FOREWORD INTRODUCTION

6-Methylhept-5-en-2-one

CAS N°: 110-93-0

SIDS Initial Assessment Report

For

SIAM 17

Arona, Italy, 11–14 November 2003

1. Chemical Name:	6-Methylhept-5-en-2-one
2. CAS Number:	110-93-0
3. Sponsor Country:	Germany Contact Point: BMU (Bundesministerium für Umwelt, Naturschutz und Reaktorsicherheit) Contact person: Prof. Dr. Ulrich Schlottmann Postfach 12 06 29 D- 53048 Bonn- Bad Godesberg
4. Shared Partnership with:	BASF AG, Germany; Hoffmann-La Roche Ltd., Switzerland; Kuraray, Japan.
5. Roles/Responsibilities of the Partners:	
• Name of industry sponsor /consortium	BASF AG, Germany Contact person: Dr. Hubert Lendle, GUP/CL - Z570 D-67056 Ludwigshafen
• Process used	see below
6. Sponsorship History	
• How was the chemical or category brought into the OECD HPV Chemicals Programme ?	by ICCA-Initiative
7. Review Process Prior to the SIAM:	last literature search (update): 8 December 2002 (Human Health): databases medline, toxline; search profile CAS-No. and special search terms 11 April 2003 (Ecotoxicology): databases CA, biosis; search profile CAS-No. and special search terms
8. Quality check process:	As basis for the SIDS-Dossier the IUCLID was used. All data have been checked and validated by BUA.
9. Date of Submission:	12 August 2003
10. Date of last Update:	

11. Comments:

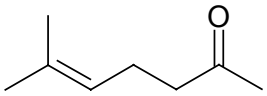
OECD/ICCA - The BUA* Peer Review Process

Qualified BUA personnel (toxicologists, ecotoxicologists) perform a quality control on the full SIDS dossier submitted by industry. This quality control process follows internal BUA guidelines/instructions for the OECD/ICCA peer review process and includes:

- a full (or update) literature search to verify completeness of data provided by industry in the IUCLID/HEDSET
- Review of data and assessment of the quality of data
- Review of data evaluation
- Check of adequacy of selection process for key studies for OECD endpoints, and, where relevant, for non-OECD endpoints by checking original reports/publications
- Review of key study descriptions according to robust summary requirements; completeness and correctness is checked against original reports/publications (if original reports are missing: reliability (4), i.e. reliability not assignable)
- Review o f validity of structure-activity relationships
- Review of full SIDS dossier (including SIAR, SIAP and proposal for conclusion and recommendation for further work)
- In case of data gaps, review of testing plan or rationale for not testing

* BUA (GDCh-Beratergremium für Altstoffe): Advisory Committee on Existing Chemicals of the Association of German Chemists (GDCh)

SIDS INITIAL ASSESSMENT PROFILE

CAS No.	110-93-0
Chemical Name	6-methylhept-5-en-2-one
Structural Formula	

SUMMARY CONCLUSIONS OF THE SIAR

Human Health

6-Methylhept-5-en-2-one was found to be of low toxicity after acute oral administration, skin contact and inhalation. The oral LD_{50} for the rat was 3,570 mg/kg bw. The main symptoms described were apathy, atonia, dyspnea, abdominal and lateral position (1,360 mg/kg bw or higher). A dose response was observed, with symptoms getting progressively worse with increasing doses and recovery occurred in survivors within 5 days at dosages of 4,250 mg/kg bw or lower. After inhalation of vapors of the substance, a LC_{50} of > 13.96 mg/l/4 hrs (> 13,960 mg/m^3/4hrs) could be estimated for rats using Haber's rule (LC_{50} > 6.98 mg/l/8 hrs, > 6,980 mg/m^3/8 hrs). The acute dermal LD_{50} for rabbits exceeded 5,000 mg/kg bw.

In rabbits, the undiluted substance was only slightly irritating to the skin and the eyes. Sensitization studies in guinea pigs (modified Draize protocol and Open Epicutaneous Test) did not reveal any indication of a skin sensitizing potential. In a human maximization test, the substance produced no skin sensitization in 25 volunteers at a test concentration of 3 % in petrolatum.

In a 90 day study (OECD TG 408), administration of 6-methylhept-5-en-2-one by gavage up to 1,000 mg/kg bw/day for 13 weeks caused substance-related effects in all dose groups. The target organs were kidney, liver and testes. Thus, under the conditions of this study, the no observed adverse effect level (NOAEL) was 50 mg/kg bw/day in females due to an increase of 21% in platelet counts at 200 mg/kg bw/day (LOAEL) and lower than 50 mg/kg bw/day in males due to an in increase of 12% and 14 % in relative and absolute kidney weights. The kidney effects in all dose groups in the males were induced by accumulation of α_{2u}-globulin which was confirmed by immunohistochemical staining. This finding is known to be a rat specific phenomenon without a toxicological correlate in humans.

No mutagenic effect was found in the Ames Test (OECD TG 471; standard plate and preincubation conditions) and *in vivo* in the mouse micronucleus test (OECD TG 474).

The results of a well conducted subchronic study with gavage administration of 6-methylhept-5-en-2-one indicate that the test compound caused testicular toxicity affecting spermatogenesis at the high dose level of 1,000 mg/kg bw/day. At the mid and low dose (50 and 200 mg/kg bw/day), no effects on sperm or testes were observed. In females, no adverse effects on reproductive organs or estrous cycle were observed up to and including the highest tested dosage of 1,000 mg/kg bw/day.
6-Methylhept-5-en-2-one was tested in a prenatal developmental toxicity study according to OECD TG 414 with gavage application. The no observed adverse effect level (NOAEL) for maternal and prenatal developmental toxicity was found at 200 mg/kg bw/day. Thus, signs of prenatal developmental toxicity in the form of mild growth retardation occurred only at a dose level which was also clearly toxic to the dams. There were no indications for teratogenicity up to and including 1,000 mg/kg bw/day.

Environment

The colorless-yellowish liquid 6-methylhept-5-en-2-one has a water solubility of 3.02 g/l (at 25 °C) and a vapor pressure of approximately 1 hPa at 18 °C. Based on measured data for vapor pressure and water solubility a Henry's

Law Constant of 6.68 Pa*m³/mole could be calculated, whereas by a model calculation a Henry's Law Constant of 21.5 Pa*m³/mole was derived. Distribution modeling using Mackay Level I indicates air (69 %) and water (30 %) to be the main targets. The substance is readily biodegradable according to OECD criteria (>70 % in 28 days; 10-days time window fulfilled; OECD 301F). Due to the chemical structure hydrolysis can be excluded. In the atmosphere 6-methylhept-5-en-2-one will be indirectly photodegraded by reaction with OH-radicals ($t_{1/2}$ = 4.2 hours) or ozone ($t_{1/2}$ = 38.4 minutes). Due to the measured $logK_{ow}$ of 2.4 (at 25 °C) and calculated $logK_{oc}$ of 1.57 and 2.04 a bio- or geoaccumulation is not to be expected.

The acute aquatic toxicity has been determined for the fish *Leuciscus idus* with a LC_{50} (96h) of 68 mg/l and for *Pimephales promelas* with a LC_{50} (96h) of 86 mg/l. Furthermore, for the waterflea *Daphnia magna* an EC_{50} (48h) of 129 mg/l and for the green alga *Scenedesmus subspicatus* an ErC_{50} (72h) of 191 mg/l (endpoint: growth rate) could be determined. Due to the moderate volatility of 6-methylhept-5-en-2-one an experiment for determining the evaporation from the test systems was performed and the effect values for fish, daphnids and algae were corrected. This resulted in an LC_{50} (96h) for *Leuciscus idus* of approximately 50 mg/l, an EC_{50} (48h) for *Daphnia magna* of approximately 83 mg/l and an ErC_{50} (72h) for *Scenedesmus subspicatus* of approximately 116 mg/l.
Results from prolonged or chronic studies are not available.
According to the EU Risk Assessment Procedure a $PNEC_{aqua}$ for the most sensitive aquatic species, the fish *Leuciscus idus*, of 50 µg/l can be calculated by applying an assessment factor of 1,000.

Exposure

The worldwide production volume of 6-methylhept-5-en-2-one in the year 2001 was between 10,000 and 30,000 t/a and has been reported to take place in closed systems.
In the Sponsor country 6-methylhept-5-en-2-one is to > 95 % used as an intermediate in closed systems for the synthesis of fine chemicals (e.g. vitamins, aroma chemicals, active ingredients used in pharmaceuticals). Only up to 5 % 6-methylhept-5-en-2-one is filled and distributed to industrial clients which are using it on the one hand as an intermediate for chemical syntheses and on the other hand as a flavouring compound and / or aroma additive in e.g. cosmetics and food. Monitoring data at the workplace are not available. However, worker protection in the Sponsor country is adequate and includes the use of appropriate technical equipment during substance handling and the use of protective equipment, etc. The risk of exposure to 6-methylhept-5-en-2-one may exist after spillages and during accidental exposure. Likewise dermal contact may result only from accidental exposure since the majority of the material (ca. 95%) is used as an intermediate in closed systems, and only small quantities (ca. 5%) are filled and distributed to industrial clients. No information is available on exposure scenarios following this use. Consumer exposure is widespread but anticipated to be low since only small amounts of 6-methylhept-5-en-2-one are contained in cosmetics at usual concentrations of up to 0.01 % and in food in maximum amounts ranging from 0.5 – 10 ppm. Some information suggests that there are products containing higher concentrations (of up to 2 %) that are assumed not to be available to the general public.
The substance naturally occurs as a biogenic volatile organic compound and shows an ubiquitous occurrence in the air due to emissions from plants or several herbs. It was also identified in several fruits as well as in drinking water- and in wastewater samples.
In European product registers products containing 6-methylhept-5-en-2-one are listed (cleaning/washing agents, cosmetics).
Exposure to workers is adequately controlled in the industry of the Sponsor country.

RECOMMENDATION

Human Health: The chemical is a candidate for further work.

Environment: The chemical is currently of low priority for further work.

RATIONALE FOR THE RECOMMENDATION AND NATURE OF FURTHER WORK RECOMMENDED

Environment:
The chemical possesses properties indicating a hazard for the environment. Although these hazards do not warrant further work as they are related to acute aquatic toxicity, which may become evident only at very high exposure levels, they should nevertheless be noted by chemical safety professionals and users.

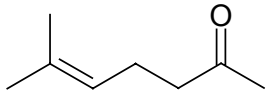
Human Health:
The chemical possesses properties indicating a hazard for human health. Testicular toxicity was induced in rats after repeated exposure at a dose of 1,000 mg/kg bw/day. Developmental effects at maternal toxic doses of 1000 mg/kg bw/day were observed. The main use is as a chemical intermediate predominantly in closed systems. However, up to 5 % of the substance are used outside the production site by industrial clients. An exposure assessment for this scenario is recommended, and, if indicated, a risk assessment should be performed.

SIDS Initial Assessment Report

1 IDENTITY

1.1 Identification of the Substance

CAS Number: 110-93-0
IUPAC Name: 6-methylhept-5-en-2-one
Molecular Formula: $C_8H_{14}O$
Structural Formula:



Molecular Weight: 126.2 g/mol
Synonyms: 6-methyl-5-hepten-2-one
2-methyl-6-oxo-2-heptene
5-hepten-2-one, 6-methyl- (8CI, 9CI)
6-methyl-δ-5-hepten-2-one
6-methyl-5-heptenone-2
sulcatone
Substance type: organic

Physical status: liquid

1.2 Purity/Impurities/Additives

Purity: approx. 98 - % w/w

Impurities: < 0.5 % (w/w) 6-methylhept-5-en-2-ol

< 0.5 % (w/w) 6-methylheptan-2-one

< 0.5 % (w/w) water

1.3 Physico-Chemical properties

6-Methylhept-5-en-2-one is a colorless-yellowish organic liquid with a strong odor (BASF AG, 2001a). The vapor pressure is 0.99 hPa and 1.99 hPa at 18.18°C and 27.99°C, respectively (BASF AG, 1999). The solubility in water was measured to be 3.02 g/l at 25°C (BASF AG, 1989a). A Henry's Law constant of 21.5 Pa*m³/mole at 25°C was calculated via HENRYWIN v3.10 (BASF AG, 2002a), whereas based on measured vapor pressure and water solubility data a HLC of 6.68 Pa*m³/mole could be calculated (Thomas, 1982).

The measured partition coefficient (log K_{ow}) was 2.4 (BASF AG, 1989b). The density of 0.851 g/cm³ at 20°C is slightly lower than that of water (Baglay et al., 1988).

The melting and boiling points of the substance are - 67.1°C – - 67.3°C and 172°C – 174°C at 1,014 hPa (Beilstein, 2003), respectively.

2 GENERAL INFORMATION ON EXPOSURE

In the year 2001 the world production volume of 6-methylhept-5-en-2-one was between 10,000 and 30,000 t/a (BASF AG, 2002b). Production ranges are 8,000 to 20,000 t/a for Europe (2 producers) and 2,000 – 10,000 t/a for Asia.

6-Methylhept-5-en-2-one is manufactured at BASF AG in a two-step synthesis using 3-methylbut-2-en-1-ol as starting substance in exclusively closed systems (BASF AG, 2002b).

In the Sponsor Country 6-methylhept-5-en-2-one is to more than 95 % used as intermediate in closed systems in the chemical industry for the synthesis of fine chemicals (e.g. vitamins, aroma chemicals, active ingredients used in pharmaceuticals). An amount of < 5 % is filled and distributed to industrial clients which are using the substance on the one hand as intermediate for chemical syntheses and on the other hand directly as a flavouring compound and/or aroma additive in e.g. cosmetics and food (BASF AG, 2002b). Worker protection is adequate and includes the use of appropriate technical equipment during substance handling and the use of protective equipment, etc. However, the risk of exposure to 6-methylhept-5-en-2-one may exist after spillages and during accidental exposure. Likewise dermal contact to the pure substance may result only from accidental exposure since the majority of the material (95 %) is used as an intermediate in closed systems, and only small quantities (ca. 5 %) are filled and distributed to industrial clients. Consumer exposure is low since only small amounts of 6-methylhept-5-en-2-one are contained in cosmetics at usual concentrations of up to 0.01 % and in food in maximum amounts ranging from 0.5 – 10 ppm. In the Swiss Product Register (2002), one unknown commercial product (not for consumer use) was listed for which a concentration range from 1 to 10 % 6-methylhept-5-en-2-one is indicated (product category cosmetics). The Danish Product Register (2002) mentions for Denmark 9 products (8 products cleaning and washing agents, 1 product not specified) with a concentration range of 0 – 2 %. For Norway the SPIN database (2003) mentions 4 preparations with use as cleaning/washing agent.

6-Methylhept-5-en-2-one naturally occurs as a biogenic volatile organic compound and shows an ubiquitous occurrence in the air due to emissions from plants, e.g. birch, or from several herbs (Ciccioli et al., 1993; Puxbaum, 1994). Further, it was found in several fruits such as apricots, apples and nectarines (Yajima et al., 1984; Takeoka et al., 1988; Mattheis et al., 1991; Gómez et al., 1993).

Releases into the environment may occur during production of 6-methylhept-5-en-2-one, during its use as chemical intermediate as well as from use of products containing the substance.

6-Methylhept-5-en-2-one was measured in the influent and the effluent of the waste water treatment plant of the BASF AG at regular intervals (24 h mixing samples) using HSGC. The concentration in the influent and in the effluent was always below the limit of quantification (influent: 0.5 mg/l; effluent: 0.05 mg/l) between 1st January 2001 and 31st August 2002 (BASF AG, 2002c). Based on the limit of detection and assuming worst case conditions less than 30 kg of 6-methylhept-5-en-2-one per day were released into the river Rhine during that period.

During production and internal processing at BASF AG Ludwigshafen (Germany) less than 25 kg/a were emitted into the air in the year 2000 (BASF AG, 2002d).

Emission data from other production and processing sites were not available.

2.1 Environmental Exposure and Fate

Distribution modeling using Mackay Level I (model V2.11) indicates air (69 %) and water (30 %) to be the main targets of 6-methyl-5-hepten-2-one. A very small part will be distributed into the soil and sediment with less than 1 % each (BASF AG, 2003b).

In the air the substance will be readily degraded according to the half-life time ($t_{1/2}$) of about 2.5 hours (measured rate constant; by Smith et al., 1996) and 4.2 hours (calculated rate constant AOP v1.90) for OH-radicals (concentration 500,000 mol/cm³) based on a 24 hours day and about 38.4 minutes for ozone molecules using the model AOP v1.90 (BASF AG, 2003c). Direct photolysis by sunlight is not relevant as the substance does not contain any functional group that would be expected to absorb light with wavelength > 290 nm

6-Methylhept-5-en-2-one is readily biodegradable according to OECD criteria. In the study using the Manometric Respirometry Test (OECD 301F) after 28 d the BOD was 91 % of the ThOD (BASF AG, 1995a). In this test within the 10 days time window more than 60 % of the test substance was degraded. Hydrolysis can be excluded due to the chemical structure of the compound.

The estimated log K_{oc} values using a) the model PCKOCWIN v1.66 and b) the equation according to TGD (2003) were 1.57 and 2.04, respectively. This indicates a very low adsorption of 6-methylhept-5-en-2-one to soil, sediments and suspended solids (BASF AG, 2002e).

No information about the bioaccumulation potential was available. Using the equation log BCF = 0.85*log K_{ow} – 0.70 as recommended in the TGD (May 2003) a BCF for fish of about 22 based on the measured log K_{ow} of 2.4 was calculated.

Due to its generally natural occurrence 6-methylhept-5-en-2-one can be measured in the air, in food and in the water. It was detected in oceanic and continental air samples in concentrations ranging from 20 – 400 pptv (September 1998, Mace Head, Ireland, average 123 pptv) (Sartin et al., 2001). In tropospheric samples of urban, suburban and forest areas in Italy 6-methylhept-5-en-2-one concentrations ranging from 0.08 – 5.44 ppbv were measured (Ciccioli et al., 1993). 6-methylhept-5-en-2-one was also found in beef and chicken meat (Shahidi et al., 1986). Further, the substance was detected in waters of lakes and rivers as a result of biotransformation processes in phytoplankton species (Jüttner, 1988 and 1992; Hayes and Burch, 1989; Cotsaris et al., 1995; Jones and Korth, 1995). 6-methylhept-5-en-2-one was also identified in 4 drinking water samples in the Netherlands with a maximum concentration of 0.01 µg/l and a detection limit of 0.005 µg/l (Zoeteman, 1980).

In a drinking water sample, taken from a reservoir in Spain, without any treatment 97.7 ng/l 6-methylhept-5-en-2-one were found (Aramendia et al., 1998). After treatment of the samples with chlorine and ozone only 5.4 ng/l could be determined, possibly due to the formation of new products. Additional treatment using physical processes like coagulation, flocculation and filtration did not completely remove the substance and resulted in similar values. In the same study 6-methylhept-5-en-2-one was found in wastewater samples from a purification plant at concentrations of 26.7 ng/l (plant failure) and 202.9 ng/l (plant worked properly). Further, 6-methylhept-5-en-2-one was identified as an ozone disinfection by-product (Richardson et al., 2000) as well as in river water samples after treatment with peracetic acid (Monarca et al., 2002).

The substance was also identified in biodegradable, in mixed household waste (< 0.1 mg/m³) and in garden waste (Wilkins, 1994; Wilkins and Larsen, 1996).

Additionally, 6-methylhept-5-en-2-one was identified in mandibular glands of the aphid hyperparasitoid wasp *Alloxysta brevis* (Völkl et al., 1994) and in defensive glands of the nymphalid butterfly *Agraulis vanillae* (Ross et al., 2001).

2.2 Human Exposure

No data on human workplace exposure are available. Exposure may occur during manufacture, transportation and industrial use. The likely primary routes of human exposure to 6-methylhept-5-en-2-one are skin contact and inhalation at the work place. Worker exposure in the Sponsor country is limited by enclosed systems, industrial hygiene controls and personal protective measures (protective gloves, safety glasses with side-shields, respiratory protection if ventilation is inadequate).

In the RIFM Monograph (1974) the usual concentrations in final consumer products are indicated to be 0.001 % (detergents), 0.01 % (soaps), 0.005 % (creams, lotions) and 0.04 % (perfumes). The substance was given GRAS (Generally Recognized As Safe) status by FEMA (1965) and is approved by FDA for food use. In Europe, it is included at a level of 1 ppm in the list of artificial flavoring substances that may be added to foodstuffs without hazard to public health (cited in RIFM Monograph 1974). The consumption as flavor is reported to be low (1995: 817 kg in Europe and 336 kg in US) (RIFM-FEMA database, 2002). Further, 6-methylhept-5-en-2-one is used in small quantities as a food flavor in maximum amounts ranging from 0.5 – 10.0 ppm in alcoholic beverages and gravies, respectively. It is also used in baked goods, frozen dairy, gelatin pudding, chewing gums, hard and soft candies and in non-alcoholic beverages at a maximum of ≤ 7.78 ppm. The cumulated intake of 6-methylhept-5-en-2-one from these types of food was estimated to be 0.54 mg per person and day according to the RIFM-FEMA database (RIFM-FEMA, 2002).

3 HUMAN HEALTH HAZARDS

3.1 Effects on Human Health

3.1.1 Toxicokinetics, Metabolism and Distribution

No specific studies are available concerning kinetic or metabolic fate of the substance. Based on the acute and the repeated dose studies, it can be concluded that the substance can be absorbed by the oral route. The centrolubular liver cell hypertrophy observed in the subchronic gavage study at a dosage of 1,000 mg/kg/d in rats suggests that the substance is metabolized by the liver (BASF AG, 2002f).

3.1.2 Acute Toxicity

In a non-guideline study (similar to OECD 401), the oral LD_{50} in rats (5 animals per sex and dose group) was found to be ca. 3,570 mg/kg bw (BASF AG, 1974). The clinical symptoms were described as apathy, atonia, dyspnea, abdominal and lateral position (1,360 mg/kg bw or higher). A dose response was observed, with symptoms getting progressively worse with increasing dose.Recovery occurred in survivors within 5 days at dosages of 4,250 mg/kg bw or lower. At necropsy, no abnormalities were detected in animals that were sacrificed after a post observation period of 7 days.

This result is broadly consistent with a further acute oral study in the rat which revealed a LD_{50} value of 4,100 mg/kg bw (Compadre et al., 1987).

Acute oral LD_{50} values in mice were reported as > 2,000, 2,410 and 3,609 mg/kg bw (Compadre et al., 1987; Migukina et al., 1988; Hoffmann-LaRoche, 1967).

LC_{50} rat (inhalation): > 13.96 mg/l/4 hrs (corresponding to 13,960 mg/m³/4 hrs), > 6.98 mg/l/8 hrs (corresponding to > 6,980 mg/m^3/8 hrs); estimated by Haber's Rule from an Inhalation Hazard Test which used a highly enriched/saturated vapor exposure system at 20°C, in which 12 rats were exposed to 6-methylhept-5-en-2-one vapor for 8 hours (calculated concentration 6,980 mg/m^3). No mortality was observed and clinical symptoms were limited to impaired balance which was reversible one day after the exposure (BASF AG, 1974).

The acute dermal LD_{50} exceeded 5,000 mg/kg bw in rabbits (Keating, 1972).

Conclusion:

6-Methyl-5-hepten-2-one was found to be of low acute toxicity after oral ingestion, skin contact and after inhalation. The oral LD_{50} for the rat was 3,570 mg/kg bw and the LD_{50} after dermal exposure was > 5,000 mg/kg bw for rabbits. From an inhalation risk test, a LC_{50} of > 13.96 mg/l/4hrs (> 13,960 mg/m^3/4hrs) can be estimated.

3.1.3 Irritation

Corrosiveness and Irritation

6-Methylhept-5-en-2-one was only slightly irritating to the skin of rabbits (4 animals) after an occlusive 20 hour application of the undiluted substance (BASF AG, 1974). The only finding was a slight redness of the skin which was fully reversible in all animals after 48 hours.

In the rabbit eye, slight corneal opacity and slight conjunctival redness 24 hours was observed after application of the undiluted substance (2 animals). All signs of irritation (including corneal opacity) were completely reversible within 8 days of observation (BASF AG, 1974).

Tested at 3 % in petrolatum, the substance produced no irritation after a 48 hours closed-patch test on human subjects (Kligman, 1972).

Conclusion:

Undiluted 6-methylhept-5-en-2-one was found to be only slightly irritating to the skin and the eyes of rabbits and not irritating when tested at 3 % in humans.

3.1.4 Sensitisation

6-Methyl-5-hepten-2-one was tested for its skin sensitizing potential in a non-guideline study using a modified Draize procedure (Sharp, 1978). 10 guinea pigs per group were used. For induction, 0.1 ml of the test substance at 0.25 % were injected intradermally at 4 sites which overlay the 2 auxillary and 2 inguinal lymph nodes. The animals were challenged 14 days later by an intradermal injection of 0.1 ml into one flank at the respective injection challenge concentration (0.1 %) and a topical application on the other flank at the application challenge concentration (20 %). Reactions were scored 24 hours later. If no sensitization reactions occurred, the procedure was repeated. 6-Methyl-5-hepten-2-one was reported to be a non-sensitizer under the conditions of the study.

An Open Epicutaneous Test was conducted with 6-methyl-5-hepten-2-one in guinea pigs (Klecak, 1985). The results of this study are available as secondary citation from a collection of data only. In a pretest the threshold irritating concentration of the test material was determined. Induction consisted of 21 daily open applications to the shaved flank of at least 6 guinea pigs per group. Open challenge applications were made on days 21 and 35. Reactions were read 24, 48 and 72 hours after

challenge. A substance was regarded allergenic if at least 1 of 6 animals of the respective concentration group showed positive results when non-irritating concentrations were used for challenge. 6-Methyl-5-hepten-2-one tested at a concentration of 3 % did not indicate a skin sensitizing potential.

A human maximization test was carried out on 25 volunteers. The material was tested at a concentration of 3 % in petrolatum and produced no sensitization reactions (Kligman, 1972).

Conclusion:

6-Methylhept-5-en-2-one was not a skin sensitizer in a guinea pig test according to a modified Draize procedure and in an Open Epicutaneous Test as well as in a human maximization test.

3.1.5 Repeated Dose Toxicity

A subchronic oral toxicity study was recently conducted with 6-methylhept-5-en-2-one under GLP conditions and according to OECD TG 408 (BASF AG, 2002f). The scope of examinations was extended to cover also effects on reproductive organs (see also chapter 3.1.7).

The compound was administered to groups of 10 male and 10 female Wistar rats by gavage for 13 weeks at dose levels of 0 (vehicle control), 50, 200 and 1,000 mg/kg bw/day. The vehicle used was olive oil and the administration volume was 5 ml/kg bw. Food consumption and body weight were determined weekly. Signs of toxicity or mortality were checked at least once a day after the treatment. Detailed clinical examinations in an open field were conducted prior to the start of the administration period and weekly thereafter. A functional observational battery (FOB) and measurement of motor activity was performed to the end of administration. Ophthalmological examinations were carried out in all animals before and in control and high dose animals at the end of the administration period. Vaginal smears for estrous cycle determination of all female animals were prepared and evaluated each day during the last 4 weeks of the study. Clinicochemical and hematological examinations as well as urinalyses were carried out towards the end of the administration period. Finally, all animals were assessed by gross pathology, followed by histopathological examinations.

The following treatment-related findings were obtained:

1,000 MG/KG BW/DAY:

- slight to moderate salivation in all animals of both sexes on several days from day 8 until the end of the study
- decreased food consumption (up to -13 %) in females from day 28 to day 49
- impairment of body weight and body weight change compared to controls in both sexes during the whole study (-7.2 % body weight in males at day 91 and -6.7 % in females at day 63; -16.4 % body weight change in females from day 35 to 84)
- decreased food efficiency in male animals on several study days (up to -56 % at day 77)
- increased platelets, calcium, total protein, albumin and cholesterol in both sexes
- decreased chloride and increased inorganic phosphate, urea, total bilirubin, globulins and magnesium in the females
- decreased aspartate aminotransferase in both sexes and slightly increased alkaline phosphatase in males

- urinalysis revealed cloudy specimens, urinary blood, renal tubular epithelial cells, degenerated transitional epithelial cells, granular casts and epithelial cell casts in the males
- significantly increased ketone levels in the urine specimens of males and females
- decreased spermatozoa in the cauda epididymis and spermatids in the testis and increased morphologically abnormal sperms in 3 out of 10 males
- diffuse tubular atrophy in the testes of three rats and focal tubular atrophy in two other rats and aspermia and debris in the lumen of the epididymides of three rats
- significantly increased mean absolute and relative liver and kidney weights in both sexes
- centrolobular hypertrophy of liver cells in almost all males and in all females
- increased accumulation of α_{2u}-globulin in the renal cortex of all male rats and multifocal dilation of renal tubular lumina in most males

200 MG/KG BW/DAY:

- increased calcium, total protein, albumin and cholesterol in the males and increased platelets in the females
- increased ketone levels in the urine specimens of the males
- significantly increased mean absolute and relative kidney weights in males
- increased accumulation of α_{2u}-globulin in the renal cortex of two male rats and multifocal dilation of renal tubular lumina in two males

50 MG/KG BW/DAY:

- significantly increased mean absolute and relative kidney weights in males
- increased accumulation of α_{2u}-globulin in the renal cortex of male rats

These observations are briefly discussed in the following:

The **clinical examination** of the animals revealed slight to moderate salivation in the high dose group in males and females which was related to the treatment but most likely caused by the intensive taste of the test compound and was regarded to be of minor toxicological importance. The decreased food consumption in females, the impairment of body weight and body weight change in both sexes during the whole study and finally, the decreased food efficiency in male animals on several study days were assessed as substance-related and obviously signs of general toxicity. The functional observational battery, the ophthalmological examinations and the estrous cycle determination did not reveal any treatment related effects.

