cell multiplication (%)
0	0
16	5
32	9
63	12
71	20
126	18
143	23
253	22
285	33
303	24
570	40
605	36
1140	55
1210	48

Significant inhibition of cell multiplication occurred at test concentrations of 63 mg/l and higher. The EC50 was 1260 mg/l. Although the inhibition at 32 mg/l was statistically significant, 32 mg/l is considered to be the NOEC, as an inhibition less than 10 % has no biological significance.
ANALYTICAL RESULTS: The concentrations found in the stock solutions were slightly above the nominal level of 1250 mg epsilon-caprolactone/l. Based on the analysis of the stock solutions, calculated test concentrations were derived.

Test condition : TEST ORGANISMS
- Strain: Pseudomonas putida (ATCC 1263)
- Source/supplier: Technical University, Delft, The Netherlands
- Pretreatment: The culture was freeze dried and stored at

4 degrees C. The bacteria were resuspended in deionised water at the beginning of the experiment
- Controls: Yes
REFERENCE SUBSTANCE
- Once a year a test with 3,5-dichlorophenol is conducted. The EC50.16h found in this test was 21.2 mg/l which was valid.
TEST MEDIUM CHEMISTRY
- Test medium: synthetic nutrient medium
- Dilution water: deionised water "Ministil"
TEST SYSTEM
- Concentrations: 0, 16, 31, 63, 125, 250, 500 and 1000 mg/l
- Renewal of test solutions: No
- Exposure vessel type: 250-ml Erlenmeyer flasks containing 100 ml test solution
- Number of replicates: 5 test vessels per concentration. The test was carried out 4 times.
- Test temperature: 21-23 degrees C
- pH: 6.8-6.9 at test initiation to 5.7-6.0 at the end of the test
- Photoperiod: 24 hours dark
TEST PARAMETER: Absorption at 600 nm
MEASUREMENTS: The absorption was measured after 16 hours. The stock solution with a concentration of 1250 mg/l was analysed.

Test substance : TESTSUBSTANCE
SOURCE: Solvay Interox S.A.
PURITY: > 99 %

Reliability : (1) valid without restriction
GLP Guideline study

10.06.2005 (19)

4.5.1 CHRONIC TOXICITY TO FISH

4.5.2 CHRONIC TOXICITY TO AQUATIC INVERTEBRATES

4.6.1 TOXICITY TO SEDIMENT DWELLING ORGANISMS

4.6.2 TOXICITY TO TERRESTRIAL PLANTS

4.6.3 TOXICITY TO SOIL DWELLING ORGANISMS

4.6.4 TOX. TO OTHER NON MAMM. TERR. SPECIES

4.7 BIOLOGICAL EFFECTS MONITORING

4.8 BIOTRANSFORMATION AND KINETICS

4.9 ADDITIONAL REMARKS

Memo : Toxicity of 6-hydroxyhexanoic acid

Remark : 6-Hydroxyhexanoic acid is formed when e-caprolactone is hydrolysed. 6-Hydroxyhexanoic acid is not listed on the European Inventory of Existing Commercial Substances (EINECS). However, the CAS number of this substance is 1191-25-9. Ecotoxicity data on this substance were not found. However, based on structural comparison the toxicological properties of this substance are expected to be similar to hexanoic acid and adipic acid.

Ecotoxicity data are available for the analogue adipic acid (OECD, 2004). The acute EC50 values for fish (Danio rerio) and water flea (Daphnia magna) were > 1000 and 86 mg/l, respectively. In an algae growth inhibition test with Desmodesmus subspicatus the 96 h-EbC50 of adipic acid was 27 mg/l and the 72 h-EbC50 was 31 mg/l.

10.06.2005 (31)

5.0 TOXICOKINETICS, METABOLISM AND DISTRIBUTION

Remark : e-Caprolactone is rapidly hydrolysed in the stomach because the half-life at a pH of 1.2 and a temperature of 37 °C is 0.4 hours (see section 3.1.2). Hydrolysis of e-caprolactone results in the formation of 6-hydroxyhexanoic acid. Specific toxicological studies with 6-hydroxyhexanoic acid could not be found in the literature but information on analogues can be found in the Annex of the SIAR and more briefly in section 5.8.3 of this IUCLID. e-Caprolactone is not only hydrolysed at low pH but is also hydrolysed in the blood. Billecke et al. (2000) reported that human serum paraoxonase (PON1) isozymes Q and R are able to hydrolyse a large group of different lactones including e-caprolactone. gamma-Butyrolactone (CAS No. 96-48-0) has a similar structure as e-caprolactone but has only 4 instead of 6 carbon atoms. gamma-Butyrolactone was rapidly hydrolysed by an enzyme found in the blood and liver and the half-life of the conversion was less than 1 minute (NTP, 1996). For this reason e-caprolactone is expected to be hydrolysed rapidly in the blood.

10.06.2005 (3) (30)

5.1.1 ACUTE ORAL TOXICITY

Type : LD50
Value : > 2000 mg/kg bw
Species : rat
Strain : Wistar
Sex : male/female
Number of animals : 10
Vehicle : other: 1.25% gum tragacanth solution in distilled water
Doses : 2000 mg/kg
Method : Directive 84/449/EEC, B.1 "Acute toxicity (oral)"
Year : 1984
GLP : yes
Test substance : other TS

Method : STATISTICAL METHOD: Not applicable
Result : MORTALITY:
None of the male rats died within the 14-day observation period, but 2 of the 5 females were found dead on day 2.
CLINICAL SIGNS:
Clinical signs observed were mostly indicative of effects on motor coordination (decreased locomotor activity, abnormal gait and posture, loss of righting reflex), on muscle tone (changes in body and limb tone), on autonomic nervous system (decreased respiratory rate, respiratory difficulties) and on central nervous system (apathy, changes in startle position). Time of onset of the first signs was within 30 minutes after dosing. All signs had disappeared within 3 days.
NECROPSY FINDINGS:
The observations did not reveal any macroscopic abnormalities.
SEX-SPECIFIC DIFFERENCES:
Female rats were more affected than males.
Test condition : TEST ORGANISMS
- Source: Harlan/CPB, Zeist, The Netherlands
- Age: not described
- Weight at study initiation: 175-200 g (males), 150-175 g (females)

- Controls: No
ADMINISTRATION
- Doses per time period: Single dose
- Volume administered or concentration: 10 ml/kg by gavage
- Post dose observation period: 14 days
EXAMINATIONS: The animals were weighed one day before dosing, at the day of dosing and at 2, 7 and 14 days after treatment. Any sign of intoxication occurring during the 14-day observation period was recorded. Gross post-mortem examination was done on rats that died during the observation period and on those surviving to the end of the 14-day observation period.

Test substance : TEST SUBSTANCE
Supplier: Solvay S.A., Brussels, Belgium
Purity: > 99 %

Reliability : (1) valid without restriction
GLP guideline study

05.09.2003 (41)

Type : LD50
Value : = 4290 mg/kg bw
Species : rat
Strain : other: Carworth-Wistar
Sex : male
Number of animals : 15
Vehicle : water
Doses : 2000, 4000 and 8000 mg/kg bw
Method : other
Year : 1953
GLP : no
Test substance : other TS

Method : STATISTICAL METHOD: Thompson's method of calculating the median-effective dose (LD50) was applied to the 14-day mortality data.

Result : EXPOSED AND CONTROLS
An LD50 of 4290 mg/kg with confidence intervals of 3070-5980 mg/kg was found.
CLINICAL SIGNS: Symptoms apparent within 4 hours after dosing included narcosis, prostration, ruffed coats and sluggishness
NECROPSY FINDINGS: Autopsies of those dying revealed congestion of the lungs, mottling of livers, paleness of kidneys and gastrointestinal tract irritation and burning.

Test condition : TEST ORGANISMS
- Age: 5 to 6 weeks
- Weight at study initiation: 90-120 g
- Controls: No
ADMINISTRATION
- Doses per time period: Single dose
- Volume administered or concentration: A 10 % aqueous dilution by stomach tube
- Post dose observation period: 14 days

Test substance : TEST SUBSTANCE
Source: S. Charleston under identification 242 RD 89
Purity: No data

Reliability : (2) valid with restrictions
Comparable to guideline study with acceptable restrictions.
Note: This study is from the pre-guideline and pre-GLP era (1953). However since acute toxicity testing methodology has not significantly changed since, and for the animal welfare reasons, this study should get reliability score 2.

10.06.2005 (40)

5.1.2 ACUTE INHALATION TOXICITY

Type	:	other
Value	:	
Species	:	rat
Strain	:	no data
Sex	:	male
Number of animals	:	6
Vehicle	:	no data
Doses	:	
Exposure time	:	8 hour(s)
Method	:	other
Year	:	1953
GLP	:	no
Test substance	:	other TS
Result	:	RESULTS No mortality was observed. The only notable response was slight skin irritation.
Test condition	:	TEST ORGANISMS - No data ADMINISTRATION - Saturated vapor, generated at room temperature by passing air at 2.5 liters/minute through a fritted glass disc immersed in 50 ml of the test substance. EXAMINATIONS - The animals were observed for a total period of 14 days.
Test substance	:	TEST SUBSTANCE Source: S. Charleston under identification 242 RD 89 Purity: No data
Reliability	:	(4) not assignable Documentation insufficient for assessment

10.06.2005 (40)

5.1.3 ACUTE DERMAL TOXICITY

Type	:	LD50
Value	:	= 5990 ml/kg bw
Species	:	rabbit
Strain	:	New Zealand white
Sex	:	male
Number of animals	:	8
Vehicle	:	other: undiluted test substance
Doses	:	5000 and 10000 mg/kg bw
Method	:	other
Year	:	1953
GLP	:	no
Test substance	:	other TS
Method	:	DEVIATIONS FROM GUIDELINE: Not described STATISTICAL METHOD: Thompson's method of calculating the median-effective dose (LD50) was applied to the 14-day mortality data.
Result	:	MORTALITY The LD50 for the undiluted compound is 5990 (4270-8420) ml/kg CLINICAL SIGNS: Skin erythema is produced which may or may not result in necrosis and desquamation NECROPSY FINDINGS: Autopsies revealed congested or hemorrhagic

lungs, and extremely congested livers.