Hematology revealed no treatment-related changes on white blood cell and red blood cell parameters. However, platelet counts were significantly increased in the high dose animals of either sex and in the mid dose females. Thrombocytosis usually is reactive or secondary to a disease process which is associated with various conditions, including inflammatory reactions, anemia etc. Since no corresponding changes were seen in the other clinical pathology examinations, the isolated finding of increased platelet counts was difficult to interpret in its original cause.

Serum enzyme examinations revealed reduced aspartate aminotransferase activities in the high dose animals of both sexes and slightly increased alkaline phosphatase activities in the high dose males, which both were considered to be test substance-related. In general, increases in serum

aspartate aminotransferase activities correlate well with hepatic diseases involving liver cell injury. Decreases of serum activities, however, are poorly understood and the assessment of enzyme reduction as an adverse toxic effect is questionable (Waner and Nyska, 1991). Since no adverse effects were observed in this study which could be associated with the fall in aspartate aminotransferase activities, a pathognomonic relevance was not assigned to this finding and was considered to be of no toxicological importance.

The slight increase in alkaline phosphatase activities in the serum of the high dose males was regarded not to represent any toxicologically relevant change, per se. Since changes in serum alkaline phosphatase activities in the rat are diet-dependent (Martins et al., 1998), the increase in enzyme activities is secondary and may be explained by the dependence of the enzyme on the nutritional state of the animals.

Changes in various blood chemistry parameters were also seen in the high dose animals and mid dose males. However, all these findings could not be assigned to a specific disease.

Urine analysis showed cloudy specimens and increased blood in the urine of the high dose males. Moreover, in the urine sediments of these animals increased numbers of degenerated renal tubular epithelial cells and transitional epithelial cells as well as granular and epithelial cell casts were detected. These findings are most likely the result of increased desquamation of tubular cells and increased excretion of mucoprotein which indicates damage of renal tubular epithelial damage. The increases in ketones are assessed to be not toxicologically relevant. Since the test substance itself is a ketone it is very likely that the test substance excreted in the urine interferes with the reagent strip method causing false positive results.

Sperm analysis revealed reduced number of spermatozoa in the cauda epididymis, decreased number of spermatids in the testis and increases in the percentage of morphologically abnormal sperms in 3 out of 10 male animals of the high dose group. These results suggest that the test compound caused testicular toxicity affecting spermatogenesis exclusively at a dose level of 1,000 mg/kg bw/day.

Concerning **pathology and histopathology**, treatment-related weight changes and microscopic findings were noted in the kidneys (significantly increased mean absolute and relative weights and diffuse accumulation of α_{2u}-globulin in the epithelia and tubular lumina of the proximal tubules of the renal cortex associated with multifocal cystic dilation of renal tubules of male rats). α_{2u}-globulin accumulation was confirmed by immunohistochemical staining. Degenerative or regenerative lesions were not noted in the proximal tubules of the renal cortex.

Under physiological conditions, α_{2u}-globulin is synthesized in the liver of male rats and excreted through the kidneys. Chemicals that bind to the protein can aggravate or prevent its excretion, thus accumulating a protein-chemical-complex in the cells of the proximal tubulus and increasing the kidney weight. Further lesions associated with and most likely related to α_{2u}-globulin accumulation, were multifocal dilation of tubular lumina. Normal female rats and higher species, such as humans, do not develop these changes and they are regarded as a specific phenomenon in male rats (see for review Haschek and Rousseaux, 1998). Therefore, α_{2u}-globulin accumulation is considered to have no toxicological impact on the human situation. Although no morphologic correlate was obtained for the significantly increased absolute and relative kidney weights in high dose females the weight increase was regarded to be treatment-related possibly due to increased metabolic activity of the renal cells associated with metabolism and/or excretion of the compound via the urine.

The centrolobular liver cell hypertrophy in most males and few females of the high dose group associated with increased mean absolute and relative liver weights is indicative for adaptive enzyme induction with the aim to increase the metabolizing and/or excretory capacity of the liver cells. As

long as no regressive cell or remarkable necrosis of liver cells occur, adaptive liver cell hypertrophy is in general reversible.

Although testicular atrophy was only noted in five animals of the high dose group and although in two cases the atrophy was only focal with an only minimal or slight degree of severity, this lesions were regarded treatment-related, as no such findings were recorded in control animals or the other dosed groups. A direct mode of action of the compound on the seminiferous epithelium was assumed, the stage of spermatogenesis, however, on which the toxic interference occurred, remained unknown.

All other findings were regarded to have developed unrelated to treatment as they were either single observations, or they occurred in control animals only, or they were recorded at low or comparable incidence and graded severity in control and high dose males and/or females.

In summary, the no observed adverse effect level (NOAEL) of this subchronic gavage study was 50 mg/kg bw/day in females due to increased platelet counts at 200 mg/kg bw/day (LOAEL) and lower than 50 mg/kg bw/day in males due to increased relative and absolute kidney weights.

Conclusion:

The administration of 6-methylhept-5-en-2-one by gavage up to 1,000 mg/kg bw/day for 13 weeks caused substance-related effects in all dosed groups. The target organs were kidney, liver and testes. Thus, under the conditions of this study, the no observed adverse effect level (NOAEL) was 50 mg/kg bw/day in females due to an increase of 21 % in platelet counts at 200 mg/kg bw /day (LOAEL) and lower than 50 mg/kg bw/day in males due to increase of 12 and 14 % in relative and absolute kidney weights. The kidney effects in all dose groups in the males were induced by accumulation of α_{2u}-globulin which is known to be a rat specific phenomenon without a toxicological correlate in humans.

3.1.6 Mutagenicity

In vitro Studies

6-Methyl-5-hepten-2-one was recently tested for its mutagenic potential at doses of up to 5,000 µg/plate in Salmonella strains TA1535, TA100, TA1537, TA98 and in E. coli WP2 uvrA with and without metabolic activation according to OECD TG 471 under GLP conditions (BASF AG, 2002g). An increase in the number of his^+ revertants was not observed in the standard plate test and in the preincubation test.

The substance was also negative in a further Ames test with and without S-9 mix using strains TA1535, TA100, TA1537 and TA98 when tested at a single concentration of 3 µmol/plate (ca. 378 µg/plate) (Florin et al., 1979).

In a forward mutation assay using Salmonella typhimurium strain TM677, 6-methylhept-5-en-2-one showed no mutagenic activity up to the highest tested concentrations of 5 mg/ml in presence and absence of metabolic activation (Compadre et al., 1987). The two latter studies are of less reliability as they did not follow guideline recommendations and the results are not sufficiently documented.

In vivo Studies

6-Methyl-5-hepten-2-one was tested for its ability to induce micronuclei in bone marrow erythrocytes in mice using two intraperitoneal doses up to 800 mg/kg bw/day under OECD TG 474 and GLP conditions (BASF AG, 2001b). This dose level produced in all treated animals evident signs of toxicity which were reversible after two days. At the two lower doses, only minor signs of clincal toxicity were observed after one hour of administration. The test substance did not have a

chromosome-damaging (clastogenic) effect and there were no indications of any impairment of chromosome distribution in the course of mitosis (aneugenic activity) in bone marrow cells in vivo.

Conclusion

6-Methyl-5-hepten-2-one gave no indication of a mutagenic effect in bacteria or a clastogenic potential in vivo. Therefore, there is no indication of a genotoxic potential in vivo.

3.1.7 Carcinogenicity

No specific study concerning the investigation of a carcinogenic potential is available.

3.1.8 Toxicity for Reproduction

Reproduction

Studies specifically designed to assess reproductive toxicity were not available for an assessment. However, in a recently well conducted subchronic oral toxicity study with 6-methyl-5-hepten-2-one, the scope of examinations was extended to cover also effects on reproductive organs (see also chapter 3.1.5; BASF AG, 2002f).

In this study, the substance was administered to groups of 10 male and 10 female Wistar rats for 13 weeks by oral gavage in olive oil at dose levels of 0 (vehicle control), 50, 200 and 1,000 mg/kg bw/day. At necropsy, the weights of the reproductive organs of the males (testes, epididymides, prostate gland) and females (ovaries, uterus) were assessed by gross pathology and a histopathological examination of the testes, epididymides, prostate gland and seminal vesicles, ovaries, uterus, oviducts and vagina was subsequently performed. Furthermore, immediately after necropsy, the right testis and cauda epididymis were taken from all male animals. Sperm motility, sperm morphology and sperm head count (cauda epididymis and testis) were examined.

Sperm analysis revealed reduced number of spermatozoa in the cauda epididymis, no spermatids per gramm testis and increases in the percentage of morphologically abnormal sperms in 3 out of 10 male animals of the high dose group. Testicular atrophy was only noted in five animals of the high dose group and although in two cases the atrophy was only focal with an only minimal or slight degree of severity it was regarded as treatment related. In the mid and low dose males, no effects on sperm or reproductive organs were observed. In the females, estrus cycle was not changed compared to controls at any dose and histopathology did not reveal any treatment related effects on sex organs.

Conclusion:

The results of the subchronic study with oral administration of 6-methylhept-5-en-2-one indicate that the test compound caused testicular toxicity affecting spermatogenesis at the high dose level of 1,000 mg/kg bw/day (LOAEL). At the mid and low dose, no effects on sperm or testes were observed (NOAEL 200 mg/kg bw/day). In females no adverse effects on reproductive organs or estrous cycle were observed up to and including the highest tested dosage of 1,000 mg/kg bw/day.

Developmental Toxicity

6-Methyl-5-hepten-2-one was recently tested for its prenatal developmental toxicity in Wistar rats according to OECD TG 414 and under GLP conditions (BASF AG, 2002h). The test substance was administered as an oily suspension to 25 time-mated female Wistar rats/group by stomach tube at doses of 50, 200 and 1,000 mg/kg bw/day on day 6 through day 19 post coitum (p.c.). A dose volume of 5 ml/kg bw was used for each group. The control group, consisting of 25 females, was

dosed with olive oil only. Food consumption and body weights of the animals were recorded regularly throughout the study period. The state of health of the animals was checked daily.

On day 20 p.c., all females were sacrificed and assessed by gross pathology (including weight determinations of the unopened uterus and the placentae). For each dam, corpora lutea were counted and number and distribution of implantation sites (differentiated as resorptions, live and dead fetuses) were determined. The fetuses were removed from the uterus, sexed, weighed and further investigated for any external findings. Thereafter, nearly one half of the fetuses of each litter was examined for soft tissue findings and the remaining fetuses for skeletal findings (incl. cartilage).

The treatment elicited clear signs of maternal toxicity at 1,000 mg/kg bw/day. Maternal toxicity was predominantly substantiated by adverse clinical findings (like abdominal position, unsteady gait and/or ataxia), statistically significant impairments in food consumption (about -7 % for the entire treatment period), and lowered absolute body weight (about -14 % for days 6 - 19) and corrected body weight gains compared to controls (about -29 % on day 20 p.c.). Moreover, all high dose and several mid dose rats showed transient salivation during the treatment phase. After cessation of treatment on day 19 p.c., however, salivation did not occur any longer in these rats. Salivation by itself was not assessed as an adverse or toxic effect. Thus, no signs of substance-induced maternal toxicity occurred at the low and the mid dose level.

There were no substance-related influences on the gestational parameters up to and including the highest dose level. Conception rate, mean number of corpora lutea, total implantations, resorptions and live fetuses, fetal sex ratio or the values calculated for the pre- and the postimplantation losses were unaffected by treatment.

Some signs of substance-induced prenatal developmental toxicity, but no indications for teratogenicity occurred exclusively at the high dose level. The mean placental and fetal body weights were diminished (-13 and –9 %, respectively if both sexes are combined). Correspondingly, the rates for certain skeletal variations (i.e. indications for delays in the ossification process) were significantly increased and outside historical control ranges. These variations mirror common findings on fetal morphology due to growth retardations, but are not indicative for selective effects on the fetal organism. No substance-induced signs of developmental toxicity were observed at 50 and 200 mg/kg bw/day. There were no indications for teratogenicity up to and including 1,000 mg/kg bw/day.

Conclusion:

Based on the results of this prenatal toxicity study with gavage application of 6-methyl-5-hepten-2-one, the no observed adverse effect level (NOAEL) for maternal and prenatal developmental toxicity was 200 mg/kg bw/day. Thus, signs of prenatal developmental toxicity in the form of mild growth retardation did only occur at a dose level, which was also clearly toxic to the dams. There were no indications for teratogenicity up to and including 1,000 mg/kg bw/day.

3.2 Initial Assessment for Human Health

6-Methyl-5-hepten-2-one was found to be of low toxicity after acute oral administration, skin contact and inhalation. The oral LD_{50} for the rat was 3,570 mg/kg bw. The main symptoms described were apathy, atonia, dyspnea, abdominal and lateral position (1,360 mg/kg bw or higher). A dose response was observed, with symptoms getting progressively worse with increasing doses and recovery occurred in survivors within 5 days at dosages of 4,250 mg/kg bw or lower. After inhalation of vapors of the substance, a LC_{50} of > 13.96 mg/l/4 hrs (> 13,960 mg/m³/4 hrs) could be estimated for rats using Haber's rule (LC_{50} > 6.98 mg/l/8 hrs, > 6,980 mg/m³/8 hrs). The acute

dermal LD_{50} for rabbits exceeded 5,000 mg/kg bw. In rabbits, the undiluted substance was only slightly irritating to the skin and the eyes. Sensitization studies in guinea pigs (modified Draize protocol and Open Epicutaneous Test) did not reveal any indication of a skin sensitizing potential. In a human maximization test, the substance produced no skin sensitization in 25 volunteers at a test concentration of 3% in petrolatum.

In a 90 days study (OECD TG 408), administration of 6-methylhept-5-en-2-one by gavage up to 1,000 mg/kg bw/day for 13 weeks caused substance-related effects in all dose groups. The target organs were kidney, liver and testes. Thus, under the conditions of this study, the no observed adverse effect level (NOAEL) was 50 mg/kg bw/day in females due to an increase of 21 % in platelet counts at 200 mg/kg bw/day (LOAEL) and lower than 50 mg/kg bw/day in males due to an increase of 12 and 14 % in relative and absolute kidney weights. The kidney effects in all dose groups in the males were induced by accumulation of α_{2u}-globulin which was confirmed by immunohistochemical staining. This finding is known to be a rat specific phenomenon without toxicological correlate in humans.

No mutagenic effect was found in the Ames Test (OECD TG 471; standard plate and preincubation conditions) and in vivo in the mouse micronucleus test (OECD TG 474).

The results of a well conducted subchronic study with gavage administration of 6-methylhept-5-en-2-one indicate that the test compound caused testicular toxicity affecting spermatogenesis at a dose level of 1,000 mg/kg bw/day. At the mid and low dose (50 and 200 mg/kg bw/day), no effects on sperm or testes were observed. In females, no adverse effects on reproductive organs or estrous cycle were observed up to and including the highest tested dosage of 1,000 mg/kg bw/day.

6-Methyl-5-hepten-2-one was recently tested in a prenatal developmental toxicity study according to OECD TG 414 with gavage application. The no observed adverse effect level (NOAEL) for maternal and prenatal developmental toxicity was found at 200 mg/kg bw/day. Thus, signs of prenatal developmental toxicity the form of mild growth retardation only occurred at a dose level which was also clearly toxic to the dams. There were no indications for teratogenicity up to and including 1,000 mg/kg bw/day.

4 HAZARDS TO THE ENVIRONMENT

4.1 Aquatic Effects

Acute Toxicity Test Results

The most sensitive studies available were considered to evaluate the toxicity on aquatic organisms of 6-methylhept-5-en-2-one.

Fish

In a static acute test, following the German DIN 38 412, to evaluate the toxicity of 6-methylhept-5-en-2-one to the golden orfe, *Leuciscus idus*, 5 concentrations ranging from 46.6 – 1,000 mg/l (nominal) plus a untreated control were tested. A LC_{50} (96h) of 68 mg/l (nominal, geometric mean of LC_0 at 46.6 mg/l and LC_{100} at 100 mg/l) could be calculated. At concentrations of 100 mg/l and above all fish were dead after 4 hours of exposure, while at the lowest test concentration (46.6 mg/l) no fish died within the test period of 96 hours. Within this test no symptoms like gasping or tumbling could be observed (BASF AG, 1989c).

To evaluate the toxicity of 6-methylhept-5-en-2-one to the fathead minnow, *Pimephales promelas*, 5 concentrations ranging from 51 – 394 mg/l (nominal) plus an untreated control were tested in a

flow-through system (Brooke et al., 1984). Based on analytical measurements a LC_{50} (96h) of 86 mg/l (83.3 – 88.2 mg/l) was calculated.

Invertebrates

In a static acute test, following the German DIN 38 412, to evaluate the toxicity of 6-methylhept-5-en-2-one to the waterflea, *Daphnia magna*, 5 concentrations ranging from 58 – 580 mg/l (nominal) were tested. An EC50 for the endpoint immobilization (48h) of 129 mg/l (nominal) was derived (BASF AG, 1990).

Algae

The acute toxicity of 6-methylhept-5-en-2-one to the green alga *Scenedesmus subspicatus*, following the German DIN 38 412, was determined using 6 concentrations ranging from 10 – 500 mg/l (nominal). The ErC50 (72h) for the endpoint growth rate was 191 mg/l (nominal) and the EbC50 (72h) for the endpoint biomass was 208 mg/l (nominal). The ErC10 (72h) was 30 mg/l and the EbC10 (72h) was 31 mg/l) (nominal) (BASF AG, 1989d).

For all aquatic toxicity tests performed at BASF AG static, open test systems were used. Since no substance specific concentration control analysis was performed, the effect values are related to the nominal concentrations. However, due to the vapor pressure of 6-methylhept-5-en-2-one and its moderate volatility an evaporation from the open test systems was likely to have occurred. Therefore, the volatility was determined by measuring the remaining test substance using TOC measurements under comparable test conditions but without the corresponding test organisms (BASF AG, 2003d). Two tests using 100 mg/l and 200 mg/l 6-methylhept-5-en-2-one and two parallels for each test system were run. The values obtained after the corresponding test periods were as follow (Table 1,2):

Table 1: Evaporation of 100 mg/l 6-methylhept-5-en-2-one in the different test systems

Species	**Test period**					**Geometric mean**
	0h	24h	48h	72h	96h	
L. idus	100 %	81 %	73 %	70 %	67 %	77 %
D. magna	100 %	65 %	50 %	/	/	69 %
S. subspicatus	100 %	69 %	52 %	42 %	/	62 %

Table 2: Evaporation of 200 mg/l 6-methylhept-5-en-2-one in the different test systems

Species	**Test period**					**Geometric mean**
	0h	24h	48h	72h	96h	
L. idus	100 %	82 %	66 %	65 %	61 %	74 %
D. magna	100 %	64 %	41 %	/	/	64 %
S. subspicatus	100 %	64 %	50 %	42 %	/	61 %

As to be seen the evaporation, on the basis of the recovery rates found by TOC measurements in the different approaches is comparable. In the test system for the fish the concentration dropped from the beginning until the end to approximately 74 % and 77 %, which result in a effective LC_{50} (96h)

of 50 mg/l and 53 mg/l, respectively. In the test system for daphnids an effective EC_{50} (48h) of 83 mg/l and 89 mg/l could be derived based on the remained test substance of 64 % and 69 %. In the test system for the algae after 72 h only 61 % and 62 % of 6-methylhept-5-en-2-one could be found. This results in a corrected ErC_{50} (72h) of approximately 116 mg/l and 119 mg/l as well as in EbC_{50} (72h) of 126 mg/l and 130 mg/l, respectively (BASF AG, 2003e).

Chronic Toxicity Test Results

No data on chronic aquatic toxicity were available.

PNEC

Using the aquatic toxic effect on the most sensitive species, *Leuciscus idus*, a $PNEC_{aqua}$ of 50 µg/l is derived by applying an assessment factor of 1,000 according to the EU Technical Guidance Document.

4.2 Terrestrial Effects

The acute oral toxicity for the red-winged blackbird, *Agelaius phoeniceus*, was investigated (Schafer et al., 1983). Over a test period of 18 hours a LD_{50} of > 111 mg/kg bw based on food consumption data could be derived. No information about dosage or application procedure was available.

4.3 Other Environmental Effects

Microorganisms

In an oxygen consumption inhibition test according to Robra the toxicity of 6-methylhept-5-en-2-one on the aquatic bacterium *Pseudomonas putida* was tested using 5 concentrations ranging from 625 – 10,000 mg/l. An EC_{10} (0.5h) of 1,800 mg/l and an EC_{50} (0.5h) of 3,000 mg/l was observed (BASF AG, 1988).

In a Microtox® toxicity assay EC_{50} (5 min) of 17.5 mg/l for *Photobacterium phosphoreum* was determined (Curtis et al., 1982).

In a respiration inhibition test following the OECD 209 the inhibition of 6-methylhept-5-en-2-one on activated sludge from laboratory waste water treatment plants treating municipal sewage was investigated. The resulting EC_{20} (0.5h) was approximately 27 mg/l and the EC_{50} (0.5h) was 800 mg/l (BASF AG, 1995b).

4.4 Initial Assessment for the Environment

The colorless-yellowish liquid 6-methylhept-5-en-2-one has a water solubility of 3.02 g/l, a vapor pressure of approximately 1 hPa at 18 °C. The measured log K_{ow} is 2.4 and the calculated log K_{oc} is 1.57. Thus, bio- and geoaccumulation are not to be expected. Based on measured data a Henry's Law Constant of 6.68 Pa*m³/mole could be calculated, whereas by a model calculation a HLC of 21.5 Pa*m³/mole was derived. Distribution modeling using the Mackay Level I indicates air (69 %) and water (30 %) to be the main targets. The substance is readily biodegradable according to OECD criteria. Hydrolysis is not expected due to the structure of the chemical. In the atmosphere 6-methylhept-5-en-2-one will be indirectly photodegraded by reactions with OH radicals (calculated $t_{1/2}$ of 4.2 h) or ozone (calculated $t_{1/2}$ of 38.4 minutes).

The acute aquatic toxicity has been determined for fish (*Leuciscus idus*: LC_{50} (96h) 68 mg/l), for invertebrates (*Daphnia magna*: EC_{50} (48h) 129 mg/l) and for a green alga (*Scenedesmus*

subspicatus: ErC_{50} (72h) 191 mg/l). Based on the moderate volatility of 6-methylhept-5-en-2-one the effect values were corrected as follows: *Leuciscus idus* LC_{50} (96h) ca. 50 mg/l, *Daphnia magna* EC_{50} (48h) ca. 83 mg/l, *Scendesmus subspicatus* EC_{50} (72h) ca. 116 mg/l. Results from prolonged or chronic studies are not available. Following the EU Risk Assessment Procedure a $PNEC_{aqua}$ of 50 µg/l can be calculated by applying an assessment factor of 1,000 on the fish, which was found to be the most sensitive species.

5 RECOMMENDATIONS

Environment: The chemical is currently of low priority for further work. 6-Methylhept-5-en-2-one possesses properties indicating a hazard for the environment. Although these hazards do not warrant further work as they are related to acute aquatic toxicity, which may become evident only at very high exposure levels, they should nevertheless be noted by chemical safety professionals and users.

Human Health: The chemical is a candidate for further work. 6-Methylhept-5-en-2-one possesses properties indicating a hazard for human health. Testicular toxicity was induced in rats after repeated exposure at a dose of 1,000 mg/kg bw/day. Developmental effects at maternal toxic doses of 1,000 mg/kg bw/day were observed. Main use is as a chemical intermediate predominantly in closed systems. However, up to 5 % of the substance are used outside the production site by industrial clients. An exposure assessment for this scenario is recommended, and, if indicated, a risk assessment should be performed.

6 REFERENCES

Aramendia MA, Borau V, Garcia I, Jimenez C, Lafont F, Marinas JM and Urbano FJ (1998). Determination of volatile organic compounds in drinking and waste water from Cordoba (Spain) by closed-loop stripping analysis in combination with gas chromatography coupled with mass spectrometry, Toxicol. Environ. Chem. **57**, 9-25.

Baglay AK, Gurarly LL and Kuleshov GG (1988). Physical properties of compounds used in vitamin synthesis, J. Chem. Eng. Data **33**, 512-518.

BASF AG (1974). Product Safety, Ergebnis der gewerbetoxikologischen Vorprüfung, unpublished study, XXIII/401, 04 Sept. 1974.

BASF AG (1988). Product Safety, Bacterial oxygen consumption test: 6-methylhept-5-en-2-one, unpublished summary-reprint of a test report, 10/0905/88, 11 Aug. 1988.

BASF AG (1989a). Analytical Laboratory, Bestimmung der Löslichkeit der Prüfsubstanz in Wasser bei 25°C, unpublished study, 106288/13, 02 May 1989.

BASF AG (1989b). Analytical Laboratory, Bestimmung des Verteilungskoeffizienten log Pow der Prüfsubstanz in 1-Octanol/Wasser bei 25°C, unpublished study, J. No. 106288/13, 03 May 1989.

BASF AG (1989c). Product Safety, Bericht über die Pruefung der akuten Toxiziät an der Goldorfe (Leuciscus idus L., Goldvariante), unpublished study, 10F0602/885325, 06 June1989.

BASF AG (1989d). Product Safety, Hemmung der Algenzellvermehrung nach DIN 38412 L9: Methylheptenon, unpublished study, 01/89/1480,, 15 Dec. 1989.

BASF AG (1990). Product Safety, Akute Toxizität für Daphnien n. DIN 38412 L 11: Methylheptenon, unpublished study, 01/89/1480 15 Jan. 1990.

BASF AG (1995a). Product Safety, Pruefung der biologischen Abbaubarkeit von 2-Methylhepten-2-on-6 im manometrischen Respirationstest, unpublished study, 95/0224/26/2, 28th April - 26th May 1995.

BASF AG (1995b). Product Safety, Prüfung der Atmungshemmung von Belebtschlamm durch 2-Methylhepten-2-on-6 im Kurzzeitatmungshemmtest, unpublished study, 95/0224/08/1, 08 May 1995.

BASF AG (1999). Substance data service, Dampfdruck von 2-Methyl-2-hepten-6-on, unpublished study, report No. 99.271, 06 April 1999.

BASF AG (2001a). Safety data sheet, 15 Nov. 2001.

BASF AG (2001b). Product safety, Cytogenetic study in vivo, mouse micronucleus test after two intraperitoneal administrations with 6-Methylhept-5-en-2-one (methylheptenon), unpublished study, 26M0874/004151, 00/0874, 10 July 2001.

BASF AG (2002a). Product Safety, SRC HENRYWIN v3.10, data assessment, 16 July 2002.

BASF AG (2002b). Internal data, pers. communications, October 2002.

BASF AG (2002c). Unpublished data, effluent concentration between 1st January – 31st August 2002, 10 Sept. 2002

BASF AG (2002d). unpublished data (emission of 2-methyl-2-hepten-6-on cited in the German Emission Register 2000), 04 Sept. 2002.

BASF AG (2002e). Product Safety, data assessment, SRC PCKOCWIN v1.66, 16 July 2002.

BASF AG (2002f). Product Safety, 6-Methylhept-5-en-2-one (methylheptenon) – Subchronic toxicity Study in Wistar rats, administration by gavage for 3 months, unpublished study 51S0874/00114, 00/0874, 29 Oct. 2002.

BASF AG (2002g). Product Safety, *Salmonella typhimurium* / **Escherichia coli** reverse mutation assay (standard plate test and preincubation test) with 6-Methylhept-5-en-2-one (methylheptenon), unpublished study, 40M0874/004193, 00/0874 18 April 2002.

BASF AG (2002h). Product Safety, 6-Methylhept-5-en-2-one (methylheptenon) - Prenatal developmental toxicity study in Wistar rats, oral administration (gavage), unpublished study, 30R0874/00111, 00/0874 29 Oct. 2002.

BASF AG (2003b). Product Safety, data assessment, Mackay Level I V2.11, 02 July 2003.

BASF AG (2003c). Product Safety, SRC AOP v1.90, data assessment, 01 July 2003.

BASF AG (2003d). Test on the volatility of 6-Methylhept-5-en-2-one (CAS: 110-93-0) in an algae, daphnia and fish test, unpublished data, 00/0874-2, 07 July 2003.

BASF AG (2003e). Calculation of recovery rates of 100 mg and 200 mg 6-methylhept-5-en-2-one from its volatility in an algae, daphnia and fish test, data assessment, 07 July 2003.

Beilstein (2003). Handbook (online), BRN 1741705, BPR 110-93-0, update date: 07 May 1999, printing date: 03 July 2003.

Brooke LT, Call DJ, Geiger DL and Northcott CE (1984). Acute toxicities of organic chemicals to fathead minnows (*Pimephales promelas*), Center for Lake Superior Environmental Studies, University of Wisconsin-Superior.

Ciccioli P, Brancaleoni E, Frattoni M, Cecinato A and Brachetti A (1993). Ubiquitous occurrence of semi-volatile carbonyl compounds in tropospheric samples and their possible sources, Atmos. Environ. **27**A, 1891-1901.

Compadre CM, Hussain RA, de Compadre RLL, Pezzuto JM, Kinghorn AD (1987). The intensely sweet sesquiterpene hernandulcin: Isolation, synthesis, characterization, and preliminary safety evaluation, J. Agric. Food Chem. **35**, 273-279.

Cotsaris E, Bruchet A, Mallevialle J and Bursill DB (1995). The identification of odorous metabolites produced from algal monocultures, Wat. Sci. Tech. **31**, 251-258.

Curtis C, Lima A, Lozano SJ and Veith GD (1982). Evaluation of a bacterial bioluminescence bioassay as a method for predicting acute toxicity of organic chemicals to fish, ASTM STP 766, American for Testing and Materials, Philadelphia, pp 170-178.

Danish Product Register (2002). Personal communication, 2002.

Florin I, Rutberg L, Curvall M, Enzell CR (1979). Screening of tobacco smoke constituents for mutagenicity using the Ames' Test, Toxicology **15**, 219-232.

Gómez E, Ledbetter CA and Hartsell PL (1993). Volatile compounds in apricot, plum and their interspecific hybrids, J. Agric. Food Chem. **41**, 1669-1676.

Haschek WM and Rousseaux CG (1998). Fundamentals of toxicologic pathology, Chapter 7: The Kidney, Academic Press, San Diego, USA, pp 173-176.

Hayes KP and Burch MD (1989). Odorous compounds associated with algal blooms in South Australian waters, Wat. Res. **23**, 115-121.

Hoffmann-LaRoche (1967). Acute toxicity, eye and skin irritation test of fragrance materials, unpublished results, 1967 cited in RIFM-FEMA data base, Oct. 2002.

Jones GJ and Korth W (1995). In situ production of volatile odour compounds by river and reservoir phytoplankton populations in Australia, Wat. Sci. Tech. **31**, 145-151.

Jüttner FR (1988). Biochemistry of biogenic off-flavour compounds in surface waters, Wat. Sci. Tech. **20**, 107-116.

Jüttner FR (1992). Flavour compounds in weakly polluted rivers as a means to differentiate pollution sources, Wat. Sci. Tech. **25**, 155-164.

Kligman AM (1972). Report to RIFM, 19 July, cited in Food Cosm. Toxicol. **13**, 859, 1974.

Keating JW (1972). Report to RIFM cited in Opdyke, Fd. Cosm. Toxicol. **13**, 859, 1975.

Klecak G (1985). The Freund's complete adjuvant test and the Open Epicutaneous Test, Curr. Probl. Derm., vol. **14**, 152-171, Karger, Basel.

Martins MJ, Dias PO and Hipólito-Reis (1998). Rat serum alkaline phosphatase electrophoretic fractions: variations with feeding, starvation and cellulose fibre ingestion, Clinical Nutrition **17** (6), 279-285.

Mattheis J.P., Fellmann J.K., Chen P.M. and M.E. Patterson (1991). Changes in headspace volatiles during physiological development of Bisbee delicious apple fruit, J. Agric. Food Chem. **39**, 1902-1906.

Migukina NB, Lipanina TB, Maltzewa NM, Stepkina LN, Schumarina NB, Karpuchina EA,and Kajutina CB (1988). To the validation of tentative safe level of methylheptenon exposure in the work zone air, Gigiena Truda i Professional'nye Zabolevaniya. Labor Hygiene and Occupational Diseases **32** (88), 52-53.

Monarca S, Richardson SD, Feretti D, Grottolo M, Thruston jr AD, Zani C, Navazio G, Ragazzo P, Zerbini I and Alberti A (2002). Mutagenicity and disinfection by-products in surface drinking water disinfected with peracetic acid, Environ. Toxicol. Chem **21**(2), 309-318.

Puxbaum H (1994). GC-MS-Analytik biogener VOC-Emissionen, Ö. Chem. Z. **9**, 220-226.

Richardson SD, Thruston jr AD, Caughran TV, Chen PH, Collette TW, Schenck K.M., Lykins jr BW, Rav-Acha C and Glezer V (2000). Identification of new drinking water disinfection byproducts from ozone, chlorine dioxide, chloramine, and chlorine, Wat. Air Soil Pollut. **123**, 95-102.

RIFM Monograph (1974). Methyl heptenone, Food Cosm. Toxicol. **13**, 859.

RIFM-FEMA (2002). RIFM (Research Institute for Fragrance Materials, USA) - FEMA (Flavor and Extract manufacturers' Association of the US) database, Oct. 2002.

Ross GN, Fales HM, Lloyd HA, Jones T, Sokoloski EA, Marshall-Batty K and Blum MS (2001). Novel chemistry of abdominal defensive glands of nymphylid butterfly Agraulis vanillae, J. Chem. Ecol. **27**, 1219-1228.

Sartin JH, Halsall CJ, Davison B, Owen S and Hewitt CN (2001). Determination of biogenic volatile organic compounds (C_8–C_{16}) in the coastal atmosphere at Mace Head, Ireland, Anal. Chim. Acta **428**, 61-72.

Schafer EW, Bowles jr WA and Hurlbut J (1983). The acute oral toxicity, repellency, and hazard potential of 998 chemicals to one or more species of wild and domestic birds, Arch. Environ. Contam. Toxicol. **12**, 355-382.

Shahidi F, Rubin LJ and D`Souza LA (1986). Meat flavor volatiles: a review of the composition, techniques of analysis and sensory evaluation, CRC Crit. Rev. Food Sci. Nutrition **24**, 141-243.

Sharp DW (1978). The sensitization potential of some perfume ingredients tested using a modified Draize procedure, Toxicology **9**, 261-271.

Smith AM, Rigler E, Kwok ESC and Atkinson R (1996). Kinetics and products of the gas-phase reactions of 6-Methyl-5-hepten-2-one and trans-cinnamaldehyde with OH and NO3 Radicals and O3 at 296+/-2K, Environ. Sci. Technol. **30**, 1781-1785.

SPIN database (2003)., http://195.215.251.229/spin.html, query June 2003.

Swiss Product Register (2002). Pers. communication, P. Bormann, 2002.

Takeoka GR, Flath RA, Güntert M and Jennings W (1988). Nectarine volatiles: vacuum steam distillation versus headspace sampling, J. Agric. Food Chem. **36**, 553-560.

TGD (2003). Technical Guidance Document , European Commission, May 2003.

Thomas RG (1982). Volatilization from water, In: Lyman, W.J., Reehl, W.F. and D.H. Rosenblatt (eds.), Handbook of Chemical Property Estimation Methods. Environmental Behavior of Organic Compounds, pp 15-1 - 15-34, American Chemical Society, Washington DC, USA, 1982.

Völkl W, Hübner G and Dettner K (1994). Interactions between *Alloxysta brevis* (Hymenoptera, Cynipoidea, Alloxystidae) and honeydew-collecting ants: how an aphid hyperparasitoid overcomes ant aggression by chemical defense, J. Chem. Ecol. **20**, 2901-2915.

Waner T and Nyska A (1991). The toxicological significance of decreased activities of blood alanine and aspartete aminotransferase, Veterinary Research Communications **15**, 73-78.

Wilkins K (1994). Volatile organic compounds from household waste, Chemosphere **29**, 47-53.

Wilkins K and Larsen K (1996). Volatile organic compounds from garden waste, Chemosphere **32**, 2049-2055.

Yajima I, Yanai T, Nakamura M, Sakakibara H and Hayashi K (1984). Volatile flavor components of Kogyoku Apples, Agric. Biol. Chem. **48**, 849-855.

Zoeteman B (1980). Sensory assessment of water quality, Pergamon Press, Oxford.

ANNEX: DETAILS OF THE LITERATURE SEARCH USED

The data banks searched are indicated below.

Toxicology

Date of last literature search: 08 December 2002

JETOC

RTECS

AGRICOLA

CABA

CANCERLIT

TOXCENTER

TOXLINE

JICST-EPLUS

LIFESCI

TOXLIT

EMBASE

ESBIOBASE

EMBAL

HEALSAFE

CSNB

MEDLINE

RIFM-FEMA database

IRIS

ATSDR TOX. PROFILES

atsdr TOX: FAQS

chemfinder

civs

gestis

ginc

nicnas

ntp

Ecology

Date of last literature search: 11 April 2003

AQUASCI

BIOSIS

EMBASE

ESBIOBASE.

LIFESCI

OCEAN

POLLUAB

SCISEARCH

TOXCENTER

TOXLINE

ULIDAT

datalog

chemfate

biodeg

aquire

HSDB

I U C L I D

D a t a S e t

Existing Chemical ID: 110-93-0
CAS No. 110-93-0
EINECS Name 6-methylhept-5-en-2-one
EC No. 203-816-7
Molecular Weight 126.2
Molecular Formula C8H14O

Producer Related Part
Company: BASF AG
Creation date: 18-FEB-1992

Substance Related Part
Company: BASF AG
Creation date: 18-FEB-1992

Memo: master

Printing date: 16-APR-2004
Revision date:
Date of last Update: **16-APR-2004**

Number of Pages: **111**

Chapter (profile): Chapter: 1, 2, 3, 4, 5, 6, 7, 8, 10
Reliability (profile): Reliability: without reliability, 1, 2, 3, 4
Flags (profile): Flags: without flag, SIDS

1.0.1 Applicant and Company Information

Type: lead organisation
Name: BASF AG
Contact Person: Dr. Hubert Lendle **Date:**
GUP/CL - Z570
Street: Carl-Bosch-Str
Town: 67056 Ludwigshafen
Country: Germany
Phone: +49 621 60 44712
Telefax: +49 621 60 58043
Email: hubert.lendle@basf-ag.de
Homepage: www.basf.com

Flag: Critical study for SIDS endpoint
22-JUL-2002

Type: cooperating company
Name: Hoffmann-La Roche Ltd.
Country: Switzerland

Flag: Critical study for SIDS endpoint
31-OCT-2002

Type: cooperating company
Name: Kuraray
Country: Japan

Flag: Critical study for SIDS endpoint
31-OCT-2002

1.0.2 Location of Production Site, Importer or Formulator

1.0.3 Identity of Recipients

1.0.4 Details on Category/Template

1.1.0 Substance Identification

Mol. Formula: C8 H14 O
Mol. Weight: 126.20 g/mol

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002

1.1.1 General Substance Information

Purity type: typical for marketed substance
Substance type: organic
Physical status: liquid
Purity: ca. 98 - % w/w
Colour: colourless - yellowish
Odour: strong

Flag: non confidential, Critical study for SIDS endpoint
16-SEP-2002 (1) (2)

1.1.2 Spectra

1.2 Synonyms and Tradenames

2-Methyl-2-hepten-6-one

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

2-Methyl-6-oxo-2-heptene

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

2-Methylhepten-2-on-6

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

2-Methylheptenone

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002

2-Oxo-6-methylhept-5-ene

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

5-Hepten-2-one, 6-methyl- (8CI, 9CI)

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

6-Methyl-.DELTA.5-hepten-2-one

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

6-Methyl-5-hepten-2-one

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

6-Methyl-5-heptenone-2

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002

Sulcatone

Flag: non confidential, Critical study for SIDS endpoint
19-FEB-1992

1.3 Impurities

CAS-No: 1569-60-4
EC-No: 216-377-1
EINECS-Name: 6-methylhept-5-en-2-ol
Mol. Formula: C8 H16 O

Remark: typically less than 0.5 % w/w.
Flag: non confidential, Critical study for SIDS endpoint
08-JUL-2003 (3)

CAS-No: 928-68-7
EC-No: 213-179-7
EINECS-Name: 6-methylheptan-2-one
Mol. Formula: C8 H16 O

Remark: typically less than 0.5 % w/w.
Flag: non confidential, Critical study for SIDS endpoint
08-JUL-2003 (3)

CAS-No: 7732-18-5
EC-No: 231-791-2
EINECS-Name: water
Mol. Formula: H2 O

Remark: typically less than 0.5 % w/w.
Flag: non confidential, Critical study for SIDS endpoint
08-JUL-2003 (3)

1.4 Additives

1.5 Total Quantity

Remark: production volumes (year 2001):

Germany: 3.000 - 10.000 t/a
Europe: 8.000 - 20.000 t/a
USA: 0 t/a
Asia: 2.000 - 10.000 t/a

World: 10.000 - 30.000 t/a
Flag: Critical study for SIDS endpoint
31-OCT-2002

1.6.1 Labelling

Labelling: no labelling required (no dangerous properties)

Flag: non confidential, Critical study for SIDS endpoint
16-SEP-2002 (2)

1.6.2 Classification

Classified: no classification required (no dangerous properties)

Flag: non confidential, Critical study for SIDS endpoint
16-SEP-2002 (2)

1.6.3 Packaging

1.7 Use Pattern

Type: industrial
Category: Chemical industry: used in synthesis

Flag: non confidential, Critical study for SIDS endpoint
31-OCT-2002

Type: use
Category: Intermediates

Remark: In the sponsor country > 95 % of the production volume of 6-methylhept-5-en-2-one is used as intermediate in closed systems in the chemical industry for the synthesis of fine chemicals (e.g. vitamins, aroma chemicals, active ingredients used in pharmaceuticals).
Flag: non confidential, Critical study for SIDS endpoint
30-JAN-2004 (4)

Type: industrial
Category: Personal and domestic use

Remark: 9 products (8 products cleaning and washing agents, 1 product not specified) with a concentration range of 0 - 2 % are known.
Flag: non confidential, Critical study for SIDS endpoint
30-JAN-2004 (5)

Type: type
Category: Use in closed system

Flag: non confidential, Critical study for SIDS endpoint
15-APR-2004

Type: use
Category: other: cosmetics and food/foodstuff additives

Remark: In the Sponsor Country < 5 % is filled and distributed to industrial clients which are using the substance on the one hand as intermediate for chemical syntheses and on the other hand directly as a flavouring compound and/or aroma additive in e.g. cosmetics and food.
Flag: non confidential, Critical study for SIDS endpoint
30-JAN-2004 (4)

1.7.1 Detailed Use Pattern

1.7.2 Methods of Manufacture

Orig. of Subst.: Synthesis
Type: Production

Remark: Addition of acetylene to acetone results in the formation of 3-methyl-1-butyn-3-ol, which is hydrogenated to 3-methyl-1-buten-3-ol in the presence of a palladium catalyst. This product is converted into its acetoacetate derivative with diketene or with ethyl acetoacetate. The acetoacetate undergoes rearrangement when heated (Carroll reaction) to give 2-methyl-2-hepten-6-one.
Flag: non confidential, Critical study for SIDS endpoint
31-OCT-2002 (6)

Orig. of Subst.: Synthesis
Type: Production

Remark: In another process, 2-methyl-2-hepten-6-one is obtained by reaction of 3-methyl-1-buten-3-ol with isopropenyl methyl ether followed by a Claisen rearrangement.
Flag: non confidential, Critical study for SIDS endpoint
31-OCT-2002 (6)

Orig. of Subst.: Synthesis
Type: Production

Remark: A third synthesis starts from isoprene, which is converted into 3-methyl-2-butenyl chloride by addition of hydrogen chloride. Reaction of the chloride with acetone in the presence of a catalytic amount of an organic base leads to 2-methyl-2-hepten-6-one.
Flag: non confidential, Critical study for SIDS endpoint
31-OCT-2002 (6)

Orig. of Subst.: Synthesis
Type: Production

Remark: In another process, 2-methyl-2-hepten-6-one is obtained by isomerization of 2-methyl-1-hepten-6-one. The latter can be prepared in two steps from isobutylene and formaldehyde. 3-Methyl-3-buten-1-ol is formed in the first step and is converted into 2-methyl-1-hepten-6-one by reaction with acetone.
Flag: non confidential, Critical study for SIDS endpoint
31-OCT-2002 (6)

Orig. of Subst.: Synthesis
Type: Production

Remark: 6-Methylhept-5-en-2-one is manufactured at BASF AG in a two-step synthesis using 3-methylbut-2-en-1-ol as starting substance in exclusively closed systems.
Flag: non confidential, Critical study for SIDS endpoint
27-JAN-2004 (7)

1.8 Regulatory Measures

1.8.1 Occupational Exposure Limit Values

Limit value: other: no occupational exposure limit values indicated

Flag: non confidential, Critical study for SIDS endpoint
27-JAN-2004

1.8.2 Acceptable Residues Levels

1.8.3 Water Pollution

Classified by: other: VwVwS (Germany), Annex 2
Labelled by: other: VwVwS (Germany), Annex 2
Class of danger: 1 (weakly water polluting)

Country: Germany
Remark: ID-Number: 1613
Flag: non confidential, Critical study for SIDS endpoint
22-APR-2003 (8)

1.8.4 Major Accident Hazards

1.8.5 Air Pollution

1.8.6 Listings e.g. Chemical Inventories

Type: EINECS
Additional Info: EINECS No. 203-816-7

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)

Type: ENCS
Additional Info: ENCS No. 2-2480

Remark: ENCS CLASSIFICATION:
Low Molecular Chain-like Organic Compounds.
Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)

Type: ECL
Additional Info: ECL Serial No. KE-24196

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)

Type: other: SWISS
Additional Info: SWISS No. G-8745

Remark: SWISS CLASSIFICATION:
Giftliste 1 (List of Toxic Substances 1), 31 May 1999.
Toxic Category 3: Acute oral lethal dose of 50 - 500 mg/kg.
Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)
Type: TSCA

Flag: non confidential, Critical study for SIDS endpoint

22-JUL-2002 (9)

Type: DSL

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)

Type: AICS

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)

Type: PICCS

Flag: non confidential, Critical study for SIDS endpoint
22-JUL-2002 (9)

1.9.1 Degradation/Transformation Products

EINECS-Name: No hazardous decomposition/degradation products.

Flag: non confidential, Critical study for SIDS endpoint
16-SEP-2002 (2)

1.9.2 Components

1.10 Source of Exposure

Remark: The risk of exposure to 6-methylhept-5-en-2-one may exist after spillages and during accidental exposure. Likewise dermal contact may result only from accidental exposure since the majority of the material (95%) is used as an intermediate in closed systems, and only small quantities (ca. 5%) are filled and distributed to industrial clients. Consumer exposure is low since only small amounts of 6-methylhept-5-en-2-one are contained in cosmetics at usual concentrations of up to 0.01% and in food in maximum amounts ranging from 0.5 - 10 ppm.

Flag: non confidential, Critical study for SIDS endpoint
29-JAN-2004 (4)

1.11 Additional Remarks

Memo: German "Flammable Liquids" classification (VbF): AIII

Flag: non confidential, Critical study for SIDS endpoint
16-SEP-2002 (2)

1.12 Last Literature Search

1.13 Reviews

2.1 Melting Point

Value: = -67.1 - -67.3 degree C

Remark: No further information is available.
Reliability: (2) valid with restrictions
scientifically accepted compilation of physical and chemical data
Flag: Critical study for SIDS endpoint
03-JUL-2003 (10)

Value: = -67 degree C

Reliability: (4) not assignable
Manufacturer / producer data without proof
03-JUL-2003 (2)

2.2 Boiling Point

Value: = 172 - 174 degree C at 1014 hPa

Remark: No further information is available.
Reliability: (2) valid with restrictions
scientifically accepted compilation of physical and chemical data
Flag: Critical study for SIDS endpoint
03-JUL-2003 (10)

Value: = 172 degree C at 1013 hPa
Decomposition: yes

Remark: Thermal decomposition: > 170 °C
Reliability: (4) not assignable
Manufacturer / producer data without proof
03-JUL-2003 (2)

2.3 Density

Type: density
Value: = .8508 g/cm³ at 20 degree C

Method: other: measured with double-capillary pycnometers
GLP: no

Remark: reason for flagging: experimental derived data
Test substance: 6-Methyl-5-hepten-2-one, purity 99.0 mol %
Reliability: (2) valid with restrictions
Scientifically acceptable study, meets basic scientific principles, but without detailed documentaion
Flag: Critical study for SIDS endpoint
26-JAN-2004 (11)

Type: density
Value: = .851 g/cm³ at 25 degree C

Reliability: (4) not assignable
Manufacturer / producer data without proof
01-JUL-2003 (2)

2.3.1 Granulometry

2.4 Vapour Pressure

Value: = .99 hPa at 18.2 degree C

Method: other (measured): dynamic with nitrogen
Year: 1999
GLP: no

Remark: reason for flagging: method and results are comprehensible
Result:

temperature (°C)	vapour pressure (hPa)
18.18	0.99
27.99	1.99
34.03	3.01
41.98	5.00
47.51	7.01
53.51	9.97
66.40	20.08
74.40	29.99
85.34	50.08
93.09	70.03
101.57	99.67
119.92	200.17
131.64	299.82
147.84	499.88
159.39	700.14
172.75	1007.05

Test substance: 6-Methyl-5-hepten-2-one, purity 99.5 % (impurity: 0.5 % Methylacetoacetate)
Reliability: (2) valid with restrictions
scientifically acceptable method
Flag: Critical study for SIDS endpoint
01-JUL-2003 (12)

Value: = 1 hPa at 20 degree C

Result: 8 hPa at 50°C
Reliability: (4) not assignable
Manufacturer / producer data without proof
23-JAN-2001 (2)

Value: = 3.3 hPa at 34.7 degree C

Method: other (measured): dynamic
Year: 1972
GLP: no

Result: Temperature (°C) versus vapour pressure (hPa):

°C	hPa
34.7	3.3
40.8	4.7
45.1	6.0
48.0	7.3
50.7	8.5
58.4	12.5
64.4	18.5
74.7	30.3
81.4	41.5
91.4	61.9
100.0	93.5
109.1	133.7
119.7	199.7
131.1	300.0
141.2	413.8
151.0	552.3

Test substance: 2-methylhepten-2-one-6; indication of purity is missing
Reliability: (2) valid with restrictions
comprehensible and acceptable
13-AUG-2002 (13)

Value: = 14.1 hPa at 55.2 degree C

Method: other (measured): static
Year: 1988
GLP: no

Result:

temperature (°C)	vapour pressure (hPa)
55.21	14.1
55.24	14.5
69.5	29.3
75.84	37.6
81.56	48.1
83.23	51.9
107.77	141.2
109.99	154.5
111.1	164.4
127.29	284.1
127.86	290.1
143.22	473.3
144.21	486.0
149.38	564.7
152.98	627.6
164.17	853.5
166.80	915.4
171.16	1024.7
174.96	1127.9
178.37	1229.4

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity 99.0 mol %
Reliability: (2) valid with restrictions
Scientifically acceptable study, meets basic scientific principles, but without detailed documentaion
01-JUL-2003 (11)

Value: = 59 hPa at 80 degree C

Method: other (measured): method unknown
Year: 1977
GLP: no

Result: Temperature in °C versus vapour pressure in hPa:

°C	hPa
80	59
110	183
170	1120
200	2260

The substance is decomposing at 230°C.
Reliability: (2) valid with restrictions
comprehensible and acceptable
26-JAN-2004 (14)

Method: other (measured): dynamic with argon atmosphaere
Year: 1989
GLP: no

Result: Temperature (°C) versus vapour pressure (hPa), measured values:

°C	hPa
34.55	3.00
42.18	5.00
47.17	7.00
55.57	10.00
66.65	20.00
74.78	30.00
85.67	50.00
93.33	70.00
101.94	100.00
120.20	200.00
131.93	300.00
148.00	500.00
159.30	700.00
171.81	1000.00

The calculated vapour pressure is 1013.25 at 172.63°C using the derived equation:
ln(p/bar)=9.9365-3704.88/(200.72+t/Celsius).
Test substance: 2-methylhepten-2-one-6; indication of purity is missing
Reliability: (2) valid with restrictions
comprehensible and acceptable
08-AUG-2002 (15)

2.5 Partition Coefficient

Partition Coeff.: octanol-water
log Pow: = 2.07 at 25 degree C

Method: other (measured): Determination of the logPow using gas chromatographic methods
Year: 1989
GLP: no

Test condition: A defined quantity of test substance was weighted into 25 mL 1-octanole and 25 mL water were added until the equilibrium was reached. The water phase was separated after the phase

separation, then centrifuged for 15 minutes and pipetted. The phase was then extracted using chloroform. The amount of the test substance was then measured in the chloroform phase and detected against an external standard using gas chromatographic methods.
Test was made 3 times.
The amount of test substance in the 1-octanole phase was calculated on the basis of the mass balance.

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one; purity: >99.5 %
Reliability: (2) valid with restrictions
comprehensible and acceptable

09-JUL-2003 (16)

Partition Coeff.: octanol-water
log Pow: = 2.1 at 25 degree C

Method: other (calculated): via SRC KOWWIN 1.66
Year: 2002

Reliability: (2) valid with restrictions
scientifically acceptable method

09-JUL-2003 (17)

Partition Coeff.: octanol-water
log Pow: = 2.4 at 25 degree C

Method: other (measured): according to EEC Directive 79/831, Annex V, Part A: Methods for Determination of Physical-Chemical-Properties.
Year: 1989
GLP: no

Test condition: The determination of the log Pow was performed using HPLC methods at 25°C. For the determination 6 reference compounds were used: benzyl alcohol, acetophenone, ethyl benzoate, benzophenone, phenyl benzoate and diphenylether. The corresponding k' values were calculated using the measured retention times of the reference compounds and their log Pow values. The resulting calibration curve was used to calculate the log Pow value of the test substance.

Substance	logPow	logk'(mean of three measurements)
benzyl alcohol	1.1	0.164
acetophenone	1.7	0.412
ethyl benzoate	2.6	0.892
benzophenone	3.2	1.001
phenyl benzoate	3.6	1.241
diphenylether	4.2	1.449

The resulting log Pow for 6-methylhept-5-en-2-one was 2.4 at 25°C.