Test condition : TEST ORGANISMS
- Age: 3 to 5 months
- Weight at study initiation: 2.5 kg

ADMINISTRATION
- Occlusion: A polyethylene sheeting was used to retain the dose in contact with the clipped skin of the trunk.
- Removal of test substance: Yes, after 24 hours the test substance was removed.

EXAMINATIONS
- The animals were observed for a total period of 14 days.

Test substance : TEST SUBSTANCE
Source: S. Charleston under identification 242 RD 89
Purity: No data

Reliability : (2) valid with restrictions
Comparable to guideline study with acceptable restrictions. Note: This study is from the pre-guideline and pre-GLP era (1953). However since acute toxicity testing methodology has not significantly changed since, and for animal welfare reasons, this study should get reliability score 2.

10.06.2005 (40)

5.1.4 ACUTE TOXICITY, OTHER ROUTES

Type : LD50
Value : = 1255 mg/kg bw
Species : mouse
Strain : ICR
Sex : male/female
Number of animals : 40
Vehicle : water
Doses : 800, 1000, 1200, 1400, 2000, 3000 mg test article/kg
Route of admin. : i.p.
Exposure time : 72 hour(s)
Method : Other
Year : 1997
GLP : yes
Test substance : other TS

Method : STATISTICAL METHOD: The LD50 (3 days) was calculated by probit analysis.

Remark : This study was performed as a preliminary experiment to set dose levels for the mouse micronucleus assay.

Result : RESULTS

Conc. (mg/kg)	Mortality (No. animals dead/total no. tested) Male	Female	Total
800	0/5	0/5	0/10
1000	0/5	0/5	0/10
1200	2/5	2/5	4/10
1400	5/5	5/5	10/10
2000	5/5	5/5	10/10
3000	5/5	5/5	10/10

Mortality occurred within three hours of dose administration in 5/5 males and 5/5 females at 1400, 2000 and 3000 mg/kg. Mortality occurred within two days of dose administration in 2/5 males and 2/5 females at 1200 mg/kg. Clinical signs, which were noted following dose administration,

included lethargy in male and female mice at 800, 1000 and 1200 mg/kg and gasping and convulsions in male and female mice at 1400, 2000 and 3000 mg/kg.

Test condition : TEST ORGANISMS
- Source: Harlan Sprague Dawley, Inc., Frederick, MD.
- Age: 6 to 8 weeks at test initiation
- Weight at study initiation: 33.8-38.8 g (males), 26.3-27.5 g (females)
- No. of animals per dose: 10 (5 per sex)

ADMINISTRATION
- Vehicle: sterile distilled water
- Frequency of treatment: single dose
- Volume applied: 20 ml test article-vehicle mixture/kg body weight
- Post dose observation period: 14 days

EXAMINATIONS: Body weights were recorded prior to dose administration and 1 and 3 days after dose administration. Mice were observed after dose administration and daily thereafter for 3 days for clinical signs of chemical effect.

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: 100 %

Reliability : (1) valid without restriction
GLP guideline study, test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

09.09.2003 (33)

Type : LD50
Value : = 1300 mg/kg bw
Species : mouse
Strain : Swiss Webster
Sex : male
Number of animals :
Vehicle :
Doses :
Route of admin. : i.p.
Exposure time : 24 hour(s)
Method : other
Year : 1979
GLP : no
Test substance : no data

Reliability : (4) not assignable
Secondary literature

10.06.2005 (38)

5.2.1 SKIN IRRITATION

Species : rabbit
Concentration : .5 g
Exposure : Open
Exposure time : 4 hour(s)
Number of animals : 3
Vehicle : other: undiluted test substance
PDII :
Result : not irritating
Classification : not irritating
Method : OECD Guide-line 404 "Acute Dermal Irritation/Corrosion"

Year : 1991
GLP : yes
Test substance : other TS

Result : AVERAGE SCORE
- Erythema: No erythema were observed in the three rabbits on 30 minutes, 24, 48 and 72 hours after removal of the patch.
- Oedema: No oedema were observed in the three rabbits on 30 minutes, 24, 48 and 72 hours after removal of the patch.

Test condition : TEST ANIMALS
- Strain: SPF-derived New Zealand White
- Sex: Male
- Source: Harlan Olac, Zeist, The Netherlands
- Weight at study initiation: 2.5-3.0 kg
- Controls: No

ADMINISTRATION/EXPOSURE
- Preparation of test substance: undiluted test material
- Area of exposure: 6 cm2
- Postexposure period: 72 hours

EXAMINATIONS
- Scoring system: According to OECD Guideline 404
- Examination time points: At 30-60 minutes and at 24, 48 and 72 hours after patch removal, the skin reactions were scored.

Test substance : TEST SUBSTANCE
Supplier: Solvay S.A., Brussels, Belgium
Purity: > 99 %

Reliability : (1) valid without restriction
GLP Guideline study

02.09.2003 (17)

Species : rabbit
Concentration : undiluted
Exposure : Open
Exposure time : 24 hour(s)
Number of animals : 5
Vehicle : other: undiluted test substance
PDII :
Result : slightly irritating
Classification :
Method : other
Year : 1953
GLP : no
Test substance : no data

Result : AVERAGE SCORE: Grade 1 of 6; slight irritation
REVERSIBILITY: Not described
OTHER EFFECTS: Not described

Test condition : TEST ORGANISMS
- No data

ADMINISTRATION/EXPOSURE
- Preparation of test substance: 0.01 ml of undiluted test substance was applied
- Postexposure period: 24 hours

EXAMINATIONS
- Scoring system: Grade 1-6; Grade 1 indicates the least visible capillary injection from undiluted test material, grade 6 indicates necrosis.

Test substance	:	TEST SUBSTANCE Source: S. Charleston under identification 242 RD 89 Purity: No data
Reliability	:	(3) invalid Significant methodological deficiencies.
10.06.2005		(40)

Species	:	rabbit
Concentration	:	500 mg
Exposure	:	Occlusive
Exposure time	:	24 hour(s)
Number of animals	:	
Vehicle	:	no data
PDII	:	
Result	:	moderately irritating
Classification	:	
Method	:	other: no data
Year	:	1986
GLP	:	no
Test substance	:	no data

Reliability	:	(4) not assignable Original reference not available
02.09.2003		(25)

5.2.2 EYE IRRITATION

Species	:	rabbit
Concentration	:	undiluted
Dose	:	.1 ml
Exposure time	:	
Comment	:	not rinsed
Number of animals	:	4
Vehicle	:	none
Result	:	irritating
Classification	:	irritating
Method	:	OECD Guide-line 405 "Acute Eye Irritation/Corrosion"
Year	:	1991
GLP	:	yes
Test substance	:	other TS

Result	:	DESCRIPTION OF LESIONS: At 1 hour very slight opacity, iritis, redness, very slight chemosis and severe discharge were observed in three out of four animals. Slight iritis, redness and chemosis were observed in the other animal. At 24 hours slight opacity and iritis were observed in all animals. Slight redness was observed in one animal and moderate redness in two animals. At 48 hours opacity was slight in 2 animals and very slight in one animal. Iritis was slight in all animals. At 72 hours slight opacity and slight iritis were observed in two animals. Very slight opacity was observed in the other animal. Slight redness was observed in two animals and moderate redness was observed in the other animal. REVERSIBILITY: All findings had disappeared at day 7.
Test condition	:	TEST ANIMALS - Strain: SPF-derived New Zealand White - Sex: male - Source: Harlan Olac, Zeist, The Netherlands - Weight at study initiation: 1.8-2.2 kg (one animal),

2.5-3.0 kg (three animals)
- Number of animals: 4. During the dosing, one animal broke its' back due to struggling in the animal restrainer. After reading the scores at 1 hour after application of the test material, this animal was killed for ethical reasons and a fourth animal was assigned to the study.
- Controls: The right eye, remaining untreated, served as control

ADMINISTRATION/EXPOSURE
- Preparation of test substance: The test substance was administered undiluted
- Amount of substance instilled: 0.1 ml

EXAMINATIONS
- Ophtalmoscopic examination: ocular reactions were examined
- Scoring system: method of Draize
- Observation period: Readings of reactions were made in all rabbits at 1, 24, 48, 72 hours and 7 days after treatment.

Test substance : TEST SUBSTANCE
Supplier: Solvay S.A., Brussels, Belgium
Purity: > 99 %

Reliability : (1) valid without restriction
GLP guideline study

05.09.2003 (18)

Species : rabbit
Concentration :
Dose :
Exposure time :
Comment : no data
Number of animals : 5
Vehicle : no data
Result : highly irritating
Classification :
Method : other
Year : 1953
GLP : no
Test substance : other TS

Result : DESCRIPTION OF LESIONS: The instillation of 0.005 ml amounts undiluted and 0.5 ml quantities of a 15 % dilution in propylene glycol caused severe corneal necrosis of the eyes of groups of 5 rabbits. A 5 % dilution in propylene glycol caused no injuries. This response places epsilon-caprolactone in Grade 8 of the 10-grade rating system.

Test condition : TEST ANIMALS
- No data

ADMINISTRATION/EXPOSURE
- Preparation of test substance: Undiluted test substance was used and a 15 % dilution in propylene glycol
- Amount of substance instilled: 0.005 ml (undiluted) and 0.5 ml of 5-15 % dilutions in propylene glycol

EXAMINATIONS
- Scoring system: 10-grade rating system for eye burns

Test substance : TEST SUBSTANCE
Source: S. Charleston under identification 242 RD 89
Purity: No data

Reliability : (3) invalid
Significant methodological deficiencies.

10.06.2005 (40)

5.3 SENSITIZATION

5.4 REPEATED DOSE TOXICITY

Type	:	Sub-chronic
Species	:	rat
Sex	:	male/female
Strain	:	Sprague-Dawley
Route of admin.	:	inhalation: vapour
Exposure period	:	9 days
Frequency of treatm.	:	6 hours per day
Post exposure period	:	no
Doses	:	45 ppm (target concentration)
Control group	:	yes, concurrent no treatment
NOAEL	:	= 45 ppm
Method	:	other
Year	:	1991
GLP	:	yes
Test substance	:	other TS

Method : STATISTICAL METHODS: Results of quantitative variables were intercompared between the exposure group and one control group by Levene's test for equal variances and t-tests. Frequency comparisons for microscopic diagnoses were made with Fisher's exact test.
ANALYTICAL METHODS: The concentrations of epsilon-caprolactone vapour were analyzed six times during the six hour exposure period by sampling the chamber atmosphere using sorbent tubes. The samples were desorbed and analyzed by GC/FID.

Result : TOXIC RESPONSE/EFFECTS BY DOSE LEVEL:
- Mortality and time to death: There were no mortalities during the study.
- Clinical signs: One male in the 45 ppm group was noted to have swollen periocular tissue, although this did not appear to be exposure related. Perinasal encrustation was observed in males and females in both the 0 and 45 ppm groups.
- Body weight gain: There were no differences in mean body weight values and in the mean body weight gains for the males and females in the 45 ppm group.
- Ophtalmoscopic examination: No lesions were observed in the animals.
- Clinical chemistry/haematology: No hematologic differences were observed for the males and females in the 45 ppm group.
- Organ weights: There were no differences in the mean absolute and relative (as percentages of body and brain weight) organ weights for the males and females in the 45 ppm group.
- Gross pathology and histopathology: There were no gross or microscopic lesions related to caprolactone exposure.

RESULTS OF CHEMICAL ANALYSIS: The mean epsilon-caprolactone concentration (+/- SD) was 44 (+/- 2.2) ppm. Epsilon-Caprolactone was not detected in the control chamber.

Test condition : TEST ORGANISMS
- Source: Harlan Sprague-Dawley, Inc., Indianapolis, IN

- Age: 56 days at test initiation
- Weight at study initiation: 181.8 g (controls) and 184.2 g (exposed)
- Number of aninmals: 20 per group (total of 40)

ADMINISTRATION/EXPOSURE
- Duration of test/exposure: Animals were exposed 6 hours per day, for 9 exposures during a 2-week period.
- Type of exposure: Vapour
- Post exposure period: No
- Type or preparation of vapour: Liquid caprolactone was metered from a piston pump into a glass evaporator.
- Vehicle: Air with an airflow rate of 300 liters/minute (13-14 air changes per hour).
- Concentrations: Target epsilon-caprolactone concentrations of 0 (control) and 45 ppm.

CLINICAL OBSERVATIONS AND FREQUENCY
- Clinical signs: All animals were individually observed for signs of toxic effects except during exposure. During the exposure, observations were recorded on a group basis.
- Mortality: Animals were observed twice a day for mortality.
- Body weight: All animals were weighed prior to the first exposure. The animals were also weighed prior to the second, fifth, sixth, and seventh exposures and immediately prior to sacrifice.
- Food/water consumption: No observations were made.
- Ophtalmoscopic examination: At test initiation and termination ophthalmic examinations were performed.
- Haematology: Hematological evaluations were performed on blood samples collected from all rats at sacrifice.

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
- Macroscopic: Organs (brain, liver, lungs, kidneys and testes) were weighed at sacrifice. Gross examinations were performed on all animals.
- Microscopic: A complete necropsy was performed on each animal. Microscopic evaluations were performed on the following tissues: adrenals, brain, eyes, heart, kidneys, larynx, liver, nasal turbinates, ovaries, spleen, stomach, testes, thymus, trachea.

Test substance : TEST SUBSTANCE
- Source: Union Carbide Chemicals and Plastics Company Inc.
- Purity: 99.7 %

Reliability : (1) valid without restriction
GLP study, test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

10.06.2005 (28)

Type	:	Sub-chronic
Species	:	rat
Sex	:	male/female
Strain	:	Sprague-Dawley
Route of admin.	:	drinking water
Exposure period	:	14 days
Frequency of treatm.	:	Not applicable
Post exposure period	:	No
Doses	:	0, 500, 2000 and 5000 ppm
Control group	:	yes, concurrent vehicle
NOAEL	:	= 2000 ppm

LOAEL : = 5000 ppm
Method : other
Year : 1991
GLP : yes
Test substance : other TS

Method : STATISTICAL METHOD: Data for continuous, parametric variables were intercompared for the dose and control groups by using Levene's test for homogeneity of variances by analysis of variance and by t-tests.
ANALYTICAL METHOD: Not applicable

Result : TOXIC RESPONSE/EFFECTS BY DOSE LEVEL:
- Mortality and time to death: There were no mortalities during the study.
- Clinical signs: No clinical signs were observed in either sex at any dose level.
- Body weight gain: Mean absolute body weight and/or body weight gain were reduced in the 5000 ppm group of males and females throughout the study. The average reduction was 24 % based on the whole exposure period. Mean body weight gain was also reduced (32%) in the 2000 ppm group of both sexes during the Day 0-4 measurement period but based on the whole exposure period day (0-14) there was no reduction (only 4 %, not statistically significant) in body weight gain.
- Food/water consumption: Dose-related decreases in mean water consumption were observed in all treated groups and were attributed to aversion to the caprolactone drinking water solutions. Decreases in mean food consumption were observed for the 2000 and 5000 ppm groups of both sexes during the Day 0-4 measurement period.
- Clinical chemistry/haematology: No treatment-related effects on hematology and clinical chemistry were observed in males or females of any treated group. The only effect in clinical chemistry that could possibly be related to treatment was an increased urea nitrogen observed in the males of the 5000 ppm group. As there was no dose-effect-relationship and there were no histological lesions observed in the kidneys, this effect was not considered of toxicological relevance.
- Organ weights: No treatment-related effects on organ weights were observed in males or females of any treated group.
- Gross pathology and histopathology: There were no gross or microscopic lesions attributed to the administration of caprolactone in the drinking water.
- Based on the report of the study, the no-observed-effect level was 2000 ppm based on effects on food and water consumption, and body weight. However, the effect on on body weight gain was only between day 0-4 and therefore 2000 ppm is considered a NOAEL.

TEST SUBSTANCE CONSUMPTION: The mean caprolactone intakes over the study period were 45, 152 and 347 mg/kg/day for males and 53, 184 and 384 mg/kg/day for females for the 500, 2000 and 5000 ppm groups, respectively.

Test condition : TEST ORGANISMS
- Source: Harlan Spargue-Dawley Inc. Indianapolis IN.
- Age: not described
- Weight at study initiation: 242 g
- Number of animals: 20 per group (total of 80)

ADMINISTRATION/EXPOSURE

- Type of exposure: Via drinking water
- Vehicle: Milli-Q water
- Concentration in vehicle: 0, 500, 2000 and 5000 ppm
- Total volume applied: The test substance consumption was calculated based on the nominal concentrations of epsilon-caprolactone in drinking water and water consumption.

CLINICAL OBSERVATIONS AND FREQUENCY
- Clinical signs: Clinical observations were performed on days 0, 2, 4, 7 and 9-14.
- Mortality: Animals were observed twice a day for mortality.
- Body weight: Animals were weighed on Days 0, 4, 7, 10 and 14.
- Food/water consumption: Water and food consumption data were collected for all animals on Days 0, 4, 7, 10 and 14.
- Haematology: After 14 days of treatment, blood was obtained for hematology and clinical chemistry determinations.

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
- Macroscopic: The liver, kidneys, lungs and testes were weighed.
- Microscopic: A complete necropsy was performed on each animal. The liver, kidneys, lungs and testes were examined microscopically for the control and high dose groups.

CHEMICAL ANALYSIS: No analyses of test solutions were performed. Concentration of the test substance in the dosing solutions was documented on a weight basis.