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one; purity: 95-98 %
Reliability: (1) valid without restriction
study was performed according to a guideline
Flag: Critical study for SIDS endpoint

09-JUL-2003 (18)

2.6.1 Solubility in different media

Solubility in: Water
Value: = 3.02 g/l at 25 degree C
pH value: = 6.6
Conc.: 25 degree C

Year: 1989
GLP: no

Test condition: A mixture consisting of demineralised water and test substance in steady state were separated. Afterwards the two phases were centrifuged for collecting the water phase. The water phase was extracted using trichloromethane. The amount of test substance was then determined via GC using an external standard.
Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one; purity: 97.2% (using GC)
Reliability: (2) valid with restrictions
scientifically acceptable method, although the values of the single measurements are missing, but method and results are comprehensible
Flag: Critical study for SIDS endpoint
08-AUG-2002 (19)

2.6.2 Surface Tension

Test type: other: capillary method
Value: = 28.47 mN/m at 20 degree C

Method: other: measured
Year: 1988
GLP: no

Result: The result refers to a neat liquid.
Test substance: 6-Methyl-5-hepten-2-one, purity 99.0 mol %
Reliability: (2) valid with restrictions
Scientifically acceptable study, meets basic scientific principles, but without detailed documentaion
26-JAN-2004 (11)

2.7 Flash Point

Value: = 56 degree C
Type: closed cup

Method: other: measured (Abel-Pensky)
GLP: no

Reliability: (2) valid with restrictions
scientifically acceptable and comprehensible
26-JAN-2004 (20)

2.8 Auto Flammability

Value: = 250 degree C

Method: other: DIN 51 794
GLP: no

Remark: Ignition temperature
Reliability: (2) valid with restrictions
National standard specification, scientifically acceptable and comprehensible
26-JAN-2004 (20)

2.9 Flammability

2.10 Explosive Properties

Result: not explosive

Remark: because of chemical structure
Reliability: (2) valid with restrictions
Expert judgement
24-JAN-2001 (21)

2.11 Oxidizing Properties

Result: no oxidizing properties

Remark: because of chemical structure
Reliability: (2) valid with restrictions
Expert judgement
24-JAN-2001 (21)

2.12 Dissociation Constant

2.13 Viscosity

2.14 Additional Remarks

Remark: The test is assumed to be performed under non-GLP conditions.
Result: Viscosity: 0.98 mPa*s at 20 °C
Reliability: (4) not assignable
Manufacturer / producer data without proof
23-JAN-2001 (2)

Remark: The test is assumed to be performed in 1985 and under non-GLP conditions.
Result: Electrical conductivity [kappa]: 0.008 µS/cm at 25°C
Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one; purity: > 99 %
Reliability: (2) valid with restrictions
scientifically acceptable and comprehensible
02-JUL-2003 (22)

Remark: Explosion limits

The test is assumed to be performed under non-GLP conditions.
Result: Explosion limits:
lower limit: 1.1 vol.% at 52.0°C and 11.73 hPa
upper limit: 6.6 vol.% at 89.0°C and 65.06 hPa
Reliability: (2) valid with restrictions
scientifically acceptable and comprehensible
02-JUL-2003 (20)

Remark: Explosion limits

The test is assumed to be performed in 1976 and under non-GLP conditions.
Result: Explosion limits:
lower limit: 1.1 vol.% at 51.5°C and 11 hPa
upper limit: 7.3 vol.% at 91.5°C and 71.5 hPa
Reliability: (2) valid with restrictions
scientifically acceptable and comprehensible
02-JUL-2003 (23)

Remark: Specific heat capacity

The test is assumed to be performed in 1995 and under non-GLP conditions.
Result: Specific heat capacity [cp] at 25°C = 2.09 J/g*K

Specific heat capacity [measured; cp]:

Temperature (°C)	cp in J/(g*K)
0	2.04
25	2.09
50	2.14
75	2.19
100	2.24
125	2.30
150	2.35

Reliability: (2) valid with restrictions
scientifically acceptable and comprehensible
09-JUL-2003 (22)

Remark: Specific thermal conductivity

The test is assumed to be performed in 1985 and under non-GLP conditions.
Result:

Temp. (°C)	Lambda (W/m*K)
0	0.2097
22.7	0.2019
56.8	0.1927
94.5	0.1839
122.5	0.1744
149.8	0.1672

Reliability: (2) valid with restrictions
scientifically acceptable and comprehensible
02-JUL-2003 (22)

3.1.1 Photodegradation

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: NO3
Rate constant: ca. .0000000000075 cm^3/(molecule * sec)

Method: other (measured)
Year: 1992
GLP: no data

Test condition: The experiments were carried out at 296+/-2 Kelvin (22.85+/-2°C) and 740 Torr (98.66 hPa) total pressure of purified air at approx. 5 % relative humidity in a approx. 6700 L Teflon chamber equipped with two parallel banks of black lamps and with a Teflon-coated fan to ensure rapid mixing of reactants during their introduction. The reference compound was trans-2-butene.
Test substance: 6-methyl-5-hepten-2-one, purity: >= 98%
Reliability: (2) valid with restrictions
study meets basic scientific principles
26-JAN-2004 (24)

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: O3
Rate constant: ca. .00000000000000039 cm^3/(molecule * sec)
Deg. products: yes

Method: other (measured)
GLP: no data

Remark: Acetone, CH3C(O)CH2CH2CHO, Cyclohexanone and cyclohexanol were observed in the O3 reaction.
Test condition: The experiments were carried out at 296+/-2 Kelvin (22.85+/-2°C) and 740 Torr (98.66 hPa) total pressure of purified air at approx. 5 % relative humidity in a approx. 6700 L Teflon chamber equipped with two parallel banks of black lamps and with a Teflon-coated fan to ensure rapid mixing of reactants during their introduction.
The reference compounds for the OH radical and the O3 reaction were trans-2-butene and 2-methyl-2-butene, respectively.
Test substance: 6-methyl-5-hepten-2-one, purity: >= 98%
Reliability: (2) valid with restrictions
study meets basic scientific principles
26-JAN-2004 (24)

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: O3
Rate constant: = .000000000000000394 cm^3/(molecule * sec)
Deg. products: yes

Method: other (measured)
Year: 1996

Remark: The degradation products found were acetone, methyl glyoxal, formaldehyde and cyclohexanone.
Test condition: The experiment was carried out at 1013.25 hPa and 24.85°C and

with a relative humidity of 55+/-10%.
Test substance: 6-methyl-5-hepten-2-one, purity: 99 %
Reliability: (2) valid with restrictions
study meets basic scientific principles
03-JUL-2003 (25)

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: O3
Conc. of sens.: 700000000000 molecule/cm³
Rate constant: = .00000000000000043 cm³/(molecule * sec)
Degradation: = 50 % after 38.4 minute(s)

Method: other (calculated): using SRC AOP v1.90
Year: 2002

Test condition: Calculated t1/2 is valid for 12 hours-day as well as for a 24 hours-day.
Reliability: (2) valid with restrictions
scientifically acceptable method
Flag: Critical study for SIDS endpoint
26-JAN-2004 (26)

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: OH
Conc. of sens.: 1500000
Rate constant: = .00000000009177 cm³/(molecule * sec)
Degradation: = 50 % after 1.4 hour(s)

Method: other (calculated): using AOP v1.90
Year: 2002

Test condition: Calculated t1/2 based on a 12 hours day.
Reliability: (2) valid with restrictions
scientifically acceptable method
09-JUL-2003 (27)

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: OH
Conc. of sens.: 500000 molecule/cm³
Rate constant: = .0000000000917782 cm³/(molecule * sec)
Degradation: = 50 % after 4.2 hour(s)

Method: other (calculated): using SRC AOP v1.90
Year: 2002

Test condition: Calculated t1/2 based on a 24 hours-day.
Reliability: (2) valid with restrictions
scientifically acceptable method
Flag: Critical study for SIDS endpoint
09-JUL-2003 (26)

Type: air
INDIRECT PHOTOLYSIS
Sensitizer: OH
Rate constant: ca. .000000000157 cm³/(molecule * sec)
Deg. products: yes

Method: other (measured)
GLP: no data

Remark: The light source to generate OH radicals was at a wavelength of 300 nm. The test duration is not stated. the OH radical concentration is not stated.

The main degradation products of the OH radical were acetone and CH3C(O)CH2CH2CHO.

Test condition: The experiments were carried out at 296+/-2 Kelvin (22.85+/-2°C) and 740 Torr (986.6 hPa) total pressure of purified air at approx. 5 % relative humidity in a approx. 6700 L Teflon chamber equipped with two parallel banks of black lamps and with a Teflon-coated fan to ensure rapid mixing of reactants during their introduction.
The reference compounds for the OH radical and the O3 reaction were trans-2-butene and 2-methyl-2-butene, respectively.

Test substance: 6-methyl-5-hepten-2-one, purity: >= 98%
Reliability: (2) valid with restrictions
study meets basic scientific principles
16-APR-2004 (24)

3.1.2 Stability in Water

Type: abiotic

Method: other: Expert Judgement

Remark: Hydrolysis can be excluded due to the chemical structure of the compound as hydrolysis itself is mostly described for esters, carbamates, epoxides, halomethanes and specific alkyl halides

Reliability: (2) valid with restrictions
scientifically acceptable procedure
05-MAR-2004 (28)

3.1.3 Stability in Soil

3.2.1 Monitoring Data (Environment)

Type of measurement: concentration at contaminated site
Medium: other: drinking water and waste water (Cordoba, Spain)
Method: via closed-loop stripping in combination with gas chromatography and detection by MS and FID

Result: For drinking water analysis five samples were taken:
A: St. Rafael reservoir; B: water treated with chlorine and ozone; C: decanted water; D: filtered water; E: drinking water. These correspond to the different physical and chemical processes that the water at the "Villa Azul" undergoes in the Cordoba water treatment plant.
After analyses using CLSA coupled with GC-MS and GC-FID methods (including internal standards) 6-methyl-5-hepten-2-one was found as follows: site A: 97.7 ng/l; site B: 5.4 ng/l; site C: 4.8 ng/L; site D: 4.2 ng/L; site E: 5.0 ng/L. The results showed that during the process of chloronation and ozonation new products were formed and that physical processes like coagulation, flocculation and filtration have no

influence in the way of water composition.
For waste water analysis two samples were taken from the "La Golondrina" purifucation plant: F: the plant worked poorly because of a failure; G: the plant worked properly (a month later). After analyses using CLSA coupled with GC-MS and GC-FID methods (including internal standards) 6-methyl-5-hepten-2-one was found as follows: site F: 26.7 ng/l; site G: 202.9 ng/l.

Reliability: (2) valid with restrictions
scientifically acceptable, well documented and comprehensible
09-JUL-2003 (29)

Type of measurement: concentration at contaminated site
Medium: drinking water
Method: Identification of disinfection byproducts (DBP) using GC/EI-MS

Result: Ozonated water was collected from three sources (two ozonation plants and one laboratory scale ozonation) and concentrated using XAD resins.
6-methyl-5-hepten-2-one was identified as an ozone disinfection byproduct.
No concentration is given.

Reliability: (2) valid with restrictions
scientifically acceptable, well documented and comprehensible.
16-JUL-2003 (30)

Type of measurement: concentration at contaminated site
Medium: other: surface drinking water (Italy)
Method: Disinfection by-products in river water samples after treatment with peracetic acid using GC/MS methods

Result: 6-Methyl-5-hepten-2-one was identified by GC/MS in treated river water samples at a pilot and a full scale plant after peracetic acid (3 mg/l) disinfection.
No concentration is given.

Reliability: (2) valid with restrictions
scientifically acceptable, well documented and comprehensible
16-JUL-2003 (31)

Type of measurement: other:
Medium: surface water

Result: 6-methylhept-5-en-2-one is indicated as one of the nor-carotinoids which is known to be a metabolite of cyanobacteria and chlorophyceae in lake waters.

Reliability: (2) valid with restrictions
scientifically acceptable method
09-JUL-2003 (32)

Type of measurement: other: agricultural and natural plants
Medium: air

Result: 6-methylhept-5-en-2-one was observed in emissions only from birch with an emission rate of 24.9 ng/g*h using GC-MS methods.

Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (33)

Type of measurement: other: air samples in ambient coastal air (Mace Head Ireland, September 1998)
Medium: air

Result: 6-methylhept-5-en-2-one was detected in air samples and was one of the most abundant oxygenates via GC-MS. The mixing ratios for this compound ranged from 20 - 400 pptv (average 123 pptv). The levels of 6-methylhept-5-en-2-one found in the continental air were by a factor of two significant higher then those in the oceanic air.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (34)

Type of measurement: other: at contaminated site
Medium: surface water

Result: Volatile organic compounds (VOC) were detected in eight small weakly polluted rivers and brooks in South-West Germany using purge and trap enrichment with GC-MS methods.
The nor-carotinoid 6-methylhept-5-en-2-one was found in the running waters of Schussen, Rickenbach, Rotach, Argen und Brunnisach.
6-methylhept-5-en-2-one and its hydrogenated derivative 6-methylheptan-2-one are indicators of extensive microbial activity in running waters.
No informations about the concentration and detection limit is available in this study.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (35)

Type of measurement: other: at contaminated site
Medium: drinking water

Result: 6-methylhept-5-en-2-one was identified as one organic compound in drinking water in The Netherlands (original reference: Zoeteman, B.C.J. 1980. Sensory Assessment of Water Quality. Pergamon Press, Oxford).
At 4 from 16 sites the substance was detected with 3 times 0.005 µg/l and 1 time 0.01 µg/l (detection limit 0.005 µg/l).
Reliability: (2) valid with restrictions
scientifically acceptable method
17-JUL-2003 (36)

Type of measurement: other: at forrested sites in SE of the USA
Medium: air

Result: 6-methylhept-5-en-2-one was detected due to emissions from vegetation via GC-MS. No concentration is indicated.
Reliability: (2) valid with restrictions
scientifically acceptable method
17-JUL-2003 (37)

Type of measurement: other: biogenic emissions
Medium: air
Result: 6-methylhept-5-en-2-one was detected for the first time as a biogenic emission from the birch using GC-MS methods.

Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (38)

Type of measurement: other: clove essential oil
Medium: biota

Result: 6-methylhept-5-en-2-one was one of about 40 components identified in the neutral fraction of clove essential oil using GC-MS methods. The substance was considered as a degradation product formed during clove bud drying and the concentration is not indicated.
Reliability: (2) valid with restrictions
scientifically acceptable method
17-JUL-2003 (39)

Type of measurement: other: detection of VOC's in fruits
Medium: food

Result: The volatile compounds were measured in tree-ripened apricots (Prunus armeniaca L.), plums (Prunus salicina) and in their interspecicific hybrid. The compounds were isolated by similtaneous extraction and analyzed by GC-MS.
The measured concentrations of 6-methylhept-5-en-2-one in the apricots were 10 - 105 µg/kg, whereas the substance was not found in the plums. In the progenies of the hybrid the concentrations ranged between 5 and 24 µg/kg
6-methylhept-5-en-2-one measured in seven several intercrosses.
Reliability: (2) valid with restrictions
scientifically acceptable method
09-JUL-2003 (40)

Type of measurement: other: emissions at three different sites in the USA
Medium: air

Result: 6-methylhept-5-en-2-one was one of 114 BVOCs identified in branch enclosure experiments from a total of 66 vegetation species sampled at three U.S. sites. The substance was found only in cotton gras from Willow Springs, WI using GC-MS methods.
Reliability: (2) valid with restrictions
scientifically acceptable method
17-JUL-2003 (41)

Type of measurement: other: head space
Medium: other: biodegradable and mixed household waste

Result: 6-methylhept-5-en-2-one was identified in one sample of biodegradable waste and in one sample of mixed waste in concentrations of < 0.1 mg/m³ using GC-MS methods.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (42)

Type of measurement: other: head space or liquid exudate
Medium: other: garden waste
Result: 6-methylhept-5-en-2-one was identified in garden waste exudate and garden waste head space in the laboratory using GC-MS

methods.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (43)

Type of measurement: other: in Bisbee Delicious apple
Medium: food

Remark: 6-methylhept-5-en-2-one was detected in Bisbee Delicious apples. The apples were harvested in weakly intervals and means of three replicate 1 kg samples are given. The concentrations in the first orchard ranged from 15.5 - 99.4 pL/kg*h and in the second orchard from 13.9 - 171.6 pL/kg*h using GC-MS methods.
Reliability: (2) valid with restrictions
scientifically acceptable method
09-JUL-2003 (44)

Type of measurement: other: in algal monoculturs
Medium: surface water

Result: 6-methylhept-5-en-2-one was detected and quantified via GC-MS at two sites of measurement in surface waters in Australia where Microcystis aeroginosa and Anabaena sp. occur. The concentrations was < 1200 ng/L at Carcoar site, but no linear significant correlation between the substance, or other, VOC's and any phytoplancton species was found.
Reliability: (2) valid with restrictions
scientifically acceptable method
26-JAN-2004 (45)

Type of measurement: other: in algal samples
Medium: biota

Result: 6-methylhept-5-en-2-one was identified in samples of the cyanobacterium Microcystis aeruginosa from Little Para Reservoir (South Australia) and is responsible for the fruity and ester-like odour.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (46)

Type of measurement: other: in defensive glands of nymphalid butterfly Agraulis vanillae
Medium: biota

Result: The abdominal defensive glands emit a pronounced odor when disturbed. 6-methylhept-5-en-2-one is one of several compounds found in the glandular exudate of this species. MHO was the only truly volatile compound found in both sexes of this butterfly as an alarm pheromone. No concentrations of MHO are indicated.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (47)

Type of measurement: other: in nectarines (Prunus persica nectarina)
Medium: food

Result: 6-methylhept-5-en-2-one was indentified in vacuum distilled blended fruits of nectarine using GC-MS methods. Concentration of the compound is not indicated.
Reliability: (2) valid with restrictions scientifically acceptable method
01-JUL-2003 (48)

Type of measurement: other: in paprika oleoresin (Spanish type)
Medium: food

Result: 6-methylhept-5-en-2-one was detected in red pepper with 0.9 mg/kg using SDE method at atmospheric pressure. The presence of the substance is attributed to as being a carotinoid derivative as indicated by the author.
Reliability: (2) valid with restrictions scientifically acceptable method
01-JUL-2003 (49)

Type of measurement: other: in phytoplankton
Medium: biota

Result: 6-methylhept-5-en-2-one was identified via GC-MS in cultures of S. subspicatus and A. granulata. The indicated odour threshold is 50 µg/L and gives a fruity odour.
Reliability: (2) valid with restrictions scientifically acceptable method
01-JUL-2003 (50)

Type of measurement: other: in several types of meat
Medium: food

Result: 6-methylhept-5-en-2-one was identified in beef and chicken meat (original references: Perrson T., von Sydow E. and C. Akesson. 1973. Aroma of canned beef: sensory properties. J. Food Sci. 38; Tang J., Jin Q.Z., Shen G.H., Ho C.T. and S.S. Chang. 1983. Isolation and identification of volatile compounds from fried chicken. J. Agric. Food Chem. 31).
Reliability: (2) valid with restrictions scientifically acceptable method
09-JUL-2003 (51)

Type of measurement: other: in the juice of Kogyoku apples
Medium: food

Result: 6-methylhept-5-en-2-one was identified in flavor concentrates from the juice and the peel of the Kogyoku apples using GC-MS methods.
Reliability: (2) valid with restrictions scientifically acceptable method
01-JUL-2003 (52)

Type of measurement: other: in the pulp of the palm
Medium: biota

Result: 6-methylhept-5-en-2-one is one of the two main volatiles (other than those derived from simple acids and alcohols) detected in the pulp of Dalieb (Borassus aethiopum L.) using GC-MS methods. Concentrations are not indicated in this study.
Reliability: (2) valid with restrictions

scientifically acceptable method
17-JUL-2003 (53)

Type of measurement: other: urban, suburban and forest areas
Medium: air

Result: 6-methylhept-5-en-2-one was detected and analysed in tropospheric samples in Italy (Monti, Cimini Forest, Rome, Montelibretti, Milan, Taranto, Lido di Ostia, Storkow). Concentrations ranging from 0.08 up to 5.44 ppbv were measured using GC-MS methods.
The substance was detected preferably in oil cell containing flowers, fruits and plants.
Reliability: (2) valid with restrictions
scientifically acceptable method
01-JUL-2003 (54)

Type of measurement: other
Medium: biota

Result: 6-methylhept-5-en-2-one was one compound identified in mandibular glands of the aphid hyperparasitoid wasp Alloxysta brevis to prevent honeydew-collecting ants from attacks. MHO was isolated in headspace samples of 30 freshly thawed A. brevis using GC-MS methods.
Reliability: (2) valid with restrictions
study meets principles basic research
03-JUL-2003 (55)

3.2.2 Field Studies

3.3.1 Transport between Environmental Compartments

Type: adsorption
Media: water - soil
Method: other: calculated using SRC PCKOCWIN v1.66

Result: Koc = 37.12; logKoc = 1.57
Reliability: (2) valid with restrictions
scientifically acceptable method, model accepted by US EPA
Flag: Critical study for SIDS endpoint
09-JUL-2003 (56)

Type: adsorption
Media: water - soil
Method: other: calculated according to TGD (May 2003)
Year: 2003

Result: Based on the equation (Sabljic and Güsten, 1995):
logKoc = 0.81*logKow + 0.10 for so called "predominantly hydrophobics" a logKoc of 2.044 (Koc = 111) by using the measured logKow of 2.4 (see chapter 2.5) can be calculated.
Reliability: (2) valid with restrictions
scientifically acceptable, method recommended in TGD
Flag: Critical study for SIDS endpoint
09-JUL-2003 (57)
Type: volatility
Media: water - air

Method: other: calculated using SRC HENRYWIN v3.10

Remark: reason for flagging this study: model accepted by the US EPA
Result: Henry's Law Constant: H = 21.481 Pa*m³/mole (bond method)
Reliability: (2) valid with restrictions
scientifically acceptable, method accepted by US EPA
Flag: Critical study for SIDS endpoint
09-JUL-2003 (58)

Type: volatility
Media: water - air
Method: other: calculated

Result: The calculation by using the vapour pressure (Ps = 160 Pa at 25°C; extrapolated from the measured data [see chapter 2.4]) and the measured water solubility (Ws = 3.02 g/L; Cs = 23.93 mol/m³ at 25°C) (as cited in Thomas, 1982) results in a Henry's Law Constant of 6.68 Pa*m³/mol at 25°C.
This indicates a moderate volatility of 6-methylhept-5-en-2-one.
Reliability: (2) valid with restrictions
scientifically acceptable calculation
Flag: Critical study for SIDS endpoint
09-JUL-2003 (59)

3.3.2 Distribution

Media: air - biota - sediment(s) - soil - water
Method: other (calculation): Mackay Lavel I V2.11
Year: 1999

Remark: The calculation is based on the following physical and chemical properties:
molecular mass (g/mole) = 126.2
log Kow = 2.4
water solubility (g/m³) = 3020
vapour pressure (Pa) = 160
melting point (°C) = -67.0.
The Henry's Law Constant calculated by the program itself is 6.69 Pa*m³/mole.

	Volume (m³)	Density (kg/m³)	org. C (g/g)	fish lipid (g/g)
Air	6.0E+09	1.185		
Water	7.0E+06	1000		
Soil	45000	1500	0.02	
Sediment	21000	1300	0.05	
susp. Sed.	35	1500	0.167	
Fish	7	1000		0.05
Aerosol	0.012	1500		

Result: The substance will be mainly distributed to the air and to the water:
air: 69 %
water: 30 %
soil: 0.6 %
sediment: 0.6 %
susp.sed.: 0.004 %
fish: 6.42E-06 %
aerosol: 5.17E-05 %

Reliability: (2) valid with restrictions
scientifically acceptable method
Flag: Critical study for SIDS endpoint
26-JAN-2004 (60)

3.4 Mode of Degradation in Actual Use

3.5 Biodegradation

Type: aerobic
Inoculum: activated sludge, domestic
Concentration: 85 mg/l related to Test substance
Degradation: ca. 91 % after 28 day(s)
Result: readily biodegradable
Control Subst.: Aniline

Method: OECD Guide-line 301 F "Ready Biodegradability: Manometric Respirometry Test"
Year: 1995
GLP: yes
Test substance: other TS: >98 % 2-methylhepten-2-one-6 with < 1% 2-methyl-2-hepten-6-ol

Method: Test mixture containing 85 mg/L test substance, anorganic medium and 10mL non-adapted activated sludge from a laboratory wastewater treatment plant treating municipal wastewater were incubated for 28 days in a respirometer. The BOD for each treatment including the controls were measured daily and compared to the ThOD. For statistical reasons seven replicates were prepared.
As controls were used:
- Inoculum blanc: control only with the inoculum and without test substance using 2 flasks
- Positive control: treatment with 85 mg/L aniline using 1 flask
- abiotic control: control with test substance and mercury chloride (250µL) but without inoculum using 1 flask to measure the abiotic elimination.

Remark: reason for flagging this study: validity criteria for a guideline study fulfilled

Result: The biodegradation of the control substance aniline was 85 % within 28 days. The degradation in the treatment for the abiotic elimination was 2 % in total.
The degradation of the test substance was as follow (CS = control substance aniline; TSx = test substance (7 replicates):

time [day]	% of ThOD							
	CS	TS1	TS2	TS3	TS4	TS5	TS6	TS7
0	0	0	0	0	0	0	0	0
3	-2	-1	0	0	2	0	0	0
4	-3	1	5	6	10	3	3	3
5	-2	11	26	27	29	21	16	25
14	80	68	78	78	77	68	70	79
25	85	89	93	98	90	80	92	95
28	85	91	95	99	90	81	93	96

The sigmoidal shape of the biodegradation curve is a strong indication for a ready biodegradation. Elimination of the test substance via adsorption processes on the sludge could not be observed. The adaptation phase was approximately 4 days. Within the 10-day window the substance was biodegraded for >60 % which indicates ready biodegradation.

Reliability: (1) valid without restriction
Guideline study

Flag: Critical study for SIDS endpoint

29-JUL-2003 (61)

3.6 BOD5, COD or BOD5/COD Ratio

3.7 Bioaccumulation

Species: other
BCF: = 7.7

Method: other: calculated via SRC BCFWIN v2.14
Year: 2002

Remark: Estimation based on the calculated logKow = 2.06 using KOWWIN v1.66.

Reliability: (2) valid with restrictions
scientifically acceptable method

02-JUL-2003 (62)

Species: other: fish
BCF: = 22

Method: other: calculated according to TGD
Year: 2002

Result: Using the equation logBCF(fish) = 0.85*logKow - 0.70 as developed by Veith et al. (1979) and the measured logKow with 2.4 [see chapter 2.5.] a BCF for fish with 21.88 could be calculated.

Reliability: (2) valid with restrictions
scientifically acceptable method

Flag: Critical study for SIDS endpoint

09-JUL-2003 (57)

Species: other: fish
BCF: = 14.06

Method: other: calculated with BCF v2.14 using measured logKow
Year: 2004

Remark: The measured logKow of 2.4 was used as input parameter for the calculation program BCF v2.14.

Reliability: (2) valid with restrictions
scientifically acceptable method

26-JAN-2004 (63)

3.8 Additional Remarks

AQUATIC ORGANISMS

4.1 Acute/Prolonged Toxicity to Fish

Type: flow through
Species: Pimephales promelas (Fish, fresh water)
Exposure period: 96 hour(s)
Unit: mg/l **Analytical monitoring:** yes
LC50: 85.7

Method: other: according to US EPA Committee on Methods for Toxicity Tests with Aquatic Organisms (1975)
Year: 1984
GLP: no data

Result: No fish died up to a concentration of 142 mg/L (nominal) over the 96 h test period. In the treatment with 236 mg/l (nominal) 6-methylhept-5-en-2-one after 1 h of exposure 7 and 5 fish died in the two replicates, respectively. Until 96 h in one replicate 25 fish and in the other 22 fish died. In the treatment with 394 mg/l (nominal) 6-methylhept-5-en-2-one all fish died after 1 h of exposure.
Further, prior to death affected fish showed sublethal effects like hypoactivity, lost of equilibrium and they stopped schooling.
The LC50 (96h) based on the analytical concentrations by using gas-liquid-chromatography was calculated with 85.7 mg/l (limit of confidence: 83.3-88.2 mg/l) using the Spearman-Kärber method.

Concentrations (mg/l)

Nominal	Analytical (Corrected avarage)
0 A	<0.9
0 B	<0.9
51 A	20.6
51 B	20.9
85 A	42.3
85 B	41.9
142 A	65.2
142 B	64.1
236 A	111.0
236 B	107.0
394 A	144.0
394 B	160.0

Test condition: The test was performed according to US EPA Committee on Methods for Toxicity Tests with Aquatic Organisms (1975).
The fathead minnow Pimephales promelas was used.
Test temperature: 24.8+/-0.47 °C
Dissolved oxygen: 7.2+/-0.46 mg/l
pH-value: 7.62+/-0.20
Hardness 44.5 mg/l CaCO3
Alkalinity: 43.2 mg/l CaCO3
Tank volume: 6.3 l
Additions: 5.70 l exchange per day
Mean length of fish: 17.6+/-3.583 mm
Mean weight of fish: 0.096+/-0.0680 g
Loading: 0.381 g fish/l test water
Test design: Five concentrations and an untreated control with

two replicates each were tested. 25 fish were placed at each replicates at concentrations of 51, 85, 142, 236 and 394 mg/l over a 96 h period without feeding. Lake Superior water was used in the tests.

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity: 98 %

Reliability: (1) valid without restriction
According to guideline study

Flag: Critical study for SIDS endpoint

09-JUL-2003 (64) (65) (66) (67) (68)

Type: static
Species: Leuciscus idus (Fish, fresh water)
Exposure period: 96 hour(s)
Unit: mg/l **Analytical monitoring:** no
NOEC: 46.4
LC50 (geometric mean) :
= 68.1
LC50 (effective) :
ca. 50

Method: other: German Industrial Standard DIN 38 412, Part 15
Year: 1982
GLP: no
Test substance: as prescribed by 1.1 - 1.4

Result: The fish were found to respond to a positive control (chloroacetamide) with an LC50 of 32 mg/l after 48 h.
No fish died in the treatment with 46.4 mg/l over the 96 h test period. After 4 h all fish died in the treatments with 100, 215, 464 and 1000 mg/l 6-methylhept-5-en-2-one.
Thus, using the geometric mean of the LC0 of 46 mg/l and the LC100 of 100 mg/l a LC50 of 68.1 mg/l (geometric mean) could be calculated. Further, no symptoms like gasping or tumbling etc. could be observed.

All values are referred to nominal concentrations. No analytics of the test compound were performed. Due to a moderate volatility in an experiment using 100 mg and 200 mg of 6-methylhept-5-en-2-one, but without test organisms, the evaporation over 96 hours was observed. After 96 h approximately 23 % and 26 % of the test substance were evaporated (geometric mean of all measured TOC values between 0 h and 96 h), respectively. Therefore, the LC50(96h) can be corrected to be approximately 50 mg/l and 53 mg/L, respectively.

Test condition: Closely followed the German Industrial Standard Guideline Number DIN 38 412, Part 15 (June 1982) using a static exposure procedure. One criteria of this guideline is that the corpulence factor K should be between 0.8 and 1.1 g/cm³ (equation for calculation: $K = 100*W/L^3$; W = weight in g; L = length in cm).
The Golden Orfe (L. idus), golden variety, was used.
Aeraton: slight
Duration of housing and adaptation: about 2 weeks
Duration of adaptation: 3 days
Withdrawal of food before exposure: 1 day before and during exposure
Body length: 6.0 cm (range: 5.5-7.1)
Body weight: 1.8 g (range: 1.2-2.8)

Corpulence factor: 0.8 g/cm³
Loading: 1.8 g fish/l test water
Test design: 10 fish were used per concentration and an untreated control, at concentrations of 46.4, 100, 215, 464 and 1000 mg/l.

measured pH values:

concentration (nominal, mg/l)	pH 1h	24 h	48 h	72 h	96 h
46.4	7.6	7.6	7.6	7.6	7.6
100	7.6				
215	7.6				
464	7.7				
1000	7.7				
control	7.8	7.6	7.9	7.7	7.5

measured oxygen concentrations

concentration (nominal, mg/l):	O2 1h	24 h	48 h	72 h	96 h
46.4	8.2	7.9	8.0	8.2	8.2
100	8.3				
215	8.3				
464	8.6				
1000	8.7				
control	8.1	7.9	8.1	8.3	7.5

The concentrations used were chosen based on a range finding study.
The test substance was added to the test water without any prior treatment. Subsequently, the fish were added to the water.
Test vessel: All-glass aquarium non-sealed (30 x 22 x 24 cm)
Dilution water chemistry: reconstituted freshwater was prepared from demineralized tap water that was resalted by the addition of 294.0 mg/l CaCl2*2H2O, 123.3 mg/l MgSO4*7H2O, 63.0 mg/l NaHCO3 and 5.5 mg/l KCl. The test water had a total hardness of 2.5 mmol/l, an acid capacity of 5.5 mmol/l and a pH of 8.0. The water temperature was 21+/-1°C. As control test water without test substance was used.

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity: 98.2 %
Reliability: (2) valid with restrictions
Guideline study, most sensitive study available on this endpoint, but no GLP and no analytics were performed
Flag: Critical study for SIDS endpoint
26-JAN-2004 (69) (70) (71)

Type: other
Species: other: three different species of fish (Petromyzon marinus, Salmo gairdnerii, Lepomis macrochirus)
Exposure period: 24 hour(s)
Unit: **Analytical monitoring:** no

Method: other
GLP: no

Result: At an initial concentration of 5 ppm (mg/l) no species were affected within 24 h of exposure. Observations about sublethal

effects caused by the test substance are not indicated.

Test condition: Tests were conducted for a 24 h period at a water temperature of 55 deg.F +/- 1.0 deg.F (12.78 deg. C).
Size of fish: 4 inches or slightly less in length
Loading: 6 animals together (two specimens of each, or lamprey larvae and one of the other fishes) in 10-liter glass battery containing 5l of water
Aeration: at near oxygen saturation using stone air-breakers
Test water: from Hammond Bay of Lake Huron (250 feet offshore at a depth of about 9 feet), over the year variations in pH from 7.5 to 8.2, dissolved oxygen from 8.6 to 13.7 ppm (mg/l) and free carbon dioxide from 5 to 9 ppm (mg/l) could be measured.
Test concentrations: 5 ppm (mg/l; initial), chemicals killed lamprey larvae in eight hours or less were further tested in 1 and 0.1 ppm (mg/l).

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one;
The authors indicated that the degree of purity was not available, thus, all samples were treated as pure preparations.

Reliability: (3) invalid
no analytics were performed; no purity of the test substance available

02-JUL-2003 (72)

4.2 Acute Toxicity to Aquatic Invertebrates

Type: static
Species: Daphnia magna (Crustacea)
Exposure period: 48 hour(s)
Unit: mg/l **Analytical monitoring:** no
EC0: = 58
EC50: = 129
EC100: = 320
EC50 (effective) :
ca. 83

Method: other: Acute Toxicity test for Daphnia according to DIN 38412/11
Year: 1989
GLP: no

Remark: Reason for flagging this study: only study available on this endpoint.

Result: Observation of immobile daphnids (mean %) after n hours:

nominal concentration (mg/l)	immobility			
	3h	6h	24h	48h
580	100	100	100	100
320	100	100	100	100
180	20	25	45	85
100	0	0	15	20
58	0	0	0	5
control	0	0	0	0

Using the Spearman-Kärber method a EC50 (24h) of 168 mg/l and

a EC50 (48h) of 129 mg/l was calculated.

All values are referred to nominal concentrations. No analytics of the test compound were performed.
Due to a moderate volatility in an experiment using 100 mg and 200 mg of 6-methylhept-5-en-2-one, but without test organisms, the evaporation over 48 hours was observed. After 48 h approximately 33 % and 35 % of the test substance were evaporated (geometric mean of of all measured TOC values between 0 h and 96 h), respectively. Therefore, the EC50(48h) can be corrected to be approximately 83 mg/l and 89 mg/L, respectively.

Test condition: Closely followed the German Industrial Standard Guideline Number DIN 38 412, Part 11 (June 1989) using a static exposure procedure.
The waterflea Daphnia magna STRAUS was used. Before testing the animals were kept separately in 100 ml glass beakers containing 70 ml water at 21°C. The water was exchanged daily and a conductivity of 658 µS/cm, a Ca: Mg ratio of 5:1 and a Na:K ratio of 20:1 were measured. The animals showed a reproduction rate of three animals per day. The animals were fed with the algae Scenedesmus subspicatus once a day.
Test design: After preparing a stock solution (1000 mg/l) 4x5 daphnids were used per concentration and an untreated control, at nominal concentrations of 58, 100, 180, 320 and 580 mg/l according to a total test volume of 20 ml each. The test conditions corresponded to that in the stock culture. The test substance was added to the test water without any prior treatment. Subsequently, the daphnids were added to the water. As control test water without test substance was used. The several treatments were prepared according to the standard guideline and therefore, no measurements of oxygen, pH and temperature at the beginning of the exposure (0h) were performed. After 48h of exposure these parameters were determined:

measured pH values, oxygen concentrations (mg/l) and temperature (°C) at the end of exposure (48h):

	pH	O2	temperature
580	7.62	8.3	21.0
320	7.63	8.6	21.0
180	7.56	8.2	21.0
100	7.56	8.3	21.0
58	7.58	8.3	21.0
control	7.59	8.4	21.0

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity 98.1%, impurity with 2-methyl-2-hepten-6-ol (<1%)

Reliability: (1) valid without restriction
Guideline study

Flag: Critical study for SIDS endpoint

29-JUL-2003 (69) (70) (73)

4.3 Toxicity to Aquatic Plants e.g. Algae

Species: Scenedesmus subspicatus (Algae)
Endpoint: growth rate
Exposure period: 72 hour(s)
Unit: mg/l **Analytical monitoring:** no
NOEC: < 10
EC10: = 30.13
EC50: = 191.42
EC50 (effective) :
ca. 116

Method: other: algal growth inhibition test according to DIN 38412/9
Year: 1989
GLP: no

Remark: Reason for flagging this study: only study available on this endpoint.
Result: Neither growth promotion nor autofluorescence were observed.
The effect values for the endpoint growth rate are:
NOEC (72h) < 10 mg/l
EμC10 (72h) = 30.13 mg/l (95% confidence: 9.1-99.9 mg/l)
EμC50 (72h) = 191. 42 mg/l (95% confidence: 56.1-653.6 mg/l)
The effect values for the endpoint biomass are:
NOEC (72h) < 10 mg/l
EbC10 (72h) = 31.12 mg/l (95% confidence: 11.4-84.5 mg/l)
EbC50 (72h) = 208.20 mg/l (95% confidence: 71.3-607.7 mg/l)

The pH values at the beginning (t0) and at the end of exposure (96h) and the temperature were measured:

	pH		°C
	t0	t96	
control	7.73	7.99	21.3
10 mg/l	7.77	9.19	21.3
25 mg/l	7.77	9.17	21.3
50 mg/l	7.77	8.51	21.3
100 mg/l	7.76	7.88	21.3
250 mg/l	7.77	7.59	21.3
500 mg/l	7.76	7.58	21.3

In spite of the increased pH values after 96h in the treatments 10 and 25 mg/l a pH effect can be excluded, because the cell density and the growth rate, as relevant parameter, were not affected.

All values are referred to nominal concentrations. No analytics of the test compound were performed.
Due to a moderate volatility in an experiment using 100 mg and 200 mg of 6-methylhept-5-en-2-one, but without test organisms, the evaporation over 72 hours was observed. After 72 h approximately 37 % and 39 % of the test substance were evaporated (geometric mean of of all TOC values between 0 h and 96 h), respectively. Therefore, the ErC50(72h) can be corrected to be approximately 116 mg/l and 119 mg/L, respectively. For the EbC50(72h) the effective values are 126 mg/l and 130 mg/l, respectively.
Test condition: Test was performed according to the German standard DIN 38412, part 9.
Test organisms:

- Scenedesmus subspicatus CHODAT (SAG 86.81)
Test conditions:
- according to the standard working procedure of A 38412, part 9
- test temperature range: 21 - 25°C
- number of cells were measured daily using fluorescence method at 300 - 780 nm (pulsed fluorometry)
Test desgin:
- range finding (0, 5, 50, 500 mg/l)
- concentrations: 10, 25, 50, 100, 250 and 500 mg/l without additional solvent
- 4 replicates per concentration
- no analytical monitoring
- test duration was 96 h
The ECx values were calculated according to Tallarida and Jacob (1979).

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity 98.1%, impurity with 2-methyl-2-hepten-6-ol (<1%)

Reliability: (2) valid with restrictions
According to national standard test procedures, comprehensible and acceptable

Flag: Critical study for SIDS endpoint

10-JUL-2003 (69) (70) (74) (75) (76)

4.4 Toxicity to Microorganisms e.g. Bacteria

Type: aquatic
Species: Photobacterium phosphoreum (Bacteria)
Exposure period: 5 minute(s)
Unit: mg/l **Analytical monitoring:** no
EC50: = 17.5

Method: other: Microtox Toxicity Assay
GLP: no

Method: Within the Microtox system the decrease in the luminescence of the bacterium Ph. phosphoreum in response to a toxicant is measured. The endpoint is the 5-min median effect concentration (EC50), which indicates a 50 percent reduction in light output.

Reliability: (2) valid with restrictions
study meets basic scientific principles

Flag: Critical study for SIDS endpoint

03-JUL-2003 (77)

Type: aquatic
Species: Pseudomonas putida (Bacteria)
Exposure period: 30 minute(s)
Unit: mg/l **Analytical monitoring:** no
EC10: = 1800
EC50: = 3000
EC90 : = 4500

Method: other: oxygen consumption inhibition test according to Robra
Year: 1988
GLP: no

Result:

Concentration	pH	oxygen consumption 1	2	Mean	Mean %
control	7.19	2.2	2.05	2.125	100
1,250 mg/l	7.19	2.4	2.35	2.375	115.15
2,500 mg/l	7.19	1.45	1.4	1.425	69
5,000 mg/l	7.18	0	0	0	0

Test condition: The stock solution (10000 mg/l) was prepared using 100 mg/l Tween 80. The test concentrations (625; 1,250; 2,500; 5,000 and 10,000 mg/l and two replicates each) were prepared under constantly stirring. As blank a control with Tween 80 was used.
The test was performed according to the ROBRA oxygen consumption inhibition test (Robra K.H. 1976. gwf Wasser-Abwasser 117, 80-86.) at 25°C using a 24 h old bacterial suspension.

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity 98.1%, impurity with 2-methyl-2-hepten-6-ol (<1%)

Reliability: (2) valid with restrictions
According to national standard test procedures, comprehensible and acceptable

02-JUL-2003 (78)

Type: aquatic
Species: other bacteria: activated sludge from laboratory waste water treatment plants treating municipal sewage
Exposure period: 30 minute(s)
Unit: mg/l **Analytical monitoring:** no
EC50: = 800
EC20 : ca. 27

Method: OECD Guide-line 209 "Activated Sludge, Respiration Inhibition Test"
Year: 1995
GLP: yes

Method: Test mixture (total volume 250 ml) was incubated at 20+/-2°C for 30 minutes, after which the respiration rate was measured. Test mixture contained test substance (1g/l), sewage feed (as prescribed by OECD 209) and activated sludge from laboratory wastewater treatment plants treating municipal wastewater.
The following controls were included:
- Inoculum blank: control without test substance but with inoculum (3 flasks)
- Positive control: 3,5-dichlorophenol with inoculum (3 flasks, 1, 10, and 100 mg/l).

Remark: A significant inhibition of the respiration rate was observed. Disturbances in the biodegradation process of activated sludge are possible.

Result:

	Respiration rate (mgO2/l*h)	Inhibition (%)
Test substance		
20 mg/l	13	-19
100 mg/l	12	-25
250 mg/l	11	-31
500 mg/l	10	-38
1000 mg/l	7	-56

Inoculum blank		16	-
Reference substance			
	1 mg/l	15	-6
	10 mg/l	9	-44
	100 mg/l	1	-94

The EC20 for the reference 3,5-dichlorophenyl was 2.5 mg/l and the EC50 was 15 mg/l. The EC50 for the reference is valid because of ranging between 5 and 30 mg/l as validity criteria.

Test substance: CAS: 110-93-0, 6-methylhept-5-en-2-one, purity 98.1%, impurity with 2-methyl-2-hepten-6-ol (<1%)

Reliability: (1) valid without restriction
Guideline study, test was performed with sludge from municipal waste water treatment plant

Flag: Critical study for SIDS endpoint

09-JUL-2003 (79)

Type: aquatic

Species: other bacteria: different species (Cytophaga johnsonae, Chromobacterium lividum, Arthrobacter spec., Enterobacter cloacae, Pseudomonas fluorescens, Bacillus mycoides

Year: 1981

Result: 6-methylhept-5-en-2-one was found to be the strongest inhibitor of bacterial growth, pigment production and glucose utilization. MHO inhibited the colonial growth of six bacteria strains tested, but E. cloacae and P. fluorescens were less sensitive. The violacein pigment production by Chromobacter lividum was inhibited by MHO. Further, the uptake and respiration of [U-14C]glucose by river microflora was strongly inhibited at 2ppm (v/v) MHO (= 18µM).

Test condition: The effects of 6-methylhept-5-en-2-one on bacterial growth and on glucose uptake and respiration of six bacteria strains and microflora of river water were investigated a) in a test using agar plates and b) in an other test containing a mineral medium plus 10mM glucose.

agar-test:
-quadruplicated pipetting of 2µl MHO on surface-dry nutrient agar plates inoculated with 0.1 ml of 10+E09 cells/ml (log phase culture)
- incubation: 3 days at 20°C
- evaluation: free areas in bacterial lawn of more than 0.5 mm diameter were scored as growth inhibition

glucose-test
- conducted with the six species of bacteria and the microflora of river water
- test duration: 60 minutes at 20°C
- 50 ml serum (culture medium) + 0.2 ml glucose (final dilution 2.5-50 µM) + 0.02 ml substance (final dilution 2*10E-08 - 2*10E-05 (v/v)) + 9 ml bacterial suspension (late log phase) or 9 ml river water sample

Reliability: (2) valid with restrictions
scientifically acceptable method, well documented and comprehensible

09-JUL-2003 (80)

Type: field
Species: other fungi: Fusarium nivale and Septoria nodorum

Result: The anitfungal activities of 6-methylhept-5-en-2-one and other five ketones was tested using F. nivale and S. nodorum. These ketones caused inhibition but were less effective compared to other groups such as aldehydes and esters. Significant results for MHO are not indicated by the author.

Test condition: - petridishes of 90 mm diameter containing 15 ml sterile medium
- inoculation of medium with pieces of mycelium of uniform size (3 mm diameter)
- sterile aluminium cup introduced to the margin of each dish
- 1, 10, 20, 40 µl compound/dish was pipetted in corresponding cup
- sealing of dishes with parafilm
- incubation at 30°C in darkness
- examination of morphological characteristics, colour, linear growth, dry matter at the end of incubation

Reliability: (3) invalid
documentation unsufficient for assessment

09-JUL-2003 (81)

4.5 Chronic Toxicity to Aquatic Organisms

4.5.1 Chronic Toxicity to Fish

4.5.2 Chronic Toxicity to Aquatic Invertebrates

TERRESTRIAL ORGANISMS

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 Toxicity to other Non-Mamm. Terrestrial Species

Species:	other: avian (red-winged blackbird; Agelaius phoeniceus)
Endpoint:	mortality
Expos. period:	18 hour(s)
Unit:	mg/kg bw
LD50 :	> 111
Method:	other: Acute oral toxicity
Year:	1972
GLP:	no data
Remark:	Estimated LD50 are based on food consumption data.
Test condition:	-birds were preconditioned to captivity for 2 to 6 weeks and dosed by gavage with solutions or suspensions of the test chemical in propylene glycole - other oral dosing methods were occasionally used like pellets or gelatine capsules
Reliability:	(2) valid with restrictions basic data are given and are acceptable, even if no informations about the number of test organisms, the way and preparation of dosed gavages and other important test parameters for the corresponding test substances are available
Flag:	Critical study for SIDS endpoint

29-JUL-2003 (82)

4.7 Biological Effects Monitoring

4.8 Biotransformation and Kinetics

Type:	other: fungi
Result:	The four test compounds were transformed into methylheptenone by Penicillium digitatum. Geraniol was completely converted after 5 h into MHO and an equimolar amount was found. Nerol was transformed at a similar rate. Citral was completely converted within 1 h into MHO, geraniol and nerol. After 5h all compounds were completely converted and a quantitative increase in the MHO concentration could be observed. Thus, the maximal MHO transformation rate from citral was three times higher than the formation rates a substrate containing geraniol or nerol. Citral lyase, a cofactor-independently enzyme, was detected as the converts citral into methylheptenone and acetaldehyde. All substances were detected via GC-MS methods.
Test condition:	- experiments were carried out in 15 mL vials, 1mM substrate was added to a spore suspension (the culture was maintained at -80°C) in a total volume of 1 mL of 50 mM phophate buffer (pH 7.0)

- after addition the suspension was shaken for 30 s and then placed in a shaking 25°C bath
- at each sampling time one vial was taken to extract the terpenes using ethyl acetate
- analysis was performed using GC-MS methods followed by

Test substance: geraniol, nerol, citral and geranic acid
Reliability: (2) valid with restrictions
scientifically acceptable method
03-JUL-2003 (83)

Type: other: fungi

Result: Geraniol and nerol were transformed into 6-methylhept-5-en-2-one (MHO) by spores of Penicilliium italicum over a up to two months period. In a experiment with both geraniol and nerol (6 x 100 µL geraniol and 2 x 100 µL nerol to one batch within a exposure period of two months) MHO was obtained with a purity of 96 %, which means that a total yield of nearly 89 % could be observed.

Test condition:
- Conical flasks with 100 mL soil medium on the bottom
- Inoculation period until the surface was completely coverd with spores was approximately two weeks
- 100 µL test substance was sprayed over the sporulated surface
- headspace samples were taken for analysis
- analysis was performed using GC-MS methods followed by quantification using added standards

Test substance: geraniol, nerol
Reliability: (2) valid with restrictions
scientifically acceptable method
03-JUL-2003 (84)

Type: other: fungi

Result: Citral was faster converted to 6-methylhept-5-en-2-one then the alcohols by Penicilliium digitatum, but only to approximately 76 %. In contrast, the alcohols and their mixture were converted to MHO to an amount of about 83 %. Longer observations showed a better bioconversion of 80-90 % in dependence to the substrate used.

Test condition:
- six batches of sporulated surface culturs in 500 mL conical flasks containing 100 mL Malt Extract Agar (MEA)
- inoculation period until the surface was completely coverd with spores was approximately two weeks
- batches were treated with 300 µL test substance: 1x geraniol, 1x nerol, 2x mixture of geraniol and nerol (= citrol), 2x mixture of geranial and neral (= citral)
- headspace samples at defined intervals were taken for analysis
- analysis was performed using GC-MS methods

Reliability: (2) valid with restrictions
scientifically acceptable method
03-JUL-2003 (85)

4.9 Additional Remarks

Memo: Prediction of toxicity to fathead minnow using QSAR's

Remark: Prediction of fathead minnow acute toxicity of organic compounds from molecular structure. 2-Methylhepten-2-on-6 was mentioned as one of 375 compounds for which a QSAR study was performed.

Reliability: (4) not assignable
Data from Handbook or collection of data

22-MAR-2004 (86)

5.0 Toxicokinetics, Metabolism and Distribution

Remark: No specific studies are available concerning kinetic or metabolic fate of the substance.

23-MAR-2004 (87)

5.1 Acute Toxicity

5.1.1 Acute Oral Toxicity

Type: LD50
Species: rat
Strain: Sprague-Dawley
Sex: male/female
No. of Animals: 10
Vehicle: CMC
Doses: 170, 1360, 2720, 3400, 4250 and 5540 mg/kg bw
Value: ca. 3570 mg/kg bw

Method: other: similar to OECD 401
Year: 1973
GLP: no
Test substance: as prescribed by 1.1 - 1.4

Result: MORTALITY
No deaths occurred at the lowest doses of 170 mg/kg bw. All animals died at 5540 mg/kg bw and above.

Time of death and number of deaths at each dose

5540 mg/kg bw: 5/5 males and 4/5 females died within 24 hrs, 1 additional female died within 7 days
4250 mg/kg bw: 4/5 males and 2/5 females died within 24 hrs
3400 mg/kg bw: 0/5 males and 0/5 females died within 7 days
2720 mg/kg bw: 0/5 males and 1/5 females died within 24 hrs
1360 mg/kg bw: 2/5 males and 0/5 females died within 24 hrs
170 mg/kg bw: 0/0 males and 0/0 females died within 7 days

From these data a LD50 value of 3570 mg/kg bw was estimated after 7 days

CLINICAL SIGNS
5540 mg/kg bw: Abdominal and lateral position, atonia, apathy, severe dyspnoea
4250 - 1360 mg/kg bw: abdominal position, apathy, atonia, gasping, smeared fur, animals were without clinical symptoms after 5 days
170 mg/kg bw: atonia, gasping, after 1 day animals were without clinical symptoms

NECROPSY FINDINGS
In animals that died due to substance application:
5540 - 1360 mg/kg bw: acute heart dilatation, congestive hyperemia; greyish discoloration of liver and kidneys

In animals that were sacrificed after post observation period: No abnormalities detected in any group

Test condition: TEST ORGANISMS

Per dose group 5 male and 5 female Sprague-Dawley (Gassner) rats with a median weight of 172 g (females) and 194 g (males), no control animals were included

ADMINISTRATION
The substance was applied at dosages of 6.4, 5.0, 4.0, 3.2, 1.6 and 0.2 ml/kg bw by gavage as aqueous emulsions in CMC (supplemented with 2-3 drops of Cremophor EL) at concentrations of 30, 16 and 2% (v/v). These values corresponded to dosages of 5540, 4250, 3400, 2720, 1360 and 170 mg/kg bw.
Post observation period: 7 days

EXAMINATIONS
Animals were inspected for signs of pharmacologic or toxicologic effects during a 7 d post observation period. Body weight was measured before dosing. At the end of the observation period survivors were sacrificed and necropsied as were animals that died.
The approximative mean lethal dose (LD50) was estimated (calculation method not mentioned)

Test substance: 6-Methyl-5-hepten-2-one, purity 98 %
Reliability: (2) valid with restrictions
Comparable to guideline study with acceptable restrictions.
Flag: Critical study for SIDS endpoint
09-MAR-2004 (88)

Type: LD50
Species: rat
Strain: no data
Sex: no data
No. of Animals: 60
Vehicle: no data
Doses: 2000, 2500, 3200, 4000, 4100, 5000 mg/kg bw
Value: 4100 mg/kg bw

Method: other
Year: 1972
GLP: no data
Test substance: other TS

Result: 2000 mg/kg bw: 0/10 deaths

2500 mg/kg bw: 2/10 deaths. Deaths occurred on days 1 and 2. Clinical signs observed were immediate stimulation followed by ataxia.

3200 mg/kg bw: 3/10 deaths which occurred on days 1 and 2. Clinical signs observed were immediate stimulation followed by ataxia.

4000 mg/kg bw: 4/10 deaths which occured on days 1 and 2. Clinical signs observed were immediate stimulation followed by ataxia.

5000 mg/kg bw: 9/10 deaths which occurred on days 1 and 2. Clinical signs observed were immediate stimulation followed by ataxia.

LD50: 4100 mg/kg bw (95% CI = 3300 - 5040 mg/kg bw)
Test condition: 10 rats per dose were used. Animals were observed for mortality and clinical signs for a period of 7 days.
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (2) valid with restrictions
Comparable to guideline study with acceptable restrictions. Restriction: method for calculation of LD50 not mentioned.
15-APR-2003 (89) (90)

Type: LD50
Species: mouse
Strain: Swiss Webster
Sex: male
No. of Animals: 10
Vehicle: CMC
Doses: 1000 and 2000 mg/kg bw
Value: > 2000 mg/kg bw

Method: other
Year: 1987
GLP: no data
Test substance: other TS

Result: 1000 mg/kg bw: no mortality observed
2000 mg/kg bw: no mortality observed
Test condition: Animals were housed in temperature-controlled rooms with a 12-hour light/dark cycle, and allowed free access to water and food. After a 3-day acclimatization period, groups of 10 animals were administered a single dose of the test material in 1% aqueous sodium carboxymethylcellulose by oral intubation. The controls were treated with the vehicle only. Mortality was determined up to 24 hours after oral administration and the LD50 values were calculated by the Miller and Tainter method. If no mortality was recorded, the animals were observed for 14 days to detect any delayed toxicity. The body weights were recorded on days 0, 1, 7 and 14.
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (2) valid with restrictions
Comparable to guideline study with acceptable restrictions.
08-JUL-2003 (91)

Type: LD50
Species: mouse
Strain: no data
Sex: no data
Vehicle: no data
Doses: no data
Value: = 2410 mg/kg bw

Method: other
Year: 1988
GLP: no
Test substance: other TS

Test substance: methyl heptenone, no data on purity mentioned
Reliability: (4) not assignable
Secondary literature, essential details of method and results not given

08-JUL-2003 (92) (93)

Type: LD50
Species: mouse
Strain: other: CF-1
Sex: male/female
No. of Animals: 10
Vehicle: no data
Doses: no data
Value: = 3609 mg/kg bw

Method: other
Year: 1967
GLP: no
Test substance: other TS

Result: LD50 3609 +/- 337 mg/kg bw
Test condition: Male and female CF-1 mice weighing 17-25 g were orally administered the test compound. Ten mice were used per dose level. The observation period was 72 hours. The LD50 was calculated per Miller and Tainter (1944). The use of a vehicle was not mentioned. Necropsy was not mentioned.
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (4) not assignable
Secondary literature, essential details of method and results not given

13-JUN-2003 (94)

5.1.2 Acute Inhalation Toxicity

Type: other: Inhalation Risk Test
Species: rat
Strain: Sprague-Dawley
Sex: male/female
No. of Animals: 12
Vehicle: other: air
Exposure time: 8 hour(s)

Method: other: BASF-method
Year: 1973
GLP: no
Test substance: as prescribed by 1.1 - 1.4

Result: MORTALITY
No mortality was observed when 12 rats were exposed for 8 hours to an atmosphere that has been saturated at 20 degree centigrade with the volatile part of the compound.

CLINICAL SIGNS
Immediately after exposure, the animals showed impaired balance. The next day, all animals were normal again.