Test substance : TEST SUBSTANCE
- Source: Union Carbide Chemicals and Plastics Company Inc.
- Purity: 99.7 %

Reliability : (1) valid without restriction
GLP study, test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

10.06.2005 (14)

Type	:	Chronic
Species	:	rat
Sex	:	male/female
Strain	:	Sprague-Dawley
Route of admin.	:	inhalation: vapour
Exposure period	:	90 days
Frequency of treatm.	:	6 hours per day, 5 days per week
Post exposure period	:	Ten additional males and females were in the 0 and 45 ppm groups for a 4-week recovery period.
Doses	:	target concentrations of 0, 15 and 45 ppm
Control group	:	yes, concurrent vehicle
NOAEL	:	= 15 ppm
LOAEL	:	= 45 ppm
Method	:	other
Year	:	1992
GLP	:	yes
Test substance	:	other TS

Method : STATISTICAL METHODS: The data for continuous, parametric variables were intercompared for the exposure and control groups by use of Levene's test for homogeneity of variance, by analysis of variance and by t-

tests. The probability value of p<0.05 (two-tailed) was used as the critical level of significance for all tests.
ANALYTICAL METHODS: The concentrations of epsilon-caprolactone vapour were analyzed six times during the six hour exposure period by sampling the chamber atmosphere using sorbent tubes. The samples were desorbed and analyzed by GC/FID.

Result : TOXIC RESPONSE/EFFECTS BY DOSE LEVEL:
- Mortality and time to death: There were no mortalities during the study.
- Clinical signs: Swollen periocular tissues were observed in the control and 45 ppm groups, but no exposure relationship was evident.
- Body weight gain: The mean values for body weight and body weight gain values were lower, but not statistically significantly lower, than control values for males and females at the 14-week sacrifice and for the males at the 18-week sacrifice.
- Food/water consumption: On week 4 in males exposed to 45 ppm, a significantly decreased food consumption value was noted.
- Ophtalmoscopic examination: No lesions were observed in the animals.
- Clinical chemistry/haematology: Changes in the hematologic values were considered to be sporadic and not related to caprolactone exposure.
- Urinalysis: No significant differences in urinalysis in male or female rats of the 15 and 45 ppm groups were noted.
- Organ weights: No caprolactone-exposure related differences in organ weights were noted.
- Gross pathology and histopathology: The only caprolactone exposure-related lesions at the 14-week sacrifice were perinasal and periocular encrustation and eyelid swelling in the males of the 45 ppm group.

RESULTS OF CHEMICAL ANALYSIS: Gas chromatographic analysis of the chamber atmosphere resulted in mean (+/-SD) concentrations of 14.2 (+/-1.13) and 42.4 (+/-4.02) ppm. Epsilon-caprolactone was not detected in the control chamber.

Test condition : TEST ORGANISMS
- Source: Harlan Sprague-Dawley, Inc., Indianapolis, IN
- Age: 49 days at test iniation
- Mean weight at study initiation: 231.8 g
- Number of animals: 20 per group (total of 100)

ADMINISTRATION/EXPOSURE
- Duration of test/exposure: Animals were exposed for 6 hours per day, 5 days a week, for 13 weeks. The animals were exposed for 3 days during the fourteenth week.
- Type of exposure: Vapour
- Post exposure period: Ten additional males and females were in the 0 and 45 ppm groups for a 4-week recovery period.
- Type or preparation of vapour: Liquid caprolactone was metered from a piston pump into a glass evaporator.
- Vehicle: air with an airflow rate of 200 liters/minute (13-14 air changes per hour).
- Concentrations: Target epsilon-caprolactone concentrations of 0 (control), 15 and 45 ppm.

CLINICAL OBSERVATIONS AND FREQUENCY
- Clinical signs: During nonexposure days, the animals were examined once a day for overt clinical signs. All animals

were individually observed for signs of toxic effects except during exposure. During the exposures, observations were recorded on a group basis.
- Mortality: Animals were observed twice a day for mortality.
- Body weight: All animals were weighed prior to the first exposure (week 0). The animals were weighed weekly throughout the exposure regime and immediately prior to sacrifice.
- Food/water consumption: Food and water consumption measurements were obtained on a weekly basis during the first 4 weeks of exposure.
- Ophtalmoscopic examination: At test initiation and termination, ophthalmic examination was performed.
- Haematology: Hematology and serum clinical chemistry evaluations were performed on blood samples collected from all rats at the end of the exposure regimen and on 10 rats per sex in the control and high exposure groups at the end of the 4-week recovery period.
- Urinalysis: Urinalysis was performed following Day 64 for the male rats and Day 65 for the female rats.

ORGANS EXAMINED AT NECROPSY (MACROSCOPIC AND MICROSCOPIC):
- Macroscopic: Organs were weighed at sacrifice. Gross examinations were performed on all animals.
- Microscopic: Microscopic evaluations were performed on the following tissues from animals of the control and high exposure groups: Adrenals, brain, eyes, kidneys, nasal turbinates, bone and bone marrow, esophagus, stomach, intestine, spleen, lungs, thymus, urinary bladder, spinal cord, larynx, epididymides, prostate, seminal vesicles, testes, thyroids, uterus, ovaries, vagina, cervix, pituitary, muscle-gastrocnemius, liver, nerve-sciatic, trachea, mammary tissue, salivary glands, skin (flank), aorta, parathyroids, heart, pancreas, lymph nodes.

Test substance : TEST SUBSTANCE
- Source: Union Carbide Chemicals and Plastics Company Inc.
- Purity: > 99 %

Reliability : (1) valid without restriction
GLP study, test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

15.09.2003 (29)

5.5 GENETIC TOXICITY 'IN VITRO'

Type	:	Mammalian cell gene mutation assay
System of testing	:	CHO/HGPRT
Test concentration	:	1000, 2000, 3000, 4000 and 5000 micrograms/ml (non-activated system); 250, 500, 1000, 2000, 3000, 4000 and 5000 micrograms/ml (S9-activated system)
Cycotoxic concentr.	:	
Metabolic activation	:	with and without
Result	:	negative
Method	:	other
Year	:	1997
GLP	:	yes
Test substance	:	other TS

Result : GENOTOXIC EFFECTS:
- With and without metabolic activation: No positive responses were observed.

CYTOTOXIC CONCENTRATION:
- With metabolic activation: No toxicity was observed
- Without metabolic activation: Toxicity was observed at doses of 2100 micrograms/ml and higher.

Test condition : SYSTEM OF TESTING
- Species/cell type: CHO-K1-BH4 cells
- Source: Oak Ridge National Laboratories, Oak Ridge, TN
- Metabolic activation system: Aroclor 1254-induced rat liver S9-activated system.

ADMINISTRATION
- Numer of replicates: 2
- Application: 50 microliters test solution in distilled water was added to the cells.
- Positive and negative control groups and treatment: Ethyl methanesulfonate (0.2 ul/mL) was used as the positive control for the non-activated test system. Benzo(a)pyrene (4 ug/ml) was used as the positive control for the S9-activated test system. The solvent (distilled water) was used as the negative control.
- Pre-incubation time: Cells were incubated at 37 +/- 1 degrees C for 18-24 hours.

DESCRIPTION OF FOLLOW UP REPEAT STUDY: An independent repeat mutation assay, with and without a metabolic activation system, was conducted. In the non-activated system, cultures were treated with concentrations of 1000, 2000, 3000, 4000 and 5000 ug/ml. The S9-activated system was treated with concentrations of 500, 1000, 1500, 1600, 1700, 1800, 1900, 2000, 2100 and 2200 ug/ml.

CRITERIA FOR EVALUATING RESULTS:
Negative Controls: The cloning efficiency of the solvent control must be >50%. The spontaneous mutant frequency must fall within the range of 0-25 mutants per 10.000.000 clonable cells.
Positive Controls: The positive control must induce a mutant frequency at least 3 times that of the solvent control and must exceed 40 mutants per 10.000.000 clonable cells.
Test Substance-treated Cultures: A minimum of 4 analyzable concentrations with mutant frequency data will be required.

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: 100 %

Reliability : (1) valid without restriction
GLP guideline study, test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

10.06.2005 (35)

Type : Mammalian cell gene mutation assay
System of testing : Chinese Hamster Ovary (CHO) cells
Test concentration : 1.0, 0.50, 0.25, 0.125 and 0.0625% v/v (without S9 activation); 0.1, 0.05, 0.025, 0.0125 and 0.00625% v/v (with S9 activation)
Cycotoxic concentr. :
Metabolic activation : with and without
Result : ambiguous
Method : other
Year : 1981
GLP : no
Test substance : other TS

Result : CYTOTOXIC CONCENTRATION:
- With and without metabolic activation: Only a moderate degree of cell killing was obtained with the top concentration of epsilon-caprolactone, in tests with or without S9 activation, in comparison to the cytotoxicity obtained with the solvent control.

FREQUENCY OF EFFECTS:

Test conc. (%, v/v)	Total Mutant Colonies	Mutants/10E+6 Viable cells
Without S9 activation		
1.0	8	18.7*
0.5	6	16.2*
0.25	4	13.4
0.125	0	0
0.0625	7	13.8*
H2O	2	3.3
DMSO (20 ul/ml)	3	5.7
EMS (200 ug/ml)	124	275.6*
With S9 activation		
0.1	6	6.4
0.05	0	0
0.025	0	0
0.0125	1	1.2
0.00625	2	3.0
H2O	5	5.2
DMSO (20 ul/ml)	18	19.9*
DMN (3700 ug/ml)	63	127.3*
DMN (740 ug/ml)	30	45.6*

*: Denotes statistical significance

GENOTOXIC EFFECTS:
- Without metabolic activation: Epsilon-caprolactone produced three statistically significant increases in the frequency of mutants. However, there was no dose-related effect.
- With metabolic activation: No significant effect on mutant frequency was obtained.