NECROPSY FINDINGS
No abnormalities
A LC50 of > 13.96 mg/l/4 hrs (> 13,960 mg/m3/4hrs) could be estimated using Haber's rule (LC50 > 6.98 mg/l/8 hrs, > 6,980 mg/m3/8 hrs).
Test substance: 6-Methyl-5-hepten-2-one, purity 98 %
Test condition: 12 rats were exposed for 8 hours to an atmosphere that has

been saturated at 20 degree centigrade with the volatile part of the compound.

A nominal test substance concentration in the air of 6.98 mg/l (6980 mg/m3) was calculated by using the weight loss of test substance and the amount of air used during exposure.

Post observation period: 7 days

Reliability: (2) valid with restrictions
Meets national standard methods with acceptable restrictions

Flag: Critical study for SIDS endpoint

22-MAR-2004 (88)

5.1.3 Acute Dermal Toxicity

Type: LD50
Species: rabbit
Strain: no data
Sex: no data
No. of Animals: 6
Vehicle: no data
Doses: 5000 mg/kg bw
Value: > 5000 mg/kg bw

Method: other
Year: 1972
GLP: no data
Test substance: other TS

Result: 1/6 deaths occurred on day 9. Acute dermal LD 50 > 5000 mg/kg bw. Dermal irritation observed.

Test condition: 6 rabbits per dose were used. Animals were observed for mortality and clinical signs over a period of 14 days.

Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned

Reliability: (2) valid with restrictions
Data from Handbook or collection of data

15-APR-2003 (89) (90)

5.1.4 Acute Toxicity, other Routes

Type: LD50
Species: mouse
Strain: NMRI
Sex: male/female
No. of Animals: 10
Vehicle: CMC
Doses: 170, 340, 680 and 1360 mg/kg bw (ca. 0.2, 0.4, 0.8 and 1.6 ml/kg bw)
Route of admin.: i.p.
Value: ca. 510 mg/kg bw

Method: other: BASF-method
Year: 1973
GLP: no
Test substance: as prescribed by 1.1 - 1.4

Result: MORTALITY
No deaths occurred at the lowest doses of 0.2 ml/kg bw.

Time of death and number of deaths at each dose

1.6 ml/kg bw: 5/5 males and 4/5 females died within 24 hrs
0.8 ml/kg bw: 3/5 males and 4/5 females died within 24 hrs
0.4 ml/kg bw: 0/5 males and 3/5 females died within 7 days
0.2 ml/kg bw: 0/5 males and 0/5 females died within 7 days

From these data a LD50 value of 0.8 ml/kg bw (= 510 mg/kg bw) after 7 days was estimated.

CLINICAL SIGNS
Abdominal position, staggering, apathy, atonia, dyspnoea

NECROPSY FINDINGS
No intraabdominal adhesions, no abnormalities

Test condition: TEST ORGANISMS
Per dose group 5 male and 5 female NMRI Ivanovas mice with a median weight of 23.8 g (males) and 23.1 g (females), no control animals were included

ADMINISTRATION
The substance was applied at dosages of 1.6, 0.8, 0.4 and 0.2 ml/kg bw by gavage as aqueous emulsions in CMC (supplemented with 2-3 drops Cremophor EL) at concentrations of 16, 8, 4 and 2% (v/v). These values correspond to dosages of 1360, 680, 340 and 170 mg/kg bw. Post observation period: 7 days

EXAMINATIONS
Animals were inspected for signs of pharmacologic or toxicologic effects during a 7 days post observation period. Body weight was measured before dosing. At the end of the observation period survivors were sacrificed and necropsied as were animals that died.
The approximative mean lethal dose (LD50) was estimated (calculation method not mentioned).

Test substance: 6-Methyl-5-hepten-2-one, purity 98 %

Reliability: (2) valid with restrictions
Meets national standard methods with acceptable restrictions

16-APR-2003 (88)

5.2 Corrosiveness and Irritation

5.2.1 Skin Irritation

Species: rabbit
Concentration: undiluted
Exposure: Occlusive
Exposure Time: 20 hour(s)
No. of Animals: 4
Vehicle: other: none
Result: slightly irritating

Method: other
Year: 1973
GLP: no
Test substance: as prescribed by 1.1 - 1.4

Method: Method applied similar to OECD 404
Deviations: exposure times: 1, 5, 15 min and 20 hrs; occlusive application; 2 animals per exposure time; examinations: after exposure (only 1, 5, 15 min), 24 hrs, 48 hrs, 5 days, 6 days and 8 days after application

Result: LOCAL EFFECTS
Application of test substance for 1 min - 15 min did not lead to any skin findings. Application for 20 hrs lead to questionable to slight redness (score = (+)) after 24 hrs which was fully reversible in all animals within 48 hrs, signs of edema were not observed, a slight scaling occurred in 1 animal.

SYSTEMIC TOXICITY
No mortality occurred. There were no signs of clinical toxicity from the dermal exposure.

Test condition: TEST ANIMALS
Strain: White Vienna rabbits
Sex: 2 males, 2 females
Source: M. Gaukler, Offenbach, Germany
Weight at study initiation: about 2.9 kg

ADMINISTRATION/EXPOSURE
Preparation of test substance: test substance was used as delivered
Area of exposure: 2.5 cm x 2.5 cm, back of the animals
Vehicle: not used
Total volume applied: cotton pad (size: 2.5 x 2.5 cm) was saturated with the test substance (approximat. 0.5 ml)
Exposure times: 1, 5, 15 min and 20 hrs
Animals per exposure time: 2
Removal of test substance: after exposure substance remnants were removed with a 50% Lutrol (polyethylenglykol) dilution.
Post exposure period: 8 days

EXAMINATION
Observation period: 24, 48 hrs, 5, 6 and 8 days
Scoring system:
Erythema and edema were scored according the below described system. Although the results in the report were originally not given as Draize scores the data can be transferred into the Draize scoring system (in brackets).

(+) = none - negligible effect (0)
+ = slight effect (1)
++ = moderate effect (2)
+++ = severe effect (>= 3)
N = necrosis

Test substance: 6-Methyl-5-hepten-2-one, purity 98 %

Conclusion: Occlusive application of 6-methyl-hept-5-en-2-one for 20 hrs to rabbit skin lead to slight signs of irritation which were almost reversible within 48 hrs.

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

Flag: Critical study for SIDS endpoint

16-APR-2003 (88)

Species: rabbit

Exposure: no data

Exposure Time: 5 day(s)
No. of Animals: 3
Vehicle: no data

Method: other
Year: 1967
GLP: no
Test substance: other TS

Result: Very slight erythmea and edema days 1 and 2.
Well defined erythema and very slight to no edema days 3, 4 and 5.
Very slight erythema day 8.
No edema observed day 8.
Test condition: A liberal amount of test material was applied twice daily for 5 days to abraded and unbraded skin areas of 3 rabbits. The reactions were graded daily during application and on day 8 per Draize method (1955).
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (4) not assignable
Documentation insufficient for assessment
13-JUN-2003 (94)

5.2.2 Eye Irritation

Species: rabbit
Concentration: undiluted
Dose: .05 ml
Comment: not rinsed
No. of Animals: 2
Vehicle: other: none
Result: slightly irritating

Method: other: comparable to OECD 405
Year: 1973
GLP: no
Test substance: as prescribed by 1.1 - 1.4

Method: Deviations from OECD 405: 2 test animals; eye examination 24 hrs before testing not mentioned in the report
Result: The treatment lead to the following effects at the different observation times (Draize score in brackets).

1 hour: conjunctical redness: + (1), chemosis: ++ (2), corneal opacity: + (1)
24 hours: conjunctival redness: + (1), corneal opacity: + (1), chemosis: none (0)
8 days: none; all effects were reversible within the post observation time.
Test condition: TEST ANIMALS
Strain: White Vienna rabbits
Sex: 1 male, 1 female
Source: M. Gaukler, Offenbach, Germany
Weight at study initiation: about 2.9 kg
Controls: left eye treated with 0.9 % saline served as control

EXPOSURE

Post exposure period: 8 days

EXAMINATION
Observation period: 1, 24, 48, 72 hrs and 8 days

Scoring system:
Conjunctival redness, chemosis and corneal opacity were scored according the below described system. Although the results in the report were originally not given as Draize scores the data can be transferred into the Draize scoring system (in brackets).

(+) = none - negligible effect (Draize score 0)
+ = slight effect (1)
++ = moderate effect (2)
+++ = severe effect (> = 3)

Test substance: 6-Methyl-5-hepten-2-one, purity 98 %
Conclusion: The test substance led to slight corneal opacity and slight conjunctival redness 24 hours after application of the test substance into the eyes. All effects were completely reversible within 8 days of observation.
Reliability: (2) valid with restrictions
Comparable to guideline study with acceptable restrictions.
Flag: Critical study for SIDS endpoint
15-APR-2003 (88)

5.3 Sensitization

Type: Open epicutaneous test
Species: guinea pig
Concentration 1st: 3 %
No. of Animals: 6
Vehicle: other: see TC
Result: not sensitizing

Method: other: Klecak et al., J. Soc. Cosmet. Chem., 28, 53-64, 1977
Year: 1985
GLP: no
Test substance: no data

Remark: The results were taken from a review which discusses the Freund's Complete Adjuvant Test and the Open Epicutaneous Test. Data for about 300 fragrance raw materials were presented in tabular format.
Result: Methylheptenone tested at a concentration of 3% was a non sensitizer.
Test condition: The applied protocol was descibed in a general manner:

TEST ANIMALS
Male and female guinea pigs, weighing 300-450 g, were used. Experimental groups of at least 6 guinea pigs for every concentration group were utilized. For controls, 12 animals are used.

ADMINISTRATION AND EXPOSURE
The test material was applied epicutaneously, uncovered and if possible and relevant dissolved, suspended or emulsified at concentrations of 30, 10, 3 and 1% or lower in ethanol,

acetone, water, PEG or petroleum. Constant volumes of each concentration were applied with a pipette or syringe on standard areas of the clipped flank of each animal.

Pretest:
1 day before starting the induction procedure, the threshold irritating concentration of the test material was estimated. A single application of 0.025 ml of each test concentration (e.g. 100, 30, 10 and 3%) was simultaneously performed on the clipped skin (area: 2 sq.cm). Reactions were read 24 hours after the application.

Induction:
On day 1, application of 0.1 ml of the test material (at the respective concentration) was performed to an area measuring 8 cm2 on the clipped flank. The applications were repeated daily for 3 weeks or done 5 times weekly during 4 weeks. The application sites were left uncovered and the reactions were read 24 hours after each application or at the end of each week. The maximal non irritating and the minimal irritating concentrations were determined.

Challenge:
To determine whether or not a contact sensitization was induced all groups previously treated for 21 days as well as 10 untreated, or only pretreated with the vehicle, controls were tested on days 21 and 35 on the contralateral flank with the test material at the minimal irritating and some lower primary non irritating concentrations. The substance was applied at 0.025 ml with a pipette of each concentration to skin areas of 2 sq.cm.

EXAMINATIONS
The reactions were read after 24, 48 and/or 72 hrs.

EVALUATION CRITERIA
A test material was considered allergenic at a concentration to which at least 1 of 6 animals of the concentration group concerned showed positive reactions when non irritating concentrations were used for challenge.

Test substance: methyl heptenone, no data on purity mentioned
Reliability: (4) not assignable
Data from Handbook or collection of data

22-MAR-2004 (95)

Type: other: modified Draize procedure
Species: guinea pig
No. of Animals: 10
Vehicle: no data
Result: not sensitizing

Method: other: modified Draize (1959) technique
Year: 1978
GLP: no
Test substance: other TS

Result: The Injection Challenge Concentration (ICC) was 0.1% and the Application Challenge Concentration (ACC) was 20%. Methylheptenone was reported to be a non-sensitizer.

Test condition: A modified Draize procedure was used to test 23 natural and 46 synthetic perfume ingredients.
Animals:
Tests were carried out on inbred Hartley albino guinea pigs. The animals weighing about 350 g at the start of testing were used in each test which comprised either 4 males and 6 females or vise versa. Total number of animals which were used in the pretest and the maintest for methylheptenon was not clearly stated in the publication.

Preliminary irritation tests:
were done to determine concentrations suitable for sensitizing testing.
Intradermal injection: 4 animals of the same sex and weighing about 450 g were each injected intradermally on the shaved flanks 0.1 ml aliquots of a range of concentrations of test material. The reactions were examined for size, erythema and edema 24 h later and the concentration giving slight but perceptible irritation with no edema was selected as the injection challenge concentration (ICC).
Topical application: Aliquots (0.1 ml) of a range of concentrations in a (not further specified) solvent were applied in small circular areas to the shaved flanks of 4 guinea pigs of the same sex and weighing 450 g. The reactions were examined for erythema 24 h later and the highest concentration which caused no irritation was selected as the application challenge concentration (ACC).

Sensitization test:
General: In comparison to the Draize sensitization procedure (10 intradermal inductions over a period of 3 weeks), the equivalent dose was administered on one occasion as 4 intradermal injections, each 2.5 times the ICC.

0.1 ml of test substance at 2.5 times the ICC were injected intradermally at 4 sites which overlie the auxillary and 2 inguinal lymph nodes. 14 days later each animal was challenged intradermally in one flank and topically in the other with 0.1 ml aliquots at the respective ICC and ACC. The topical application area was not covered. 24 hours later the reactions were scored and apparent sensitization reactions confirmed 7 days later by a second challenge with controls included. In the absence of sensitization reactions at first challenge, the induction and challenge procedures were repeated, but this time confirmatory challenge with controls was included irrespective of any apparent sensitization reactions at the previous challenge.

Controls: At each challenge with controls, 4 previously untreated animals of the same sex and similar weight to the test animals were treated intradermally and topically on opposite flanks with 0.1 ml aliquouts of test substance at the ICC and ACC respectively.

Scoring: Reactions were examined under a Philips colour-matching unit. Each injection reaction was given a total score based on size (2 largest diameters), erythema and edema. Individual reactions were considered positive when their total score was significantly greater than the average

total score for control reactions. Application reactions were scored on a 0 to +++ scale and individual reactions were considered positive if (a) they were + or greater and (b) there were no erythema reactions in controls.

Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned

Reliability: (2) valid with restrictions
Meets generally accepted scientific standards, well documented and acceptable for assessment

Flag: Critical study for SIDS endpoint

13-JUN-2003 (96)

5.4 Repeated Dose Toxicity

Type: Sub-chronic
Species: rat **Sex:** male/female
Strain: Wistar
Route of administration: gavage
Exposure period: 3 months
Frequency of treatment: once daily
Post exposure period: no
Doses: 50, 200, 1000 mg/kg bw
Control Group: yes, concurrent vehicle
NOAEL: < 50 mg/kg bw
LOAEL: = 50 mg/kg bw
other: NOAEL females : = 50 mg/kg bw

Method: OECD Guide-line 408 "Subchronic Oral Toxicity - Rodent: 90-day Study"
Year: 2001
GLP: yes
Test substance: as prescribed by 1.1 - 1.4

Result: ANALYSES
- Stability of the test subtance: was demonstrated in olive oil over a period of 7 days at room temperature
- Concentration control: correctness of the concentrations were confirmed. The recovery rates were within a range of 94% - 98% of the target concentrations.

CLINICAL EXAMINATIONS
- Mortality: No animal died during the administration period.

- Clinical signs: All animals of the high dose group showed slight to moderate salivation on several days from day 8 until the end of the study. Other treatment related findings were not observed.

- Food consumption: Food consumption in females of the high dose group was statistically significantly decreased (up to -13%) from day 28 to day 49. This finding was assessed as being related to treatment. In the other dose groups no treatment dependent changes were observed.

- Body weight data: Body weight in male animals of the high dose group was throughout the whole study period decreased with a maximum of -7.2% on study day 91. The body weight change of these high dosed males was also decreased. Although these effects were not statistically significant, the

impairment of body weight as well as body weight change in male animals of the high dose group was assessed as compound-related. Body weight in females of the high dose group was statistically significantly decreased (-6.7%) on day 63, only. Body weight change in females of this dose group was statistically significantly decreased up to -16.4% from day 35 to day 84, with exception of day 70. These findings were also assessed as being related to treatment.

- Food efficiency: was significantly decreased in males of the high dose group on days 21, 35, 63, 77 of the study compared to controls (-23%, -31%, -37% and -56%, respectively). This was assessed as being related to treatment.

- Ophthalmoscopy: No substance-related effects were obtained.

- Functional observational battery and motor activity measurement
All findings were assessed as being incidental, as they occurred in single animals, only, or were equally distributed between treated groups and controls.

- Estrous cycle determination: No substance-related effects were obtained.

CLINICAL PATHOLOGY
- Hematology: At the end of the administration period increased platelets were found in the peripheral blood of the high dose animals of either sex. Platelet counts were also higher in the mid dose females. The other hematology examinations did not reveal treatment-related changes.

- Clinical chemistry: After 4 weeks of test substance administration aspartate aminotransferase activities were decreased in the high dose animals of either sex. A slight increase in alkaline phosphatase activity was also seen in the high dose males. No treatment-related changes were observed in the other serum enzyme examinations. Blood chemistry examinations revealed increased calcium, total protein, albumin and cholesterol concentrations in the high dose animals of both sexes. Furthermore, in the sera of the high dose females chloride concentrations were decreased and inorganic phosphate, urea, total bilirubin, globulins and magnesium levels were increased. No test substance-related changes were noted in the other blood chemistry examinations.

- Urinalyses: At the end of the administration period significantly increased ketone levels were detected in the urine specimens of the mid and high dose males and in the high dose females. Moreover, the urine samples of 5 out of 10 high dose males appeared cloudy and the reagent strip test indicated increased blood in the urine specimens of the high dose males. Microscopic examination of the urine sediments of the high dose males revealed increased numbers of degenerated renal tubular epithelial cells and degenerated transitional epithelial cells as well as granular casts and epithelial cell casts at the end of the study. In the urine specimens of the high dose females increased urobilinogen levels were also detected. The test compound did not affect the other urine

parameters.

- Sperm analysis: In 3 out of 10 male animals of the high dose group no spermatids per gram testis and a significant reduction in the number of spermatozoa per gram cauda epididymis were observed. In these animals, a significant increase in sperm with abnormal morphology was seen and the sperm motility could not be evaluated due to insufficient number of motile sperm.

PATHOLOGY
- Absolute organ weights: The mean liver weight was significantly increased in males (+29.6%) and in females (+21.9%) of the high dose group. The mean kidney weight was significantly increased in males of the high (+28.0%), mid (+16.5%) and low dose groups (+14.3%) in a dose-related fashion and in females of the high dose group (+14.3%). The mean weight of the epididymides was incidentally although significantly increased in males of the low dose group (+8.1%).

- Relative organ weights (related to terminal body weight): The mean liver weight was significantly increased in males (+40.7%) and in females (+29.7%) of the high dose group. The mean kidney weight was significantly increased in males of the high (+38.7%), mid (+16.3%) and low dose groups (+11.6%) in a dose-related fashion and in females of the high dose group (+23.4%). In females of the mid dose group, the mean kidney weight was comparable to the control (100.6%), whereas the mean kidney weight in the low dose group was slightly although significantly decreased (-6.8%). The mean weights of the adrenal glands were significantly increased in males (+17.6%) and in females (+15.2%) of the high dose group, and in females, only, the mean spleen weight was also slightly although significantly increased (+17.2%). The mean heart weight was incidentally although significantly decreased in females of the mid dose group (-4.8%).
- Gross lesions: were noted in the epididymides and testes in 3 of the high dose males (organ size reduced), glandular stomach (erosion/ulcer or hyperemia), skin (sparse hair), thyroid glands (organ size reduced) and vagina (inflammation and malformation in one high dose female). With the exception of reduced organ sizes of testes and epididymides in high dose males, they were either single observations or they were biologically equally distributed over the control and treatment groups with no obvious relationship to treatment.

- Histopathology
Not all of the gross lesions could be correlated with a meaningful microscopic finding: the grossly decreased in size thyroid glands (unilateral) in one control and one mid dose male were in deed smaller than their contralateral organ mate, however, morphologically, no abnormalities were detected. Also, in the skin, areas described grossly to have shown sparse hair, were without a morphologic correlate. Finally, the grossly described inflammation and malformation in the vagina of one high dose female was without a histopathologic counterpart. However, regardless of whether or not they had a microscopic correlate, all these gross lesions were considered

anyway to have developed spontaneously and unrelated to treatment.

Treatment related microscopic findings were detected in the kidneys. They consisted of an increased accumulation of alpha-2u-globulin (1/1/1/10) in the epithelia and tubular lumina of the proximal tubules of the renal cortex. While minimal (2/4/0/0) alpha-2u-globulin accumulation was only seen in control and low dose animals, slight accumulation occurred more often in the mid dose group (5/5/8/0), and moderate accumulation was most often recorded in the high dose group (1/0/2/10). The accumulation of alpha-2u-globulin was regarded to have caused the significantly increased mean absolute and relative kidney weights in males of all dose groups, although morphology did not distinguish kidneys of the control group from those of the low dose group. Only the number of males with no or virtually no alpha-2u-globulin accumulation was two animals in the control group and one in the low dose group. Comparative immunohistochemistry for a2-u globulin between male control and low dose animals showed that the Mallory-positive material in the renal cortex epithelia correlated with alpha-2u-globulin immunoreactivity and that there was a quantitative higher amount of alpha-2u-globulin in the renal cortex of treated (low dose) males as compared to the control males.
The accumulation of alpha-2u-globulin was associated with focal or multifocal cystic dilation of tubulus lumina in treated male rats (0/1/3/8). The graded severity was minimal to moderate. While the minimal and/or unilateral occurrence of this finding was interpreted possibly also spontaneous, slight or moderate cystic tubular dilation (0/0/2/7) was regarded most likely treatment-related. Any further alterations of the tubular epithelia like cell sloughing into lumen, necrobiosis, necrosis and/or regenerative proliferation were not recorded.

Although no correlate was obtained that might explain the significantly increased mean absolute and relative kidney weights in high dose females, a relationship of this finding to treatment was assumed.

Treatment related microscopic findings were also detected in the liver. They consisted of minimal or slight centrolobular hypertrophy of the liver cells in males (0/0/0/10) and in females (0/0/0/10). This finding correlated well with the recorded significantly increased mean absolute and relative liver weights of either sex. No further relevant liver lesions were noted, histopathologically.

Three males of the high dose group revealed extreme diffuse atrophy of the testes, associated with aspermia and luminal debris in the epididymides. This was regarded to be treatment-related. Two other males of the high dose group revealed minimal or slight focal tubular atrophy in the testes, with two or ten tubuli being affected, respectively. Although minimal or slight focal atrophy are also known to occur spontaneously, this finding was also interpreted treatment-related, as no such finding was noted in any of the control, low or mid dose group animals.

Histopathology failed to correlate the significantly altered weights of epididymides (absolute, increased, low dose group), adrenal glands (relative, increased, males and females, high dose group), kidneys (relative, decreased, females, low dose group), heart (relative, decreased, females, mid dose group) and spleen (relative, increased, females, high dose group) with a meaningful microscopic finding. A relationship of these weight alterations to treatment was, however, denied for the following reasons:

- slightly although significantly increased mean absolute weight of epididymides in low dose males (+8.1%):
-- no dose-response relationship
-- slight, although not significant increase of the mean terminal body weight (+3.0%) and, hence, no such observation in the more reliable relative weight
-- no morphologic correlate;

- significantly decreased mean relative kidney weight in females of the low dose group (-6.8%) and significantly decreased mean relative heart weight in females of the mid dose group (-4.8%), because of
-- no dose-response relationship
-- reverse trend (kidneys)
-- lack of a microscopic correlate in the respective organs of the high dose group;

- significantly increased mean relative adrenal weights in males and in females of the high dose group, as there was
-- a slight, although not significant decrease of the mean terminal body weight in males (-7.9%) and in females (-6.2%)
-- no morphologic correlate for the weight increase in this organ in either sex.

- significantly increased mean relative spleen weight in high dose females, as
-- The slight although not significant decrease of terminal body weight might have interfered
-- no such observation was made in males
-- no relevant histopathologic finding was noted.

All other microscopic findings recorded were either single observations, or they were recorded at a low incidence, or they occurred in control animals only, or at comparable incidence and graded severity in control and high dose males and/or females.

Test condition: TEST ORGANISM
- Strain: CrlGlxBrl/Han:WI (Supplier: Charles River, Germany)
- Age at study initiation (day 0): 41 - 43 days
- Weight at study initiation: males: ca. 140-170 g (mean ca. 157 g), females: ca. 115-130 g (mean ca. 123 g)
- Number of animals per group: 10 per dose and sex

ADMINISTRATION / EXPOSURE
- Duration of test/exposure: 90 days
- Treatment: orally by gavage
- Dosages: 50, 200, 1000 mg/kg bw/day (selected by a 4 weeks range finding study)
- Administration volume: 5 ml/kg bw

- Vehicle: olive oil (Ph.Eur./DAB)
- Preparation of test formulation: olive oil was taken in a graduated flask. The appropriate amount of test substance was weighed in, filled up to the desired volume with the vehicle, and mixed using a magnetic stirrer. These solutions were prepared in intervals of no longer than 7 days and stored under N2.
- Stability of test substance in vehicle: was determined over a period of 7 days at room temperature prior to the start of the study. As the preparations were clear solutions, no homogeneity analyses were carried out. Concentration control analyses of the test substance preparations were performed in all concentrations at the start and the end of the administration period.

REGULATORY GUIDELINES
OECD No. 408 (adopted Sept. 21, 1998) and EC Commission Directive 87/302/EEC of. Nov. 18, 1987, Official J. Europ. Communities No. L 133, p. 8 - 11, 1988

GLP
This study was conducted in accordance with the OECD Principles of Good Laboratory Practice and with the GLP regulations of the German "Chemikaliengesetz" (Chemicals Act).

CLINICAL OBSERVATIONS
- Clinical signs: Twice a day for evident signs of toxicity or mortality on weekdays (morning and afternoon), once at weekends (morning) and additionally daily after application of the test substance. Detailed clinical observations outside the home cage in an open field (50x50 cm with sides of 25 cm high) were performed prior to the start of the administration period and weekly thereafter. The findings were ranked according to the degree of severity, if applicable. The following parameters were examined: behavior during "handling", fur, skin, posture, salivation, respiration, activity/arousal level, tremors, convulsions, abnormal movements, impairment of gait, lacrimation, palpebral closure, exophthalmus, feces (appearance/consistency), urine and pupil size.

- Mortality: twice daily (monday - friday), once daily (saturday and sunday)
- Body weight: before the start of administration, thereafter once weekly.
- Food consumption: once weekly
- Food efficiency: was calculated based upon individual values for body weight and food consumption.

- Ophthalmoscopic examination: Prior to the start of the administration period the eyes of all animals were examined for any changes using an ophthalmoscope after administration of a mydriatic. At the end of the study, the animals of the control and high dose group were examined.

- Functional observational battery (FOB):
was performed in all animals towards the end of the study, starting at about 10.00 a.m.. The FOB started with passive observations without disturbing the animals, followed by removal from the home cage, open field observations in a

standard arena and sensorimotor tests as well as reflex tests. The findings were ranked according to the degree of severity, if applicable.
-- Home cage observations:
The animals were observed in their closed home cages; any disturbing activities were avoided during these examinations in order not to in-fluence the behavior of the animals. Attention was paid to posture, tremor, convulsions, abnormal movements, impairment of gait and general observations.
-- Open field observations:
The animals were transferred to a standard arena (50x37.5 cm with sides of 25 cm high) and observed for at least 2 minutes.

Following parameters were examined: behavior when removed from cage, fur, skin, salivation, nose discharge, lacrimation, eyes / pupil size, posture, palpebral closure, respiration, tremors, convulsions, abnormal movements / stereotypics, impairment of gait, activity/arousal level, feces, urine and number of rearings.
-- Sensorimotor tests / reflexes:
The animals were removed from the open field and subjected to following sensorimotor or reflex tests:
approach response, touch response, vision, pupillary reflex, pinna reflex, audition, coordination of movements, behavior during, vocalization, pain perception, grip strength of forelimbs, grip strength of hindlimbs, landing foot-splay test

- Motor activity assessment:
was measured on the same day as FOB was performed. The measure-ment was performed in the dark using the Multi-Varimex-System with 4 infrared beams per cage. During the measurement the animals were kept in Polycarbonate cages with absorbent material. The measurements started at about 2.00 p.m. and the number of beam interrupts were counted over 12 intervals, each lasting 5 minutes. The period of assessment for each animal started when the first beam was interrupted by pushing the cage into the rack. Measurements ended exactly 60 minutes thereafter.

- Estrus cycle determination:
Vaginal smears for cycle determination were prepared in the morning and evaluated from day 63 until day 91 of the study. The differentiation was conducted to following stages:

Cycle stage Appearance in vaginal smear

Diestrous Leucocytes, few nucleated, epithelial cells
Proestrous Single leucocytes, many nucleated and few horny epithelial cells
Estrous Only horny epithelial cells
Metestrous Leucocytes, some horny epithelial cells and some nucleated epithelial cells

CLINICAL PATHOLOGY
Blood was taken from the retroorbital venous plexus in the morning from fasted animals without anesthesia. For urinalysis the animals were transferred to metabolism cages. At necropsy specimen were sampled from fasted anesthetized male animals in a randomized sequence for sperm analyses. The following

examinations were carried out in 10 animals per test group and sex at the end of the application period.

- Hematology
The following parameters were determined in blood with EDTA-K3 as anticoagulant using a hematology analyzer: leukocytes, erythrocytes, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, platelets, differential blood count. Prothrombin time was determined using a ball coagulometer.

- Clinical chemistry
An automatic analyzer was used to examine the following clinicochemical parameters.
alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, serum gamma-glutamyltransferase, sodium, potassium, chloride, inorganic phosphate, calcium, urea, creatinine, glucose, total bilirubin, total protein, albumin, globulins, triglycerides, cholesterol, magnesium.

- Urinalysis
The following examinations were carried out: volume, color, turbidity, pH, protein, glucose, ketones, urobilinogen, bilirubin, blood, specific gravity, sediment. With the exception of volume, color, turbidity, sediment examination and the specific gravity, all the uine constituents were determined semiquantitatively using test strips and a reflection photometer. The specific gravity was determined using a urine refractometer. The sediment was evaluated microscopically.

- Sperm parameters
Immediately after necropsy and organ weight determination the right testis and cauda epididymis were taken from all male animals. The following parameters were determined: sperm motility, sperm morphology, sperm head count (cauda epididymis), sperm head count (testis)

PATHOLOGY
- Necropsy
The animals were sacrificed by decapitation under CO2 anesthesia. The exsanguinated animals were necropsied and assessed by gross pathology.

- Organ weights
The following weight parameters from all animals sacrificed at scheduled dates were determined: Anesthetized animals, liver, kidneys, adrenal glands, testes, epididymides, ovaries, uterus, spleen, brain, heart, thymus, prostate gland

- Histopathology
The following organs were fixed in 4% formaldehyde solution, histopathologically processed and examined by light microscopy:
All gross lesions, salivary glands (mandibular and sublingualis), esophagus, stomach (forestomach and glandular stomach), duodenum, jejunum, ileum, cecum, colon, rectum, liver, pancreas, brain, pituitary gland, sciatic nerve, spinal cord (cervical, thoracic and lumbar cord), eyes, adrenal

glands, thyroid glands, parathyroid glands, trachea, lungs, pharynx, larynx, nose (nasal cavities), aorta, heart, bone marrow (femur), lymph nodes (mandibular and mesenteric), spleen, thymus, kidneys, urinary bladder, ovaries, oviducts/uterus/vagina, prostate gland, seminal vesicles, female mammary gland, skin, skeletal muscle, sternum with marrow, femur with knee joint, extraorbital lacrimal glands The left testis and the left epididymides were fixed in BOUIN's solution and embedded in paraplast. In the kidneys immunohistochemical staining and according to Mallory Heidenhain for alpha-2u-globulin detection was performed.

STATISTICAL METHODS
Means and standard deviations of each test group were calculated for several parameters. Further statistical analyses were performed.

- Dunnett test:
Food consumption, body eight, body weight change, food efficiency, mean estrous stages

- Kruskall-Wallis test:
Feces, rearing, grip strength length forelimbs, grip strength length hindlimbs,
landing foot-splay test, motor activity, clinical pathology except differential blood count, pathological weight parameters (if p-value < = 0.05 Wilcoxon test was additionally performed)

- Fishers exact test:
Urinalysis except volume, color, turbidity and specific gravity; abnormal sperm > 4%

- Wilcoxon test
Total spermatids/g testis, total sperm/g cauda epi., % motility

Test substance: purity: 99.1% (GC-method)
Reliability: (1) valid without restriction
Well conducted guideline study conducted under GLP conditions.
Flag: Critical study for SIDS endpoint
23-MAR-2004 (97)

5.5 Genetic Toxicity 'in Vitro'

Type: Bacterial forward mutation assay
System of testing: Salmonella typhimurium TM677
Concentration: 0.31 - 5 mg/ml
Metabolic activation: with and without
Result: negative

Method: other: Pezzuto et al., Proc. Natl. Acad. Sci. USA, 82, 2478, 1985
Year: 1987
GLP: no data
Test substance: other TS

Result: 0.31 - 5 mg/ml: No significant mutagenic activity was

observed.
Test condition: Forward mutation assays were conducted using Salmonella thyphimurium strain TM677 carrying the R-factor plasmid pKM101, in the presence and absence of S9 actvation. Duplicate 1 ml reaction mixtures containing 1 mg NADP+, 1 mg of glucose 6-phosphate, 0.8 unit glucuse-6-phosphate dehydrogenase, 0.67 mg of MgCl2, S-9 mix and approx. 7x10(6) bacteria were prepared in minimal essential medium. If metabolic acticvation was not required, only the bacterial and minimal essential medium were mixed. The test material was dissolved in 20 µl DMSO. After the test substance was added, the mixtures were slowly rotated for 2 hours at 37°C. The reaction was quenched by adding 4 ml phosphate-buffered saline. The bacteria were recovered by centrifugation, then resuspended, diluted and plated in triplicated in the absence and presence of 8-azaguanine. The plates were allowed a 36-to 40-hour growth period at 37oC, after which they were scored. The mutant fraction was expressed as the average number of colonies observed on plates containing 8-azaguanine divided by the average number of colonies observed on plates not containing 8-azaguanine.
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (3) invalid
Significant methodological deficiencies; unusual tester strain, bacterial strains recommended in OECD 471 not used, insufficient documentation of method and results
08-JUL-2003 (91)

Type: Ames test
System of testing: Salmonella typhimurium TA98, TA100, TA1535, TA1537
Concentration: 3 µmol/plate (ca. 378 µg/plate)
Metabolic activation: with and without
Result: negative

Method: other: nach Ames et al.: Mut. Res., 31, 347, 1975
Year: 1979
GLP: no
Test substance: other TS

Result: Results given in tabular form. Methylheptenone was found to be not mutagenic.
Test condition: The S-9 fractions for metabolic activation were prepared from Aroclor induced male rats (500 mg/kg intraperitoneally for 5 days). Vehicle was ethanol. The positive controls were N-Methyl-N'-nitro-N-mitrosoguanidin (without activation) and 2-aminoanthracene (with activation).
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (3) invalid
Significant methodological deficiencies, guideline dose levels not achived, insufficient documentation of results
08-JUL-2003 (98)

Type: Ames test
System of testing: Salmonella typhimurium TA 1535, TA 100, TA 1537, TA 98 and E. coli WP2 uvrA
Concentration: 20 - 5,000 µg/plate (SPT and PIT)
Cytotoxic Concentration: > 2,500 µg/plate (SPT); > 1,000 - 2,000 µg/plate (PIT)
Metabolic activation: with and without
Result: negative

Method: OECD Guide-line 471
Year: 2002
GLP: yes
Test substance: as prescribed by 1.1 - 1.4

Result: STABILITY OF THE TEST SUBSTANCE PREPARATION:
Has been verified at room temperature in the vehicle DMSO over a period of 4 hours.
SOLUBILITY
No test substance precipitation was found.

TOXICITY
A weak bacteriotoxic effect (slight decrease in the number of revertants and/or slight reduction in the titer) was occasionally observed in the standard plate test depending on the strain and test conditions from about 2,500 µg/plate onward. In the preincubation assay bacteriotoxicity (reduced background growth, decrease in the number of revertants, reduction in the titer) was observed depending on the strain and test conditions from about 1,000 µg - 2,000 µg/plate onward.

MUTAGENICITY
An increase in the number of his+ revertants was not observed both in the standard plate test and in the preincubation test either without S-9 mix or after the addition of a metabolizing system (see tables below).

REVERSION FREQUENCIES

Results as mean values from 3 plates

MNNG = M-methyl-N'-nitro-N-nitrosoguanidine
AA = 2-aminoanthracene
NQO = 4-nitroquinoline-N-oxide
AAC = 9-aminoacridine
NOPD = 4-nitro-o-phenylendiamine
B = reduced background growth

1st Experiment: Standard plate test

Strain	TA 1535		TA 100		TA 1537	
Dose (µg)	-S-9	+S-9	-S-9	+S-9	-S-9	+S-9
Vehicle	17	18	103	111	10	10
20	18	16	94	121	8	12
100	16	15	100	107	8	14
500	15	14	108	120	13	9
2,500	21	16	115	106	11	8
5,000	16	13	119	44	9	5
MNNG	556		584			
AA		149		1072		169
AAC					488	

Strain	TA 98		E. coli WP2 uvrA	

Dose (µg)	-S-9	+S-9	-S-9	+S-9
Vehicle	25	34	26	29
20	32	25	23	25
100	34	27	23	23
500	32	25	22	21
2,500	23	24	20	18
5,000	17	8	13	18
NOPD	961			
AA		613		223
NQO			622	

2nd Experiment: Preincubation test

Strain	TA 1535		TA 100		TA 1537	
Dose (µg)	-S-9	+S-9	-S-9	+S-9	-S-9	+S-9
Vehicle	17	18	110	103	8	11
20	16	16	122	109	9	9
100	15	17	116	115	10	10
500	15	13	130	111	7	10
2,500	11	3	98	42	6	5
5,000	0B	0B	0B	0B	0B	0B
MNNG	554		584			
AA		125		581		106
AAC					401	

Strain	TA 98		E. coli WP2 uvrA	
Dose (µg)	-S-9	+S-9	-S-9	+S-9
Vehicle	25	31	39	33
20	24	30	31	28
100	20	25	26	30
500	14	22	31	27
2,500	10	7	18	9
5,000	0B	0B	0B	0B
NOPD	862			
AA		674		246
NQO			551	

3rd Experiment: Preincubation test

Strain	TA 1535		TA 100		TA 1537	
Dose (µg)	-S-9	+S-9	-S-9	+S-9	-S-9	+S-9
Vehicle	18	16	108	111	9	9
125	14	15	107	100	10	12
250	13	10	97	85	11	11
500	14	10	104	80	8	8

1,000	14	12	100	85	7	6
2,000	13	9	81	54	4	6

MNNG	1143		1393			
AA		214		674		118
AAC					555	

Strain	TA 98		E. coli WP2 uvrA	
Dose (µg)	-S-9	+S-9	-S-9	+S9
Vehicle	26	32	31	28
125	24	24	36	28
250	24	25	33	28
500	18	22	31	23
1,000	14	18	26	20
2,000	12	20	20	11

NOPD	667			
AA		574		230
NQO			793	

Test condition: SYSTEM OF TESTING
- Metabolic activation system: S-9 mix from rat liver, induced with Aroclor 1254,
- Standard Plate Test and Preincubation Test (SPT and PIT)

ADMINISTRATION
Number of replicates: 3 experiments (1 x standard plate test +/- S-9 mix; 2 x preincubation test +/- S-9 mix
Plates per test: 3 per dose or per control
Negative controls: sterility control (soft agar, S-9 mix, buffer vehicle or the test substance but without tester strains) and vehicle control were carried out
Positive control groups and treatment: - S-9 mix: 5 µg N-methyl-N'-nitro-N-nitrosoguanidine for TA 100 and TA 1535, 10 µg 4-nitro-o-phenylendiamine for TA 98, 100 µg 9-aminoacridine chloride monohydrate for TA 1537; 5 µg 4-nitroquinoline-N-oxide for E. coli WP2 uvrA
+ S-9 mix: 2.5 µg 2-aminoanthracene for TA 1535, TA 100, TA 1537, TA 98, 60 µg 2-aminoanthracene for E. coli WP2 uvrA
Solvent: DMSO

REGULATORY GUIDELINES
OECD No. 471 (July 21, 1997) and EEC Directive 2000/32, B.13 / B.14 (May 19, 2000)

GLP
The study was conducted in accordance with the OECD Principles of Good Laboratory Practice and with the GLP regulations of the German "Chemikaliengesetz" (Chemicals Act).

CRITERIA FOR EVALUATION
The test chemical was considered positive if the following criteria were met:
- A dose-related and reproducible increase in the number of revertant colonies, i.e. about doubling of the spontaneous mutation rate in at least one tester strain either without S-9 mix or after adding a metabolizing system.

A test substance was considered nonmutagenic if the number of revertants for all tester strains were within the historical negative control range under all experimental conditions in two experiments carried out independently of each other.

Test substance: purity: 99.1%

Conclusion: The test substance 6-methyl-5-en-2-one was not mutagenic in the Ames test under the conditions chosen.

Reliability: (1) valid without restriction
GLP guideline study

Flag: Critical study for SIDS endpoint

23-MAR-2004 (99)

5.6 Genetic Toxicity 'in Vivo'

Type: Micronucleus assay
Species: mouse **Sex:** male
Strain: NMRI
Route of admin.: i.p.
Exposure period: 2 injections at a 24-hour interval
Doses: 200, 400 and 800 mg/kg bw
Result: negative

Method: OECD Guide-line 474 "Genetic Toxicology: Micronucleus Test"
Year: 2001
GLP: yes
Test substance: as prescribed by 1.1 - 1.4

Result: MORTALITY
No mortality occurred in all groups.

CLINICAL SIGNS
The administration of the test substance at 2 x 800 mg/kg bw led to evident signs of toxicity in all treated animals (poor general state, abdominal position, squatting posture, staggering) which were reversible after 2 days. At the 2 lower doses only minor signs of clincal toxicity were observed after 1 hour of administration of the test substance (squatting posture).

EFFECT ON PCE/NCE RATIO
No inhibition of erythropoiesis, determined from the PCE/NCE ratio was detected. The vehicle and the the positive control substances, CPP and VCR, caused no evident signs of toxicity.

Mean number of PCEs and NCEs (Interval: 24 hrs)

	PCEs	NCEs
vehicle	10,000	2,746
200 mg/kg bw	10,000	2,348
400 mg/kg bw	10,000	2,750
800 mg/kg bw	10,000	2,627
CPP (20 mg/kg bw)	10,000	4,129
VCR (0.15 mg/kg bw)	10,000	4,212

GENOTOXIC EFFECTS
Mean number of PCEs containing MN per 1,000 PCE at 24 hrs:
vehicle: 0.3
200 mg/kg bw: 1.0

400 mg/kg bw: 0.9
800 mg/kg bw: 0.7
CPP (20 mg/kg bw): 16.0 (p < = 0.01)
VCR (0.15 mg/kg bw): 52.9 (p < = 0.01)

STATISTICAL EVALUATION
The administration of the test substance did not lead to any statistical significant increase in the number of polychromatic erythrocytes containing either small or large micronuclei. The rate of micronuclei was nearly the range of the concurrent negative control in all dose groups and within the range of the historical control data.

Test condition: TEST ORGANISM
Male healthy Crl:NMRI mice (breeder: Charles River Deutschland GmbH, GER) with a mean weight of about 28 g (with an age range of about 5-8 weeks according to the information of the breeder), 5 animals per dose and group

ADMINISTRATION
Vehicle: olive oil (quality: Ph.Eur/DAB)
Frequency of dosing: 2 injections at a 24 hr interval
Dosing volume: 10 ml/kg bw
Control groups:
negative: 2 x vehicle control (10 ml/kg bw olive oil)
positive: 1 x 20 mg/kg bw cyclophosphamide (CPP) for clastogenic effects (10 ml/kg bw), 1 x 0.15 mg/kg bw vincristine (VCR) for aneugenic effcts (10 ml/kg bw)

TEST CONDITIONS

Sampling times: 24 hrs after the last treatment samples of bone marrow of the 2 femora were taken and prepared.
Preparation of the bone marrow: according to the method of Schmidt (1976 and 1977) and Salamon et al. (1980)

Microscopic evaluation: 2000 polychromatic erythrocytes (PCEs) from each animal of every test group were investigated for micronuclei (MN). The normochromatic erythrocytes (NCEs) were also scored. The ratio of polychromatic to normochromatic erythrocytes was determined.

Clinical observations: after administration of the vehicle, test substance and positive controls, the animals were examined for clinical signs of toxicity.

Criteria for selection of M.T.D.:
In a pretest for determination of the acute i.p. toxicity, deaths were observed down to a dose of 1,000 mg/kg bw. 800 mg/kg bw were survived by all animals but led to signs of clinical toxicity, such as staggering abdominal position, and a poor general health state. No distinct differences between male and female animals were observed. Therefore, doses of 800, 400 and 200 mg/kg bw were selected.
Statistical method: Wilcoxon test

GLP:
The study was conducted in accordance with the GLP regulations of the German Chemicals Act (Fed. Law Gazette 1994, Part I, July 29, 1994) and with the OECD Principles of Good Laboratory

Practice (Paris, 1981)

REGULATORY GUIDELINES
OECD No. 474 (July 21, 1997)
EEC Directive 2000/32, B. 12 (May 19, 2000)

EVALUATION CRITERIA
The test chemical was considered positive if the following criteria were met:
- A dose-related and significant increase in the number of micronucleated poly-chromatic erythrocytes was observed.
- The proportion of cells containing micronuclei exceeded both the values of the concurrent negative control range and the negative historical control range.

A test substance was considered negative if
- There was no significant increase in the number of micronucleated polychromatic erythrocytes at any dose above concurrent control frequencies.
- The frequencies of cells containing micronuclei were within the historical control range.

Test substance: Purity: 99.1%
Conclusion: Under the experimental conditions chosen, the test substance did not have a chromosome-damaging (clastogenic) effect, and there were no indications of any impairment of chromosome distribution in the course of mitosis (aneugenic activity) in bone marrow cells in vivo.
Reliability: (1) valid without restriction
GLP guideline study
Flag: Critical study for SIDS endpoint
23-MAR-2004 (100)

5.7 Carcinogenicity

5.8.1 Toxicity to Fertility

Type: other: sub-chronic
Species: rat
Sex: male/female
Strain: Wistar
Route of administration: gavage
Exposure Period: 3 months
Frequency of treatment: once daily
Doses: 50, 200, 1000 mg/kg bw/d
Control Group: yes, concurrent vehicle
Result: In males, testicular toxicity affecting spermatogenesis at 1000 mg/kg bw/d. No adverse effects on female reproductive organs or estrous cycle up to and including 1000 mg/kg bw/d.

Method: other: OECD TG 408
Year: 2001
GLP: yes
Test substance: as prescribed by 1.1 - 1.4

Result: ANALYSES
- Stability of the test subtance: was demonstrated in olive

oil over a period of 7 days at room temperature
- Concentration control: correctness of the concentrations were confirmed. The recovery rates were within a range of 94% - 98% of the target concentrations.

The study results concerning CLINICAL OBSERVATIONS, CLINICAL PATHOLOGY and PATHOLOGY are fully described in chapter 5.4 (Repeated Dose Toxicity). In the following only observations relevant for the endpoint reproductive toxicity are included.

CLINICAL OBSERVATIONS
- Estrous cycle determination: No substance-related effects were obtained.

CLINICAL PATHOLOGY

- Sperm analysis: In 3 out of 10 male animals of the high dose group no spermatids per gram testis and a significant reduction in the number of spermatozoa per gram cauda epididymis were observed. In these animals, a significant increase in sperm with abnormal morphology was seen and the sperm motility could not be evaluated due to insufficient number of motile sperm.

PATHOLOGY
- The mean weight of the epididymides was incidentally although significantly increased in males of the low dose group (+8.1%).

- Relative organ weights (related to terminal body weight): The relative reproductive organ weights were not statistically significantly changed.

- Gross lesions: were noted in the epididymides and testes in 3 of the high dose males (organ size reduced), and vagina (inflammation and malformation in one high dose female).

- Histopathology
The grossly described inflammation and malformation in the vagina of one high dose female was without a histopathologic counterpart.

Three males of the high dose group revealed extreme diffuse atrophy of the testes, associated with aspermia and luminal debris in the epididymides. This was regarded to be treatment-related. Two other males of the high dose group revealed minimal or slight focal tubular atrophy in the testes, with two or ten tubuli being affected, respectively. Although minimal or slight focal atrophy are also known to occur spontaneously, this finding was also interpreted treatment-related, as no such finding was noted in any of the control, low or mid dose group animals.

Histopathology failed to correlate the significantly altered weights of epididymides (absolute, increased, low dose group).

Test condition: TEST ORGANISM
- Strain: CrlGlxBrl/Han:WI (Supplier: Charles River, Germany)
- Age at study initiation (day 0): 41 - 43 days
- Weight at study initiation: males: ca. 140-170 g (mean ca.

157 g), females: ca. 115-130 g (mean ca. 123 g)
- Number of animals per group: 10 per dose and sex

ADMINISTRATION / EXPOSURE
- Duration of test/exposure: 90 days
- Treatment: orally by gavage
- Dosages: 50, 200, 1000 mg/kg bw/day (selected by a 4 weeks range finding study)
- Administration volume: 5 ml/kg bw
- Vehicle: olive oil (Ph.Eur./DAB)
- Preparation of test formulation: olive oil was taken in a graduated flask. The appropriate amount of test substance was weighed in, filled up to the desired volume with the vehicle, and mixed using a magnetic stirrer. These solutions were prepared in intervals of no longer than 7 days and stored under N2.
- Stability of test substance in vehicle: was determined over a period of 7 days at room temperature prior to the start of the study. As the preparations were clear solutions, no homogeneity analyses were carried out. Concentration control analyses of the test substance preparations were performed in all concentrations at the start and the end of the administration period.

REGULATORY GUIDELINES
OECD No. 408 (adopted Sept. 21, 1998) and EC Commission Directive 87/302/EEC of. Nov. 18, 1987, Official J. Europ. Communities No. L 133, p. 8 - 11, 1988

GLP
This study was conducted in accordance with the OECD Principles of Good Laboratory Practice and with the GLP regulations of the German "Chemikaliengesetz" (Chemicals Act).

The study conditions concerning CLINICAL OBSERVATIONS, CLINICAL PATHOLOGY, PATHOLOGY and statistical methods are fully described in chapter 5.4 (Repeated Dose Toxicity). In the following only examinations elevant for the endpoint reproductive toxicity are included.

CLINICAL OBSERVATIONS
- Estrus cycle determination:
Vaginal smears for cycle determination were prepared in the morning and evaluated according to the timetable.

CLINICAL PATHOLOGY
- At necropsy specimen were sampled from fasted anesthetized male animals in a randomized sequence for sperm analyses.

- Sperm parameters
Immediately after necropsy and organ weight determination the right testis and cauda epididymis were taken from all male animals. The following parameters were determined: sperm motility, sperm morphology, sperm head count (cauda epididymis), sperm head count (testis)

PATHOLOGY
- Necropsy
The animals were sacrificed by decapitation under CO2

anesthesia. The exsanguinated animals were necropsied and assessed by gross pathology.

- Organ weights
The following weight parameters from all animals sacrificed at scheduled dates were determined: Anesthetized animals, testes, epididymides, ovaries, uterus, prostate gland

- Histopathology
The following organs were fixed in 4% formaldehyde solution, histopathologically processed and examined by light microscopy:
All gross lesions, ovaries, oviducts/uterus/vagina, prostate gland, seminal vesicles. The left testis and the left epididymides were fixed in BOUIN's solution and embedded in paraplast.

STATISTICAL METHODS

Means and standard deviations of each test group were calculated for several parameters. Further statistical analyses were performed.

- Dunnett test:
mean estrous stages

- Kruskall-Wallis test:
pathological weight parameters (if p-value < = 0.05 Wilcoxon test was additionally performed)

- Fishers exact test:
abnormal sperm > 4%

- Wilcoxon test
Total spermatids/g testis, total sperm/g cauda epi., % motility

Test substance: purity: 99.1% (GC-method)
Reliability: (1) valid without restriction
Well conducted guideline study conducted under GLP conditions.
Flag: Critical study for SIDS endpoint
09-JUL-2003 (97)

5.8.2 Developmental Toxicity/Teratogenicity

Species: rat **Sex:** female
Strain: Wistar
Route of administration: gavage
Exposure period: Day 6-19 post conception
Frequency of treatment: once daily
Duration of test: until gestation day 20
Doses: 0, 50, 200 and 1000 mg/kg bw
Control Group: yes, concurrent vehicle
NOAEL Maternal Toxity: = 200 mg/kg bw
NOAEL Teratogenicity: > 1000 mg/kg bw
other: NOAEL developmental toxicity :
= 200 mg/kg bw
LOAEL Maternal Toxicity : = 1000 mg/kg bw

other: LOAEL developmental toxicity : = 1000 mg/kg bw
Result: not teratogenic, signs of prenatal developmental toxicity at maternal toxic dose

Method: OECD Guide-line 414 "Teratogenicity"
Year: 2002
GLP: yes
Test substance: as prescribed by 1.1 - 1.4

Result: - TEST SUBSTANCE ANALYSES
The stability of the test substance suspensions over a period of 7 days at room temperature, the homogeneity of the test substance in the vehicle and the correct concentration of the test substance in the preparation was demonstrated.

MATERNAL TOXIC EFFECTS BY DOSE LEVEL:
- Mortality and day of death: There were no substance-related or spontaneous mortalities in any of the groups.
- Clinical examinations: Each test group including the controls contained a sufficient number of females with implantation sites at necropsy (20 or more).
- Clinical symptoms: Several high dose rats showed abdominal position, ataxia and/or unsteady gait shortly after treatment. These findings were only observed on the first days of dosing. Moreover, all high dose and several mid dose rats showed transient salivation. The observed salivation was considered to be substance-induced. It is very likely, that this finding was induced by bad taste of the test substance, local affection of the upper digestive tract or as a conditioning phenomenon. Salivation itself is not assessed as an adverse or toxic effect. No disturbances of the general behavior occurred in the dams of control and low dose group.
- Food consumption: The mean food consumption of the high dose dams was statistically significantly reduced on most days of the treatment and during the posttreatment period. If calculated for the entire treatment phase (days 6 - 19 p.c.), it was about 7% below the concurrent control value. Food consumption of the mid and low dose rats was not affected by the test substance administration.
- Body weight data: The mean body weight of the substance-treated rats in all groups were not affected. The mean body weight gains of the high dose dams were statistically significantly lowered and was about 14% below the concurrent control value if calculated for days 6 - 19 p.c.. As the food consumption of these rats was also diminished and the corrected body weight gain was also decreased, this was considered to be a clear substance-related sign of maternal toxicity. Body weight gains of the dams at 50 and 200 mg/kg bw were similar to those of controls.
- Corrected body weight gain (net maternal body weight change): The corrected body weight gains (terminal body weight on day 20 p.c. minus weight of the unopened uterus minus body weight on day 6 p.c.) of the dams at 50 and 200 mg/kg bw revealed no differences of any biological relevance to the corresponding control group. The net weight change of the high dose rats was about 29% below the concurrent control value. As food consumption and body weight gain were also diminished in these animals, the effect on net body weight at the top dose

is considered to be a substance-related sign of straight maternal toxicity.

EXAMINATION OF THE DAMS AT TERMINATION
- Uterus weight: The mean gravid uterus weights of the animals of all test groups were not influenced by the administration of the test substance.
- Necropsy findings: There were no substance-related observations at necropsy in any of the dams. Two control animals and one mid dose animal showed congested lungs. In one low dose female a hemorrhagic thymus was observed. These gross findings are considered to be spontaneous in nature and are probably related to the method how the rats were killed
- Reproduction data of dams: The conception rate reached 84% at 50 mg/kg bw, 92% at 200 mg/kg bw and 96% in the controls and the high dose group. As all rats, which became pregnant had implantation sites at necropsy, a sufficient number of females for the purpose of the study was available.
There were no substance-related and/or biologically relevant differences between the different test groups in conception rate, in the mean number of corpora lutea and implantation sites or in the values calculated for the pre- and the postimplantation losses, the number of resorptions and viable fetuses. All differences observed were considered to reflect the normal range of fluctuations for animals of this strain and age.