Test condition : SYSTEM OF TESTING
- Species/Cell type: CHO-K1-BH4-D1
- Source: Oak Ridge National Laboratory
- Metabolic activation system: S9 homogenate prepared from Aroclor-1254 induced Sprague-Dawley male rats.
- Test duration: 16 hours (without S9 activation), 5 hours (with S9 activation).

ADMINISTRATION
- Dosing: 1.0, 0.50, 0.25, 0.125 and 0.0625% v/v (without S9 activation); 0.1, 0.05, 0.025, 0.0125 and 0.00625% v/v (with S9 activation).
- Application: A preliminary experiment was performed to select a range of test concentrations in which the maximum concentration would allow survival of about 10 % of the treated cells.
- Positive and negative control groups and treatment: Sterile water was used as the solvent and solvent control. DMSO was used as the negative control.

Dimethylnitrosamine (DMN) or ethylmethanesulfonate (EMS) were used as positive controls with or without S9 metabolic activation.
MEASUREMENTS: The surviving fraction was determined at 20 to 24 hours after treatment and the mutant fraction was determined after a 7-day period to allow "expression" of the mutant phenotype.
CRITERIA FOR EVALUATING RESULTS: In this assay forward mutations are detected from TG-sensitivity to TG-resistance caused by a direct loss of the activity of the HGPRT enzyme. The number of mutants should be statistically higher than the spontaneous mutation frequency of about 4 to 5 mutants/10E6 viable cells.

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: 100%

Reliability : (1) valid without restriction
Test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

10.06.2005 (39)

Type : Sister chromatid exchange assay
System of testing : Chinese Hamster Ovary (CHO) cells
Test concentration : Dosing: 1.0, 0.50, 0.25, 0.125 and 0.0625% v/v (without S9 activation); 0.1, 0.05, 0.025, 0.0125 and 0.00625% v/v (with S9 activation).
Cycotoxic concentr. :
Metabolic activation : with and without
Result : negative
Method : other
Year : 1981
GLP : no
Test substance : other TS

Result : CYTOTOXIC CONCENTRATION:
- With and without metabolic activation: Only a moderate degree of cell killing was obtained with the top concentration of epsilon-caprolactone, in tests with or without S9 activation, in comparison to the cytotoxicity obtained with the solvent control.

FREQUENCY OF EFFECTS:

Test conc. (%, v/v)	SCE/Cell	Mean no. SCE/ chromosome
Without S9 activation		
1.0	11.67	0.608
0.5	10.87	0.561
0.25	8.60	0.480
0.125	7.60	0.392
0.0625	8.87	0.454
H2O	10.20	0.549
DMSO (5 ul/ml)	8.53	0.425
EMS (100 ug/ml)	22.60	1.141
With S9 activation		
0.1	13.27	0.697
0.05	12.40	0.635
0.025	10.93	0.561
0.0125	10.80	0.591
0.00625	13.07	0.674

H2O 15.40 0.785
DMSO (5 ul/ml) 14.27 0.743
DMN (2220 ug/ml)27.40 1.430

GENOTOXIC EFFECTS:
- With and Without metabolic activation: No statistically significant increase in the frequency of SCE was obtained at any concentration tested with or without the presence of a metabolic activation system.

Test condition : SYSTEM OF TESTING
- Species/Cell type: CHO-K1-BH4-D1
- Source: Oak Ridge National Laboratory
- Metabolic activation system: S9 homogenate prepared from Aroclor-1254 induced Sprague-Dawley male rats.
- Test duration: 5 hours (without S9 activation), 2 hours (with S9 activation).

ADMINISTRATION
- Dosing: 1.0, 0.50, 0.25, 0.125 and 0.0625% v/v (without S9 activation); 0.1, 0.05, 0.025, 0.0125 and 0.00625% v/v (with S9 activation).
- Application: A preliminary experiment was performed to select a maximum dose level which would permit survival of at least 50% of the treated cells.
- Positive and negative control groups and treatment: Sterile water was used as the solvent and solvent control. DMSO was used as the negative control. Dimethylnitrosamine (DMN) or ethylmethanesulfonate (EMS) were used as positive controls with or without S9 metabolic activation.

CRITERIA FOR EVALUATING RESULTS: An increase in the frequency of Sister Chromatid Exchange (SCE) can be observed in cells treated with physical or chemical mutagenic agents. The number of SCE/cell in the cells exposed to the test substance should be statistically higher than the SCE/cell of the solvent control.

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: 100 %

Reliability : (1) valid without restriction
Test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

10.06.2005 (39)

Type : Unscheduled DNA synthesis
System of testing : Hepatocyte suspension
Test concentration : 0.1, 0.03, 0.01, 0.003, 0.001 and 0.0001 % (v/v)
Cycotoxic concentr. :
Metabolic activation :
Result : ambiguous
Method : other
Year : 1981
GLP : no
Test substance : other TS

Result : CYTOTOXIC CONCENTRATION:
Only a moderate degree of cell killing was obtained with the top concentration of epsilon-caprolactone.
GENOTOXIC EFFECTS:
In hepatocytes treated with epsilon-caprolactone, 3 concentrations tested for potential activity produced a statistically significant increase in the amount of tritiated-thymidine incorporation. Also, all six concentrations

produced numerical increases in the amount of UDS in comparison to the solvent control. However, there was no distinct dose-related increase in the amount of UDS, characteristic of strong mutagenic agents.

Test condition : SYSTEM OF TESTING
- Species/Cell type: Hepatocyte suspension prepared from Hilltop-Wistar rats
- Test duration: 2 hours

ADMINISTRATION
- Number of replicates: The test was conducted in duplicate
- Dosing: 0.1, 0.03, 0.01, 0.003, 0.001 and 0.0001 % (v/v)
- Positive and negative control groups and treatment: 4-nitroquinoline (NQO) and dimethylnitrosamine (DMN) are run in duplicate as positive controls. The solvent control (DMSO) is run in quadruplicate.
- Measurements: Determination of UDS activity was performed by analysis of incorporation of radioactive thymidine into isolated hepatocyte nuclei or in DNA.

CRITERIA FOR EVALUATING RESULTS: The stimulation of incorporation of tritiated thymidine into both purified hepatocyte nuclei and DNA is used as the indicator of chemically induced DNA damage. The average disintegrations per minute (DPM) is calculated for each dose level and the controls and the final results are expressed as DPM/10E+6 viable hepatocytes. The test is positive if the increase in the amount of UDS activity is statistically significant.

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: 100 %

Reliability : (1) valid without restriction
Test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

10.06.2005 (39)

Type : Mouse lymphoma assay
System of testing : L5178Y cell
Test concentration : not described
Cycotoxic concentr. : not described
Metabolic activation : without
Result : negative
Method : other
Year : 1983
GLP : no
Test substance : no data

Result : GENOTOXIC EFFECTS:
- Without metabolic activation: The mutation frequency was not significantly higher than the background.

Test condition : SYSTEM OF TESTING
- Species/cell type: L5178Y cell line

ADMINISTRATION
- Positive and negative control groups and treatment: Negative control is the same as solvent control

CRITERIA FOR EVALUATING RESULTS: The test system is based on the quantitation of forward mutations occurring at the heterozygous thymidine kinase (TK) locus. An agent is active if the mutation frequency is twice the background at a survival >10 %.

Reliability : (4) not assignable
Documentation insufficient for assessment

10.06.2005 (4)

Type : Ames test

System of testing	:	Salmonella typhimurium
Test concentration	:	10, 100 and 500 or 1000 micrograms
Cycotoxic concentr.	:	
Metabolic activation	:	with
Result	:	negative
Method	:	other
Year	:	1975
GLP	:	no
Test substance	:	other TS
Remark	:	In the publication of McCann et al. (1975) the most complete description of the study is given. In the publication of Kier et al. (1986) only a short overview is given.
Result	:	GENOTOXIC EFFECTS: - With metabolic activation: The number of revertants/nmol was <0.0008 (<70 revertant colonies per 10 micrograms tested).
Test condition	:	SYSTEM OF TESTING - Species/cell type: Salmonella strains TA 98, 100, 1535, 1537 - Metabolic activation system: Aroclor 1254-induced rat liver S9-activated system. CRITERIA FOR EVALUATING RESULTS: - The test is negative if the number of revertants/nmol is < 0.01.
Test substance	:	TEST SUBSTANCE - Supplier: Aldrich - Purity: not described
Reliability	:	(4) not assignable Documentation insufficient for assessment

10.06.2005 (21) (26)

Type	:	other: Chromosome Aberrations and Sister Chromatid Exchanges
System of testing	:	Chinese Hamster Cells
Test concentration	:	10E-5, 10E-4, 10E-3 M
Cycotoxic concentr.	:	
Metabolic activation	:	no data
Result	:	negative
Method	:	other
Year	:	1977
GLP	:	no
Test substance	:	no data
Remark	:	In the publication of Abe and Sasaki (1977) the most complete description of the study is given. In the publication of Latt et al. (1981) only a short overview is given.
Test condition	:	SYSTEM OF TESTING - Species/cell type: Pseudo-diploid Chinese hamster cell line - No. of metaphases analyzed: Chromosome aberrations were examined on 100 metaphase plates for each dose and the frequency of aberrations was indicated by the number of breaks per cell. The number of SCE per cell was determined on the basis of 20-50 intact metaphases. CRITERIA FOR EVALUATING RESULTS: The number of chromosome aberrations is defined as positive when the number of breaks or SCE per cell was more than twice the control value.
Reliability	:	(4) not assignable Documentation insufficient for assessment

10.06.2005 (1) (22)

Type : other: cell transformation
System of testing : Golden syrian hamster embryo cells
Test concentration : 0, 0.1, 1.0, 10 and 100 micrograms/ml
Cycotoxic concentr. :
Metabolic activation : no data
Result : negative
Method : other
Year : 1977
GLP : no
Test substance : no data

Remark : In the publication of Pienta et al. (1977) the most complete description of the study is given. In the publication of Heidelberger et al. (1983) only a short overview is given.

Result : GENOTOXIC EFFECTS:

Conc. (ug/ml)	Transformed colonies/surviving colonies*	
0	0/479	0/268
0.1	0/483	0/485
1.0	0/478	0/475
10	0/474	0/447
100	0/194	0/205

*The test was performed with two different cell cultures

Test condition : SYSTEM OF TESTING
- Species/cell type: Golden syrian hamster embryo cells
- Source: Lakeview Hamster Colony, Newfield, NJ
- Metabolic activation system: Aroclor 1254-induced rat liver S9-activated system.

ADMINISTRATION
- Numer of replicates: 6, the test was conducted in duplicate
- Application: Test chemicals were dissolved in Dulbecco's modified Eagle medium immediately prior to use.
- Positive and negative control groups and treatment: Benzo(a)pyrene and 3-methylcholanthrene were used as positive controls.
- Pre-incubation time: 4 Days befor use, cryopreserved cells were seeded.

CRITERIA FOR EVALUATING RESULTS:
The endpoint is the presence of fibroblast-like colonies morphologically altered beyond that observed in normal cells.

Reliability : (4) not assignable
Documentation insufficient for assessment

10.06.2005 (13) (32)

Type : other: intraperitoneal host-mediated assay
System of testing : mice implanted with Salmonella Typhimurium and Saccharomyces cerevisiae
Test concentration : 1st group: 432 mg/kg; 2d group: 1300 mg/kg
Cycotoxic concentr. :
Metabolic activation : without
Result : negative
Method : other
Year : 1979
GLP : no data
Test substance : no data

Remark	:	In the publication of Simmon et al. (1979) the most complete description of the study is given. In the publication of Legator et al. (1982) only a short overview is given.
Test condition	:	SYSTEM OF TESTING - Species/cell type: Salmonella typhimurium strains TA 1530, TA 1535, TA 1538; Saccharomyces cerevisiae D3; Swiss-Webster mice ADMINISTRATION - Application: In 1st group, epsilon-caprolactone was administered to mice by im injection at 432 mg/kg; In 2d group, caprolactone was administered to mouse by oral intubation at a single dose of 1300 mg/kg in 0.2 ml dimethyl sulfoxide. 2 ml of an overnight culture of the microorganisms was injected i.p. into the mouse; after 4 hours, the mice were killed; peritoneal fluids were recovered to perform the plate-tests. CRITERIA FOR EVALUATING RESULTS: The mutant organisms per ml and the total organisms per ml were measured for each mouse. With Salmonella a compound was judged mutagenic if a two-tailed t-test of statistical comparison showed P at 0.05 or less. With S. cerevisiae, compounds were regarded as mutagenic if the mutation frequency of the treated animals was > 10 times that of the controls.
Reliability	:	(4) not assignable Documentation insufficient for assessment

10.06.2005 (23) (38)

Type	:	Mitotic recombination in Saccharomyces cerevisiae
System of testing	:	Saccharomyces cerevisiae D3
Test concentration	:	Concentration of 5 % was the highest dose tested
Cycotoxic concentr.	:	
Metabolic activation	:	without
Result	:	negative
Method	:	other
Year	:	1979
GLP	:	no data
Test substance	:	no data
Remark	:	In the publication of Simmon (1979) the most complete description of the study is given. In the publication of Zimmerman et al. (1984) only a short overview is given.
Result	:	GENOTOXIC EFFECTS: At the highest concentration tested (5 %) the number of mitotic recombinants was 9 per 100.000 survivors.
Test condition	:	SYSTEM OF TESTING - Species/cell type: Saccharomyces cerevisiae D3 ADMINISTRATION - Application: Dimethyl sulfoxide was used as solvent - Positive and negative control groups and treatment: Appropriate positive and negative controls were included. CRITERIA FOR EVALUATING RESULTS: A positive response in this assay is indicated by a dose-related increase of more than threefold in the number of mitotic recombinants per 100.000 survivors.
Reliability	:	(4) not assignable Documentation insufficient for assessment

10.06.2005 (36) (50)

Type	:	Chromosomal aberration test
System of testing	:	Chinese hamster cells
Test concentration	:	Maximum effective dose was 0.5 mg/ml

Cycotoxic concentr.	:	
Metabolic activation	:	without
Result	:	negative
Method	:	other
Year	:	1977
GLP	:	no
Test substance	:	other TS
Result	:	GENOTOXIC EFFECTS: The test was negative because 4.0 % of the cells was found to have chromosomal aberrations.
Test condition	:	SYSTEM OF TESTING - Species/cell type: Chinese hamster fibroblast cell line (CHL) - Source: Cancer Institute, Tokyo - No. of metaphases analyzed: The number of cells with chromosomal aberrations was recorded on 100 well-spread metaphases. - Test duration: 48 hours ADMINISTRATION - Application: A growth inhibition test was carried out before the chromosome tests were started. For the chromosome test three different doses, including the 50% inhibition dose, were prepared. Ethanol was used as solvent. CRITERIA FOR EVALUATING RESULTS: CHL cells commonly have < 3% cells with chromosomal aberrations. The test was negative (-) if less than 4.9% of the aberration was detected, suspicious (+/-) if between 5.0 and 9.9%, and positive if between 10 and 19.9% (+), 20.0 and 49.9% (++) or more tan 50.0% (+++).
Reliability	:	(4) not assignable Documentation insufficient for assessment

10.06.2005 (16)

Type	:	Ames test
System of testing	:	Salmonella Typhimurium
Test concentration	:	0, 1 and 10 microliters compound per plate
Cycotoxic concentr.	:	
Metabolic activation	:	with and without
Result	:	negative
Method	:	other
Year	:	1979
GLP	:	no
Test substance	:	other TS
Result	:	GENOTOXIC EFFECTS: The test was negative because the number of mutants per plate was not increased at 1 or 10 microliters compound per plate.
Test condition	:	SYSTEM OF TESTING - Species/Cell type: Salmonella typhimurium strains TA1535 and TA1538 - Metabolic activation system: Livers of uninduced Sprague-Dawley rats were used (S9-fraction). - Test Duration: 48-54 hours ADMINISTRATION - Numer of replicates: 2, on at least three occasions - Application: 0, 1 and 10 microliters compound per plate - Positive and negative control groups and treatment: Plates containing known base-substitution mutagens and known frame-shift mutagens were included as positive

group in the test without metabolic activation. Plates containing 2-fluorenamine were included as positive group in the test with metabolic activation. Plates incubated with buffer were used as negative controls.
CRITERIA FOR EVALUATING RESULTS
- The number of revertants to histidine independence was determined.

Reliability : (4) not assignable
Documentation insufficient for assessment

06.01.2004 (34)

Type : DNA damage and repair assay
System of testing : Normal and DNA polymerase-deficient Escherichia coli
Test concentration : 10 microliters
Cycotoxic concentr. :
Metabolic activation : with and without
Result : negative
Method : other
Year : 1979
GLP : no
Test substance : other TS

Remark : In the publication of Rosenkranz and Poirier (1979) the most complete description of the study is given. In the publication of Leifer et al. (1981) only a short overview is given.
Result : GENOTOXIC EFFECTS:
The test was negative because there was no difference in the zone of inhibition between pol A- and pol A+.
Test condition : SYSTEM OF TESTING
- Species/Cell type: normal (pol A+) and DNA polymerase I-deficient (pol A-) Escherichia coli indicator strains.
- Metabolic activation system: Livers of uninduced Sprague-Dawley rats were used (S9-fraction).
- Test Duration: 7-12 hours
ADMINISTRATION
- Numer of replicates: 2, on at least three occasions
- Application: 10 microliters compound per plate
- Positive and negative control groups and treatment: Methyl methanesulfonate (10 microliters) and chloramphenicol (30 micrograms) were used as positive and negative controls respectively.
CRITERIA FOR EVALUATING RESULTS: The diameters of the zones of growth inhibition were determined.
Reliability : (4) not assignable
Documentation insufficient for assessment

10.06.2005 (24) (34)

Type : Ames test
System of testing : Salmonella typhimurium
Test concentration : up to and including 250 micrograms
Cycotoxic concentr. :
Metabolic activation : with and without
Result : negative
Method : other
Year : 1979
GLP : no
Test substance : other TS

Result : GENOTOXIC EFFECTS:
The test was negative because at a dose of 250 micrograms no revertants

were observed in any of the tested strains.

Test condition : SYSTEM OF TESTING
- Species/cell type: Salmonella typhimurium strains TA1535, TA1536, TA1537, TA1538, TA98 and TA100
- Source: University of California, Berkeley, California
- Metabolic activation system: Aroclor 1254-induced rat liver S9-activated system.

ADMINISTRATION
- Application: up to and including 250 micrograms
- Positive and negative control groups and treatment: Solvent controls as well as known direct-acting mutagens and a mutagen that required metabolic activation were included.

CRITERIA FOR EVALUATING RESULTS: A positive response was defined as a reproducible, dose-related increase in the number of revertants.

Reliability : (4) not assignable
Documentation insufficient for assessment

06.01.2004 (37)

5.6 GENETIC TOXICITY 'IN VIVO'

Type	:	Micronucleus assay
Species	:	mouse
Sex	:	male/female
Strain	:	ICR
Route of admin.	:	i.p.
Exposure period	:	24-72 hours
Doses	:	Vehicle control, 250, 500, 1000 mg/kg
Result	:	negative
Method	:	other
Year	:	1997
GLP	:	yes
Test substance	:	other TS

Result : MORTALITY: No mortality occurred at any dose level during the course of the micronucleus study.
CLINICAL SIGNS: Lethargy in male and female mice at all dose levels and prostration in male and female mice at 1000 mg/kg.
GENOTOXIC EFFECTS: Reductions of 2 to 6 % in the ratio of polychromatic erythrocytes to total erythrocytes were observed in some of the test article-treated groups relative to their respective vehicle controls. The number of micronucleated polychromatic erythrocytes per 1000 polychromatic erythrocytes was not statistically increased in either male or female mice, regardless of dose level or bone marrow collection time. The positive control (CP) induced a significant increase in micronucleated polychromatic erythrocytes in both male and female mice.

Test condition : TEST ORGANISMS
- Source: Harlan Sprague Dawley, Inc., Frederick, MD.
- Age: 6 to 8 weeks at test initiation
- Weight at study initiation: 33.7-39.6 g (males), 24.1-28.6 g (females)
- No. of animals per dose: 30 (15 per sex)

ADMINISTRATION
- Vehicle: sterile distilled water
- Duration of test: 24-72 hours
- Frequency of treatment: single dose
- Volume applied: 20 ml test article-vehicle mixture/kg

body weight
- Sampling times and number of samples: Bone marrow cells of five animals per sex were collected 24, 48, 72 hours after treatment
- Control groups and treatment: Cyclophosphamide (60 mg/kg) was used as the positive control.

EXAMINATIONS
- Clinical observations: After dose administration mice were observed for clinical signs.
- Criteria for evaluating results: The mean incidence of micronucleated polychromatic erythrocytes must not exceed 5/1000 polychromatic erythrocytes (0.5 %) in the vehicle control. The test article was considered to induce a positive response if a dose-responsive increase in micronucleated polychromatic erythrocytes was observed and one or more doses were statistically elevated relative to the vehicle control (P <0.05, Kastenbaum-Bowman Tables).

Test substance : TEST SUBSTANCE
- Supplier: Union Carbide Corporation
- Purity: 100 %

Reliability : (1) valid without restriction
GLP guideline study, test procedure in accordance with generally accepted scientific standards and described in sufficient detail.

02.09.2003 (33)

5.7 CARCINOGENICITY

Species : mouse
Sex : no data
Strain : no data
Route of admin. : other: skin painting
Exposure period : 24 months
Frequency of treatm. : daily
Post exposure period : No information
Doses : undiluted test substance
Result :
Control group : no data specified
Method : other
Year : 1961
GLP : no
Test substance : other TS

Remark : Epsilon-caprolactone was studied amongst 9 other chemicals.
Result : No pappilomas or carcinomas were observed in any of the mice. 38, 32 or 5 out of 40 mice were still alive after respectively 12 months, 17 or 24 months treatment.
Test substance : TEST SUBSTANCE
- Supplier: commercial
- Purity: not indicated

Reliability : (4) not assignable
Documentation insufficient for assessment

06.01.2004 (27)

5.8.1 TOXICITY TO FERTILITY

Remark : See section 5.8.3 of IUCLID.
10.02.2005

5.8.2 DEVELOPMENTAL TOXICITY/TERATOGENICITY

Remark : See section 5.8.3 of IUCLID
10.02.2005

5.8.3 TOXICITY TO REPRODUCTION, OTHER STUDIES

Remark : No studies are available with regard to reproduction and developmental toxicity of e-caprolactone. However, a well conducted 90-day inhalation study showed no macroscopic and histopathological effects on reproductive organs and also other systemic effects were not found during this study. The lack of systemic effects can be explained by the rapid hydrolysis in stomach and blood, resulting in the formation of 6-hydroxyhexanoic acid (see section 5.0 of IUCLID).

Specific toxicological studies could not be located for 6-hydroxyhexanoic acid or other hydroxyhexanoic acids. However, the toxicological properties of 6-hydroxyhexanoic acid can be predicted based on the chemical structure. Information is available for analogues of 6-hydroxyhexanoic acid (see Annex of the SIAR). In conclusion:
- 1-Hexanol was not teratogenic to rats,
- For 1,6-hexanediol there is no indication of toxic effects on reproductive function or developmental toxicity,
- Adipic acid was not teratogenic and there is no reason to expect specific reproductive toxicity and
- Aliphatic carboxylic acids show no significant evidence of either reproductive or developmental toxicity.

gamma-Butyrolactone (CAS No. 96-48-0) has a similar structure as e-caprolactone but has only 4 instead of 6 carbon atoms. This chemical was evaluated in 14-day, 13-week and 2-year toxicology and carcinogenesis studies and no organ-specific toxicity was observed (NTP, 1996). Furthermore gamma-butyrolactone was rapidly hydrolysed by an enzyme found in the blood and liver and the half-life of the conversion was less than 1 minute.

d-Valerolactone (CAS No. 542-28-9) also has a similar structure as e-caprolactone but has 5 instead of 6 carbon atoms. Based on a "Commission Decision of 23 January 2002 amending Commission Decision 1999/217/EC as regards the register of flavouring substances used in or on foodstuffs" this substance is allowed in the European Union as a flavouring substance.

Conclusion

No studies are available with regard to reproduction and developmental toxicity of e-caprolactone. However, a well conducted 90-day inhalation repeated dose study showed no macroscopic and histopathological changes on reproductive organs and also other systemic effects were not found during this study. The lack of systemic effects can be explained by

the rapid hydrolysis in stomach and blood, resulting in the formation of 6-hydroxyhexanoic acid. Analogues of 6-hydroxyhexanoic acid show no evidence of reproductive or developmental toxicity. For this reason there is no indication for a reprotoxic concern. This is supported by the toxicological profile of structurally similar lactones, where also no organ specific toxicity was observed in long term studies (with up to 2-year exposure).

13.06.2005 (30)

5.9 SPECIFIC INVESTIGATIONS

5.10 EXPOSURE EXPERIENCE

5.11 ADDITIONAL REMARKS

6.1 METHODS HANDLING AND STORING

Safe handling	:	- Carry out industrial operations in closed, but vented, piping circuits and equipment. - Preferably transfer by pump or gravity. - Use only equipment and materials which are compatible with the product.
Fire/exp. protection	:	
Storage requirement	:	- In a ventilated, cool area. - Keep in original packaging, closed. - Containment bund around storage containers and transfer installation. - For bulk storage, consult the producer.
Common storage	:	
Container	:	- Lacquered steel drums
Unsuitable container	:	
Add. information	:	
Transport code	:	

29.06.2004 (42)

6.2 FIRE GUIDANCE

Hazards	:	- Combustible - Gas/vapours mix with air, producing flammable mixtures. - Gas/vapours are heavier than air and so may travel along the ground; remote ignition possible.
Protective equipment	:	
Extinguishing medium	:	- Powder - Foam, AFFF alcohol resistant - CO2 - Water, water spray
Unsuit. exting. medium	:	
Add. information	:	
Fire class	:	
Products arising	:	

29.06.2004 (42)

6.3 EMERGENCY MEASURES

6.4 POSSIB. OF RENDERING SUBST. HARMLESS

6.5 WASTE MANAGEMENT

Memo	:	other
Remark	:	- Dispose in compliance with local/federal and national regulations. - Large quantities: Send the product to an authorized industrial waste incinerator. - Small quantities: The product can be discharged into a biological treatment plant.

29.06.2004 (42)

29.06.2004

6.6 SIDE-EFFECTS DETECTION

6.7 SUBSTANCE REGISTERED AS DANGEROUS FOR GROUND WATER

6.8 REACTIVITY TOWARDS CONTAINER MATERIAL

Memo : Plastic materials may deteriorate on contact with the product.

29.06.2004 (42)

(1) Abe, S. and Sasaki, M. (1977), "Chromosome Aberrations and Sister Chromatid Exchanges in Chinese Hamster Cells Exposed to Various Chemicals", J. Nat. Cancer, Vol. 58, pp. 1635-1640

(2) Anderson, E.M. (1991), "Stability of Dicaprolactone and Caprolactone in Aqueous systems", Bushy Run Research Centre, report 54-104

(3) Billecke, S., Draganov, D., Counsell, R., Stetson, P., Watson, C., Hsu, C. and La Du, B.N. (2000), Human serum paraoxonase (PON1) isozymes Q and R hydrolyze lactones and cyclic carbonate esters. Drug Metabolism and Disposition, 28, (11), 1335-1342.

(4) Clive, D., McCuen, R., Spector, J.F.S, Piper, C. and Mavournin, K.H. (1983), "Specific gene mutations in L5178Y cells in culture", Mutation Research, 115: pp. 225-51

(5) Daubert, T.E. and Danner, R.P. (1989), "Physical And Thermodynamic Properties Of Pure Chemicals: Data Compilation", Design Institute for Physical Property Data, American Institute of Chemical Engineers. Hemisphere Pub. Corp., New York, NY., 4 Vol.

(6) De Groot, W.A. (2004), "Estimation of environmental partitioning of epsilon-caprolactone", Internal Memorandum AdG/HES/im/04.316 of Solvay Pharmaceuticals

(7) Dolich. T. (2003), Hydrolysis as a function of pH of Epsilon-caprolactone. BASF GKA Analytik, Study No. 03L00213

(8) EPA (2000), "AOPWIN v1.90"

(9) EPA (2000), "BCFWIN v2.14"

(10) EPA (2000), "HENRYWIN v1.90"

(11) EPA (2000), "KOWWIN v1.66"

(12) Groeneveld, A.H.C., Van Dijk, N.R.M. and Van den Berg, G. (1992), "The acute toxicity of epsilon-caprolactone to the guppy Poecilia reticulata", Solvay Duphar Int. Doc. No. 56635/19/92

(13) Heidelberger, C., Freeman, A.E., Pienta, R.J., Sivak, A., Bertram, J.S., Casto, B.C., Dunkel, V.C., Francis, M.W., Kakunaga, T., Little, J.B. and Schechtman, L.M. (1983), "Cell transformation by chemical agents-a review and analysis of the literature: A report of the U.S. EPA Gene-Tox Program", Mutation Research 114, pp. 283-385

(14) Hermansky, S.J., Chun, J.S. and Loughran, K.A. (1991), "Caprolactone: Two-Week Drinking Water Study in Sprague-Dawley Rats", Bushy Run Research Center, Report No. 54-40

(15) Hisgen (2003), "Epsilon-Caprolactone - Determination of the acute effect on the swimming ability of the water flea Daphnia magna Straus", BASF Experimental Toxicology and Ecology Project No. 03/0034/50/1

(16) Ishidate, M. and Odashima, S. (1977), "Chromosome tests with 134 compounds on Chinese hamster cells in vitro - A screening for chemical carcinogens", Mutation Research 48, pp. 337-354

(17) Janssen, P.J.M. (1991), "Primary irritation of epsilon-caprolactone to the skin of male rabbits", Solvay Duphar Int. Doc. No. 56645/69/91

(18) Janssen, P.J.M. (1991), "Primary irritation study of epsilon-caprolactone to the eye of male rabbits", Solvay Duphar Int. Doc. No. 56645/106/91

(19) Janssen, W.A.J. and Van den Berg, G. (1992), "The toxicity of epsilon-caprolactone to the bacteria Pseudomonas putida" Solvay Duphar Int. Doc. No. 56635/47/92

(20) Keetelaar-Jansen, W.A.J. and Thus, J. (1993), "Determination of the biodegradability of epsilon-caprolactone in a modified Sturm test", Solvay Duphar Int. Doc. No. 56835/2/93

(21) Kier, L.E., Brusick, D.J., Auletta, A.E., Von Halle, E.S., Brown, M.M., Simmon, V.F., Dunkel, V., McCann, J., Mortelmans, K., Prival, M., Rao, T.K. and Ray, V. (1986), "The Salmonella typhimurium/mammalian microsomal assay: A report of the U.S. EPA Gene-Tox Program", Mutation Research 168, pp. 69-240

(22) Latt, S.A., Allen, J., Bloom, S.E., Carrano, A., Falke, E., Kram, D., Schneider, E., Schreck, R., Tice, R. Whitfield, B. and Wolff, S. (1981), "Sister-chromatid exchanges: a Report of the EPA Gene-Tox Program", Mutation Research 87, pp. 17-62

(23) Legator, M.S., Bueding, E., Batzinger, R., Connor, T.H., Eisenstadt, E., Farrow, M.G., Ficsor, G., Hsie, A., Seed, J. and Stafford, R.S. (1982), "An evaluation of the host-mediated assay and body fluid analysis: A report of the U.S. EPA Gene-Tox Program", Mutation Research 98, pp. 319-374

(24) Leifer, Z., Kada, T., Mandel, M., Zeiger, E., Stafford, R. and Rosenkranz, H.S. (1981), "An evaluation of tests using DNA repair-deficient bacteria for predicting genotoxicity and carcinogenicity: A report of the U.S. EPA Gene-Tox Program", Mutation Research 87, pp. 211-297

(25) Marhold J. (1986) Avicenum, p.802 (in RTECS dd 8/3/89).

(26) McCann, J., Choi, E., Yamasaki, E. and Ames, B.N. (1975), "Detection of carcinogens as mutagens in the Salmonella/microsome test: Assay of 300 chemicals", Proc. Nat. Acad. Sci. USA, Vol. 72, No. 12, pp. 5135-5139

(27) Mellon Institute of Industrial Research (1961). Studies on Carcinogenesis Completed in 1961. Union Carbide Chemicals Co Report 24-117.

(28) Norris, J.C. and Kintigh, W.J. (1991), "Caprolactone: Nine-Day Vapor Inhalation Study on Rats", Bushy Run Research Center, Report No. 54-2

(29) Norris, J.C. and Kintigh, W.J. (1992), "Caprolactone: Ninety-Day Vapor Inhalation Study on Rats", Bushy Run Research Center, Report No. 54-94

(30) NTP (1996), "NTP Summary report on the metabolism, disposition and toxicity of 1,4-butanediol". Toxicity Report Series Numbers 54. NIH Publication 96-3932. See: http://ehp.niehs.nih.gov/ntp/docs/tox54/tx54cov.pdf

(31) OECD (2004), "Sids Initial Assessment Profile (SIAP) of adipic acid". Available via: http://cs3-hq.oecd.org/scripts/hpv/

(32) Pienta, R.J., Poiley, J.A. and Lebherz, W.B. (1977), "Morphological transformation of early passage golden syrian hamster embryo cells derived from cryopreserved primary cultures as reliable in vitro bioassays for identifying diverse carcinogens", Int. J. Cancer: 19, pp. 642-55

(33) Ramadevi, G. and Ritter, P. (1997), "Micronucleus cytogenetic assay in mice", Microbiological Associates Inc., Laboratory Study Number G96CJ23.122

(34) Rosenkranz, S. and Poirier, A.L. (1979), "Evaluation of the Mutagenicity and DNA-Modifying Activity of Carcinogens and Noncarcinogens in Microbial Systems", J. Natl. Cancer Inst. Vol. 62, No. 4, pp. 873-891

(35) San, H.C. and Clarke, J.J. (1997), "In Vitro Mammalian Cell Gene Mutation Test with an Independent Repeat Assay", Microbiological Associates Inc., Study No. G96CJ23.782001

(36) Simmon, V.F. (1979), "In Vitro Assays for Recombinogenic Activity of Chemical Carcinogens and Related Compounds with Saccharomyces Cerevisiae D3", J. Natl. Cancer Inst., Vol. 62, No. 4, pp. 901-909

(37) Simmon, V.F. (1979), "In Vitro Mutagenicity Assays of Chemical Carcinogens and Related Compounds with Salmonella typhimurium", J. Natl. Cancer Inst., Vol. 62, No. 4, pp. 893-899

(38) Simmon, V.F., Rosenkranz, H.S., Zeiger, E. and Poirier, L.A. (1979), "Mutagenic activity of chemical carcinogens and related compounds in the intraperitoneal host-mediated assay", J. Natl. Cancer Inst., Vol. 62, No. 4, pp. 911-918

(39) Slesinski, R.S., Gaunt, M.W., Guzzie, P.J. and Hengler, W.C. (1981), "In Vitro Mutagenesis Studies: 3-Test Battery", Bushy Run Research Center, Project Report 43-125

(40) Smyth H.F., Carpenter, C.P., Weil, C.S. and Pozzani, U.C. (1953), "Range-finding Tests on Epsilon-caprolactone", University of Pittsburgh, Report No. 16-59

(41) Snoeij, N.J. and Buse-Pot, T.E. (1992), "Acute oral toxicity with epsilon-caprolactone in the rat", Solvay Duphar Int. Doc. No. 56645/92/91

(42) Solvay S.A. (2004), Safety data sheet of epsilon-caprolactone

(43) Tolboom, J. (2004), LC50 determination for fish study. Internal Memorandum of Solvay Pharmaceuticals, Weesp, The Netherlands.

(44) Tremain, S.P. (2004), "E-Caprolactone: Determination of vapour pressure", SafePharm Laboratories Ltd Project Number 1524/004

(45) Waggy, G.T. and Payne, J.R. (1974), "Environmental Impact Analysis Product Biodegradability Testing", Research and Development Department, Chemicals and Plastics Union Carbide Corporation, South Charleston, West Virginia, Report No. 910F44.

(46) Waggy, G.T. and Payne, J.R. (1974), "Environmental Impact Product Analysis Acute Aquatic Toxicity Testing", Research and Development Department, Chemicals and Plastics Union Carbide Corporation, South Charleston, West Virginia, Report No. 910F44

(47) Weast, R.C. (1986), "CRC Handbook of Chemistry and Physics, 66th edition"

(48) Werner, D.I. (2003), "Epsilon-Caprolactone - Determination of the inhibitory effect on the cell multiplication of unicellular green algae", BASF Experimental Toxicology and Ecology Project No. 03/0034/60/1

(49) White, D.F. and Mullee D.M. (2004), "E-Caprolactone: Determination of boiling point and water solubility", SafePharm Laboratories Ltd Project Number 1524/003

(50) Zimmerman, F.K., Von Borstel, R.C., Von Halle, E.S., Parry, J.M., Siebert, D., Zetterberg, G., Barale, R. and Loprieno, N. (1984), "Testing of chemicals for genetic activity with Saccharomyces cerevisiae: a report of the U.S. EPA Gene-Tox Program", Mutation Research 133, pp. 199-244