EXAMINATION OF FETUSES
- Sex distribution of fetuses: The sex distribution of the fetuses in all test groups was comparable with that of the control fetuses.
- Weight of placentae: The mean placental weights at 1,000 mg/kg bw were statistically significantly reduced. The value was 13% lower than the control values if both sexes are combined. The impaired mean placental weight at 1,000 mg/kg bw has to be seen in association with the reduced mean fetal body weights in this group. The mean placental weights at the low and mid dose were not influenced.
- Weight of fetuses: The mean body weights of the high dose fetuses were statistically significantly reduced (about 9% below the concurrent control values if both sexes are combined). The mean fetal body weights at 50 and 200 mg/kg bw were not influenced.
- Fetal external, soft tissue and skeletal observations: The scattered occurrence of the few observed external, soft tissue and skeletal malformations in single fetuses of all test groups including the controls without a consistent pattern, without a clear dose-response relationship and/or at incidences, which are similar to historical control rates did not suggest any substance-induced origin of these findings. If all different types of malformations are summarized, in total 2 of the 196 examined control fetuses [= 1.0%] in 2 out of 24 litters [= 8.3%], 2 of the 187 examined low dose fetuses [= 1.1%] in one out of 21 litters [= 4.8%], 2 out of 219 mid dose fetuses [= 0.9%] in 2 out of 23 litters [= 8.7%] and 3 out of 204 high dose fetuses [= 1.5%] in 2 out of 23 litters [= 8.7%] showed malformations. The mean percentages of affected fetuses/litter with total malformations amounted to 1.1, 1.1, 0.9 and 1.4% at 0; 50; 200 or 1,000 mg/kg bw respectively.

These incidences do not suggest any treatment-relationship. External variations did not occur in any of the fetuses in this study. Soft tissue variations, exclusively in the form of dilated renal pelvis and ureter, occurred in all test groups including the controls without a clear relation to dosing and at incidences, which are fully within the historical control data range.
There were, however, some indications for substance-induced effects on the high dose group fetuses if the fetal and litter incidences as well as mean percentages of affected fetuses/litter with skeletal variations are taken into account. Several skeletal variations, primarily delays in the ossification process of skull, vertebral column and sternum, occurred at statistically significantly increased rates in the high fetuses at incidences, which were above the upper historical control values. These delays in skeletal maturation are in-line with marked impairments of the fetal body weights at the high dose level.

If all variations are summarized, in total 111 of the 196 examined control fetuses [= 57%] in all 24 litters [= 100%], 105 of the 187 examined low dose fetuses [= 56%] in all 21 litters [= 100%], 125 out of 219 mid dose fetuses [= 57%] in all 23 litters [= 100%] and 118 out of 204 high dose fetuses [= 58%] in all 23 litters [= 100%] showed variations. The mean percentages of affected fetuses/litter with total variations amounted to 59.4, 56.5, 57.5 and 57.8% at 0; 50; 200 or 1,000 mg/kg bw respectively. The incidences at 50 and 200 mg/kg do not suggest any treatment-relationship, but reflect the usual biological variation inherent in the strain of rats used for this experiment. The increased occurrence of some skeletal variations at the top dose level is considered to be substance-induced and related to the lower fetal body weights in this group, although the rate of overall variations does not suggest a treatment-relationship.

Thus, the oral administration of 1,000 mg/kg bw 6-Methylhept-5-en-2-one to pregnant Wistar rats caused marginal effects on fetal morphology in the presence of maternal toxicity, but no indications for teratogenicity. These variations mirror common findings on fetal morphology most probably due to fetal growth retardations and/or due to maternal stress. They are, however, not indicative for selective effects on the fetal organism. No substance-induced effects on fetal morphology occurred at the low and the mid dose level.

Test condition: TEST ORGANISMS
Strain: Sexually mature, virgin Wistar rats (CrlGlxBrlHan:WI) supplied by Charles River Laboratories (Germany)
Number: 25 female animals per group
Age at study initiation: about 70-84 days
Weight at study initiation: 149.2-184.6

REGULATORY GUIDELINES
- OECD No. 414 (proposal for updating, January 22, 2001)
- EEC Directive 87/302, November 18, 1987, Offic. J. Europ. Communities, No. L 133, pp 24-26 (1988)
- US EPA, Health Effects Test Guidelines; OPTTS 870.3700: Prenatal Developmental Toxicity Study (August 1998)

GLP
This study was conducted in accordance with the OECD Principles of Good Laboratory Practice and with the GLP regulations of the German "Chemikaliengesetz" (Chemicals Act).

ADMINISTRATION / EXPOSURE
- Duration of test/exposure: from implanation to one day prior to the expected day of parturition (day 6 to day 19 post conception). On day 20 p.c., all surviving females were sacrificed.
- Treatment: orally by gavage always at approx. the same time of day (in the morning)
- Control group and treatment: gavage application of 5 ml/kg bw olive oil
- Vehicle: olive oil (Ph.Eur./DAB)
- Test substance preparation: At the beginning of the administration period and thereafter at intervals which took into account the analytical results of the stability verification. For the preparation of the suspensions, an appropriate amount of the test substance was weighed depending on the dose group, in calibrated beakers and subsequently suspended in the vehicle using a high - speed homogenizer. A magnetic stirrer was used to keep the suspensions homogeneous during treatment of the animals.
- Concentration in vehicle: 1000, 4000 and 20000 mg/100 ml
- Total volume applied: 5 ml/kg bw
- Doses: 50, 200, 1000 mg/kg bw
- Analyses: check of stability, homogeneity and concentration control was performed by GC

MATING PROCEDURES:
The animals were mated by the breeder ("time-mated") and supplied on day 0 post coitum (= detection of vaginal plug / sperm). The animals arrived on the same day (i.e. day 0 p.c.) at the experimental laboratory. The following day was designed "day 1" post coitum (p.c.). Animals were assigned to the test groups by taken random selection.

PARAMETERS ASSESSED DURING STUDY:
- Mortality: A check was made twice a day on working days or once a day (Saturday, Sunday or on public holidays) (days 0 - 20 p.c.).
- Clinical symptoms: The animals were examined for clinical symptoms at least once a day, or more often when clinical signs of toxicity were elicited (days 0 - 20 p.c.).
- Body weight gain: All animals were weighed on days 0, 1, 3, 6, 8, 10, 13, 15, 17, 19 and 20 p.c.. The body weight change of the animals was calculated from these results.
- Food consumption: With the exception of day 0, the consumption of food was determined on the same days as was body weight.
- Corrected body weight gain (net maternal body weight change) Furthermore, the corrected body weight gain was calculated after terminal sacrifice (terminal body weight on day 20 p.c. minus weight of the unopened uterus minus body weight on day 6 p.c.).
- Examination of uterine content: Gravid uterine weight, number of corpora lutea, number and distribution of implantation sites classified as live fetuses, dead

implantations, early resorptions, late resorptions and dead fetuses. Calculations of conception rate and pre- and postimplantation losses were carried out.
- Examination of fetuses after dissection from the uterus: Litter size, fetal weight, sex ratio, grossly visible/external/soft tissue/skeletal abnormalities. The viability of the fetuses and the condition of the placentae, the umbilical cords, the fetal membranes and fluids were examined. Individual placental weights were recorded. After these examinations, approximately one half of the fetuses per dam were eviscerated, skinned and placed in ethyl alcohol, the other half was placed in BOUIN's solution for fixation and further evaluation.
- Soft tissue examination of the fetuses: The fetuses fixed in BOUIN's solution were examined for any visceral findings according to the method of BARROW and TAYLOR (1969).
- Skeletal examination of the fetuses
The skeletons of the fetuses fixed in ethyl alcohol were stained according to a modified method of KIMMEL and TRAMMELL (1981). Thereafter, the skeletons of these fetuses were examined under a stereomicroscope. After this examination the stained fetal skeletons were retained individually.
- Evaluation criteria for assessing fetuses: the glossary of WISE et al. (1997) was used a s much as possible to describe fiondings in fetal morphology.

STATISTICAL METHODS:
Statistical analyses were performed according to following schedule:
- DUNNETT-test (two-sided): Food consumption, body weight, body weight change, corrected body weight gain, carcass weight, weight of unopened uterus, number of corpora lutea, number of implantations, number of resorptions, number of live fetuses, proportions of preimplantation loss, pro-portions of postimplanta-tion loss, proportions of resorptions, proportion of live fetuses in each litter, litter mean fetal body weight, litter mean placental weight
- FISHER'S EXACT test (one-sided): Female mortality, females pregnant at terminal sacrifice, number of litters with fetal findings
- WILCOXON-test (one-sided): Proportions of fetuses with malformations, variations and/or unclassified observations in each litter

HISTORICAL CONTROL DATA
The historical control data used for interpretation of findings refer to the same test facility, the same rat strain and supplier of the animals and cover a period of about 5 months (July 2001 - November 2001, 6 studies).

Test substance: purity: 99.1% (GC-method)

Conclusion: Based on the results of this prenatal developmental toxicity study, the no observed adverse effect level (NOAEL) for maternal and prenatal developmental toxicity is 200 mg/kg bw/day. Thus signs of prenatal developmental toxicity did only occur at a dose level, which was also clearly toxic to the dams. There were no indications for teratogenicity up to and including 1,000 mg/kg bw/day.

Reliability: (1) valid without restriction
Well conducted guideline study conducted under GLP

conditions. Chosen as key study for SIDS endpoint.
Flag: Critical study for SIDS endpoint
09-JUL-2003 (101)

5.8.3 Toxicity to Reproduction, Other Studies

5.9 Specific Investigations

5.10 Exposure Experience

Remark: Tested at 3% in petrolatum, it produced no irritation after a 48-hr closed-patch test on human subjects. A maximization test was carried out on 25 volunteers. The material was tested at a concentration of 3 % in petrolatum and produced no sensitization reactions.
Reliability: (2) valid with restrictions
basic data given, restrictions
Flag: Critical study for SIDS endpoint
06-SEP-2002 (102)

5.11 Additional Remarks

Type: Cytotoxicity

Remark: The cytotoxic effects of the substance were studied in 4 different in vitro test systems. The cytotoxic potential was scored in a system from 0 - 9.
Inhibition of cell growth in Ascites sarcoma BP8 cells: 9
Inhibition of oxidative metabolism in hamster brown fat cells: 3
Membrane damage of human diploid embryonic lung fibroblasts: 1
Inhibition of ciliar activity in embryonic trachea from chicken: 0
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (3) invalid
Unsuitable test system

08-JUL-2003 (103)

Type: Cytotoxicity

Remark: The incubation of chicken tracheal organ cultures with the test substance at 5 mM concentration for 60 min. did not lead to ciliostatic effects.
Test substance: 6-methyl-5-hepten-2-one, no data on purity mentioned
Reliability: (3) invalid
Unsuitable test system

14-APR-2003 (104)

Type: Cytotoxicity

Remark: Effect of tobacco smoke compounds on the plasma membrane of cultured human lung fibroblasts.

Result: This treatment ruptured the cell membranes leaving the nuclei intact. The results notes section indicates the percentage of nucleotide released. The nucleotide release for 6-methyl-5-hepten-2-one was 12%.
Test condition: The ability of the test material to increase the permeability of the membranes of human lung fibroblasts was studied by measuring the release of an intracellular nucleotide marker. Human diploid embryonic lung fibroblasts (line MRC-5) were cultivated to a cell density of 10 to the fifth cells/cm2. The cells were then labeled with [3H]uridine. The labelled cultures were incubated with 25 mM of the test material for 30 minutes at 37 C. 464 compounds were tested. Vehicle was Tris-buffered saline.
Reliability: (3) invalid
Significant methodological deficiencies

14-APR-2003 (105)

Type: Cytotoxicity

Remark: Title: Effects of tobacco and tobacco smoke constituents on cell multiplication in vitro.
Result: 0.1 mM: 9% inhibition, not statistically significant
1 mM: 96% inhibition
Test condition: Stem cell cultures, strain Ascites sarcoma BP8, originating from inoculated C3H mice were used to determine the toxicity of tobacco and tobacco smoke constituents. The stem cell cultures were grown in test tubes in Hams F10 medium sterilized by filtration,, wtih fetal calf serum (15w/w%), penicillin (100IU) and streptomycin (100IU) added. The test tubes were gassed with sterilized air containing 5% carbon dioxide and capped air-tight to maintain a stable pH of approximately 7.3. The cell cultures were reinoculated to a cell density of 0.1 X 10 4 cells/ml every 5th day. For the tests, the cell suspension was diluted with sterile medium to an initial cell density of 0.4 X 10 4 cells/ml. Test material was dissolved in 10 ul of ethanol, unless otherwise noted, and added to suspension. Tests were run in duplicate. All compounds were incubated at 37 degrees for 48 hours. 10 ul of solvents were added to the controls. The growth rate of an incubated cell culture was calculated and compared to the average value of 8-10 controls performed in each series. The doubling time for control cultures was approximately 24 hours. No systematic distinction was made between viable and total cell count. The effect of the tested compound is given as the ratio between the growth rates of the incubated cell culture and the controls, expressed as a percentage.
Reliability: (3) invalid
Significant methodological deficiencies

15-OCT-2002 (106)

Type: Cytotoxicity

Remark: Title: Effects of tobacco smoke compounds on the noradrenaline induced oxidative metabolism in isolated brown fat cells.
Result: 1mM: 39% inhibition
Test condition: The inhibition of noradrenaline induced respiration in isolated hamster brown fat cells was measured for 320

individual smoke components as an indication of effect on cell metabolism. The oxygen consumption rates of the cells were measured at 37 C using a Clark-type oxygen electrode fitted in a Perspex vessel of 1-ml volume. Test material was dissolved in ethanol or dimethyl sulfoxide and were incubated with the cells for exactly 5 minutes during which period the oxygen consumption was registered. After this preincubation, noradrenaline was added and the oxygen consumption of the cells was registered for another 5 minutes. The noradrenaline concentration was 1 uM which is approximately twice the dose known to induce maximal respiratory rate.

Reliability: (3) invalid

15-OCT-2002 (107)

Type: other: QSAR

Remark: Comparison of Tetrahymena and Pimephales toxicity based on mechanism of action.The toxicity data of 256 chemicals tested both in the 96 h Pimephales promelas mortality assay and the 2 d Tetrahymena pyriformis growth inhibition assay were evaluated using QSARs.

Reliability: (4) not assignable
Data from Handbook or collection of data

22-MAR-2004 (108)

Type: other: degeneration of olfactory cells

Remark: Rats were exposed to a variety of odorous compounds from 2 weeks of age for periods from 1-12 weeks.
6-Methyl-5-hepten-2-one was one of the 44 test substances. Each substance was introduced into an air stream from a glass bottle, the content was weighed before and after the experiments to measure the concentration of substance in the stream. The rats weighed between 28 - 39 g and were about 2 weeks old when placed in the cages. The animals were sacrificed at about 1, 2 and 3 months of age for microscopical examination of the olfactory tissue. For every 5 substances there was a group of control animals. For each of the 44 different odours a specific pattern of mitral cell degeneration was observed in the olfactory bulb. The pattern and extent of this degeneration did not appear to be correlated with the concentration of the odorant. The pattern of degeneration in coronal sections was maintained through the antero-posterior extent of the bulb in most cases. The results suggest a topological representation of different odours in the olfactory bulb.

Test substance: 6-methyl-5-hepten-2-one; no data on purity

Reliability: (3) invalid
Significant methodological deficiencies and insufficient documentation

09-MAR-2004 (109)

Type: other: intake via food

Remark: According to the RIFM-FEMA database (2002) the cumulated intake of 6-methylhept-5-en-2-one via various types of food

(alcoholic and non-alcoholic beverages, baked goods, chewing gum, frozen dairy, gelatin pudding, gravies, hard and soft candy) is about 0.54 mg per day (when added as a flavouring substance).

Reliability: (2) valid with restrictions

09-MAR-2004 (110)

6.1 Methods Handling and Storing

Fire/Exp. Prot.: Ensure thorough ventilation of stores and work areas.
Storage Req.: Keep tightly closed in a dry, cool and well-ventilated place.

Remark: Transport information

Land transport

ADR/RID Class: 3 Packaging group: III
Warning panel Hazard-no: 30 Substance no.: 1224
UN-No: 1224
Description of the goods: KETONE, FLUESSIG, N.A.G.
(2-METHYLHEPTEN-2-ON-6).

Inland waterway transport

ADN/ADNR Class: 3 Packaging group: III
Description of the goods: KETONE, FLUESSIG, N.A.G.
(2-METHYLHEPTEN-2-ON-6).

Sea transport

IMDG/GGVSee Class: 3 UN-No: 1224 PG: III
EMS: 3-07 MFAG: 300
Marine pollutant: no
Proper technical name: KETONES, LIQUID, N.O.S.

(2-METHYLHEPTEN-2-ON-6).

Air transport

ICAO/IATA Class: 3 UN/ID-No.: 1224 PG: III
Proper technical name: KETONES, LIQUID, N.O.S.
(2-METHYLHEPTEN-2-ON-6).

Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

Safe Handling: Worker exposure is limited by enclosed systems, industrial hygiene controls and personal protective measures (protective gloves, safety glasses with side-shields, respiratory protection if ventilation is inadequate).

Flag: non confidential, Critical study for SIDS endpoint
23-MAR-2004 (4)

6.2 Fire Guidance

Prot. Equipment: In case of fire wear a self contained breathing apparatus.
Ext. Medium: dry extinguishing media, foam
Unsuit. Ex. Med.: water
Add. Information: Fire debris must be disposed of in accordance with local regulations.

Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

6.3 Emergency Measures

Type: other: general advice

Remark: Remove contaminated clothing.
Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

Type: injury to persons (skin)

Remark: Wash with soap and water.
Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

Type: injury to persons (eye)

Remark: Wash affected eyes for at least 15 minutes under running water with eyelids held open.
Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

Type: injury to persons (oral)

Remark: Rinse mouth and then drink plenty of water.
Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

Type: injury to persons (inhalation)

Remark: keep patient calm, remove to fresh air
Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

Type: accidental spillage

Remark: Personal precautions: Ensure adequate ventilation.

Environmental precautions: Do not let product enter drains.

Methods for cleaning up: Large spillages should be dammed-off and pumped into containers; soak up remainder with absorbent material and dispose of in accordance with local regulations.
Flag: non confidential, Critical study for SIDS endpoint
12-NOV-2002 (2)

6.4 Possib. of Rendering Subst. Harmless

6.5 Waste Management

6.6 Side-effects Detection

6.7 Substance Registered as Dangerous for Ground Water

6.8 Reactivity Towards Container Material

(1) BASF AG, Fine Chemicals Product Database, status 22.07.2002

(2) BASF AG, Safety Data Sheet, METHYLHEPTENONE, 15.11.2001

(3) BASF AG, Operating Division Fine Chemicals, personal communication, 08.07.2003

(4) BASF AG, Operating Division Fine Chemicals, personal communication, 2003

(5) Danish Product Register, 2002, pers. communication, 2002

(6) Ullmann's Encyclopedia of Industrial Chemistry, Sixth Edition, 2000 Electronic Release, 2000 Wiley-VCH Verlag GmbH, Weinheim, Germany

(7) BASF AG, 2002b, internal data, pers. communications, October 2002

(8) Catalogue of Substances Hazardous to Water - Umweltbundesamt Berlin, status 13.03.2003

(9) National Chemical Inventories, 2001 Issue 2

(10) Beilstein, Handbook (online), BRN 1741705, BPR 110-93-0, update date 07 May 1999, printing date 03 July 2003

(11) Baglay A.K., Gurarly L.L. and G.G. Kuleshov, 1988, Physical properties of compounds used in vitamin synthesis, J. Chem. Eng. Data, 33, 512-518

(12) BASF AG, 1999, Substance data service, Dampfdruck von 2-Methyl-2-hepten-6-on, unpublished study, report No. 99.271, 06 April 1999

(13) BASF AG, 1972, Department of Technical Development and Technology, Dampfdruckmessungen, unpublished study, report No. 172.524.1, 29 Nov. 1972

(14) BASF AG, 1977, Analytical Laboratory, Dampfdruckmessung von 2-Methylbuten-2-ol-1 und 2-Methylhepten-2-on-6, unpublished study, BRU 77.8, 21 Jan. 1977

(15) BASF AG, 1989, Analytical Laboratory, Dampfdruck: 2-Methyl-2-hepten-6-on, unpublished study, BRU 89.117, 09 May 1989

(16) BASF AG, 1989, Analytical Laboratory, Bestimmung des Verteilungskoeffizienten log Pow der Prüfsubstanzen in 1-Octanol/Wasser bei 25°C, unpublished study, J. No. 106288/13, 08 May 1989

(17) BASF AG, 2002, Product Safety, data assessment, SRC KOWWIN 1.66, 16 July 2002

(18) BASF AG, 1989, Analytical Laboratory, Bestimmung des Verteilungskoeffizienten log Pow der Prüfsubstanz in 1-Octanol/Wasser bei 25°C, unpublished study, J. No.

106288/13, 03 May 1989

(19) BASF AG, 1989, Analytical Laboratory, Bestimmung der Löslichkeit der Prüfsubstanz in Wasser bei 25°C, unpublished study, 106288/13, 02 May 1989

(20) BASF AG, Department of Technical Safety, Sicherheitstechnische Kenndaten: 2-Methylheptenon, unpublished report, SIK-No. 76/1169

(21) BASF AG, Department of Technical Safety, Absence of explosive and oxidizing properties of 6-methylhept-5-en-2-one, internal communication, expert judgement, 11 Dec. 2000

(22) BASF AG, 1985, Analytical Laboratory, Physikalische Daten von Di-n-pentylether und 2-Methylhepten-2-on-6, unpublished study, BRU 85.54, 20 March 1985

(23) BASF AG, 1984, Department of Technical Development and Technology, Sicherheitstechnische Kenndaten: 2-Methylheptenon, unpublished study, report No. SIK 84/1222, 19 Dec. 1984

(24) Smith, A.M., Rigler, E., Kwok, E.S.C. and R. Atkinson, 1996, Kinetics and products of the gas-phase reactions of 6-Methyl-5-hepten-2-one and trans-cinnamaldehyde with OH and NO3 Radicals and O3 at 296+/-2K, Environ. Sci. Technol. 30, 1781-1785

(25) Grosjean E., Grosjean D. and J.H. Seinfield. 1996. Int. J. Chem. Kinet. 28., 373-382.

(26) BASF AG, 2003, Product Safety, SRC AOP v1.90, data assessment, 01 July 2003

(27) BASF AG, 2002, Product Safety, SRC AOP v1.90, data assessment, 16 July 2002

(28) BASF AG, Department of Product Safety, Expert Judgement, 26. Jan 2004

(29) Aramendia M.A., Borau V., Garcia I., Jimenez C., Lafont F., Marinas J.M. and F.J. Urbano, 1998, Determination of volatile organic compounds in drinking and waste water from Cordoba (Spain) by closed-loop stripping analysis in combination with gas chromatography coupled with mass spectrometry, Toxicol. Environ. Chem. 57, 9-25

(30) Richardson S.D., Thruston jr. A.D., Caughran T.V., Chen P.H., Collette T.W., Schenck K.M., Lykins jr. B.W., Rav-Acha C. and V. Glezer, 2000, Identification of new drinking water disinfection byproducts from ozone, chlorine dioxide, chloramine, and chlorine, Wat. Air Soil Pollut. 123, 95-102

(31) Monarca S., Richardson S.D., Feretti D., Grottolo M., Thruston jr. A.D., Zani C., Navazio G., Ragazzo P., Zerbini I. and A. Alberti, 2002, Mutagenicity and disinfection by-products in surface drinking water disinfected with

peracetic acid, Environ. Toxicol. Chem, 21(2), 309-318

(32) Jüttner F, 1988, Biochemistry of biogenic off-flavour compounds in surface waters, Wat. Sci. Tech. 20, 107-116

(33) König G., Brunda M., Puxbaum H., Hewitt C.N., Duckham S.C. and J. Rudolph, 1995, Relative contribution of oxygenated hydrocarbons to the total biogenic VOC emissions of selected mid-european agricultural and natural plant species, Atmos. Environ. 29, 861-874

(34) Sartin J.H., Halsall C.J., Davison B., Owen S. and C.N. Hewitt, 2001, Determination of biogenic volatile organic compounds (C8-C16) in the coastel atmosphere at Mace Head, Ireland, Anal. Chim. Acta 428, 61-72

(35) Jüttner F., 1992, Flavour compounds in weakly polluted rivers as a means to differentiate pollution sources, Wat. Sci. Tech. 25(2), 155-164

(36) Kool H.J., Krejil C.F. and B.C.J. Zoeteman, 1982, Toxicology assessment of organic compounds in drinking water, Crit. Rev. Env. Control 12, 307-357

(37) Guenther A., Zimmermann P., Klinger L., Greenberg J., Ennis C., Davis K. and W. Pollock, 1996, Estimates of regional natural volatile organic compound fluxes from enclosure and ambient measurements, J. Geophys. Res. 101, 1345-1359

(38) Puxbaum H., 1994, GC-MS-Analytik biogener VOC-Emissionen, Ö. Chem. Z. 220-226

(39) Muchalal M. and J. Crouzet, 1985, Volatile components of clove essential oil (Eugenia caryophyllus SPRENG): Neutral fraction, Agric. Biol. Chem. 49, 1583-1589

(40) Gomez E.J., Ledbetter C.A. and P.L. Hartsell, 1993, Volatile compounds in apricot, plum, and their interspecific hybrids, J. Agric. Food Chem. 41, 1669-1676

(41) Helmig D., Klinger L.F., Guenther A., Vierling L., Geron C. and P. Zimmermann, 1999, Biogenic volatile organic compound emissions (BVOCs) I. Identifications from three continental sites in the U.S., Chemosphere 38, 2163-2187

(42) Wilkins K., 1994, Volatile organic compounds from household waste, Chemosphere 29, 47-53

(43) Wilkins K. and K. Larsen, 1996, Volatile organic compounds from garden waste, Chemosphere 32, 2049-2055

(44) Mattheis J.P., Fellmann J.K., Chen P.M. and M.E. Patterson, 1991, Changes in headspace volatiles during physiological development of Bisbee Delicious apple fruit, J. Agric. Food Chem. 39, 1902-1906

(45) Jones G.J. and W. Korth, 1995, In situ production of volatile odour compounds by river and reservoir phytoplankton populations in Australia, Wat. Sci. Tech. 31,

145-151

(46) Hayes K.P. and M.D. Burch, 1989, Odorous compounds associated with algal blooms in south australian waters, Wat. Res. 23, 115-121

(47) Ross G.N., Fales H.M., Lloyd H.A., Jones T., Sokoloski E.A., Marshall-Batty K., and M.S. Blum, 2001, Novel chemistry of abdominal defensive glands of nymphalid butterfly Agraulis vanillae, J. Chem. Ecol. 27, 1219-1228

(48) Takeoka G.R., Flath R.A., Güntert M. and W. Jennings, 1988, Nectarine volatiles: Vacuum steam distillation versus headspace sampling, J. Agric. Food Chem. 36, 553-560

(49) Guadayol J.M., Caixach J., Ribe J., Cabanas J. and J. Rivera, 1997, Extraction, separation and identification of volatile organic compounds from paprika oleoresin (Spanish type), J. Agric. Food Chem. 45, 1868-1872

(50) Cotsaris E., Bruchet A., Mallevialle J. and D.B. Bursill, 1995, The identification of odorous metabolites produced from algal monocultures, Wat. Sci. Tech. 31, 251-258

(51) Shahidi F., Rubin L.J. and L. A. D'Souza, 1986, Meat flavor volatiles: A review of the composition, techniques of analysis, and sensory evaluation, CRC Crit. Rev. Food Sci. Nutrition 24, 141-243

(52) Yajima I., Yanai T., Nakamura M., Sakakibara, H. and K. Hayashi, 1984, Volatile flavor components of Kogyoku apples, Agric. Biol. Chem. 48, 849-855.

(53) Harper D.B., Nour A.A.M. and R.H. Thompson, 1986, Volatile flavour components of Dalieb (Borassus aethiopum L.), J. Sci. Food Agric. 37, 685-688

(54) Ciccioli,P., Brancaleoni, E., Frattoni, M., Cecinato, A. and A. Brachetti, 1993, Ubiquitous occurence of semi-volatile carbonyl compounds in tropospheric samples and their possible sources, Atmos. Environ. 27A, 1891-1901

(55) Völkl W., Hübner G. and K. Dettner, 1994, Interactions between Alloxysta brevis (Hymenoptera, Cynipoidea, Alloxystidae) and honeydew-collecting ants: How an aphid hyperparasitoid overcomes ant aggression by chemical defense, J. Chem Ecol. 20, 2901-2915

(56) BASF AG, 2002, Product Safety, data assessment, SRC PCKOCWIN v1.66, 16 July 2002

(57) TGD, 2003, Technical Guidance Document on Risk Assessment, European Chemicals Bureau, Chapter 4 "Use of (quantitative) structure activity relationshiipsa ((Q)SARS) in risk assessment"

(58) BASF AG, 2002, Product Safety, SRC HENRYWIN v3.10, data assessment, 16 July 2002

(59) Thomas, R.G., 1982, Volatilization from water, In: Lyman, W.J., Reehl, W.F. and D.H. Rosenblatt (eds.), Handbook of Chemical Property Estimation Methods. Environmental Behavior of Organic Compounds, pp 15-1 - 15-34, American Chemical Society, Washington DC, USA

(60) BASF AG, Product Safety, data assessment, Mackay Level I V2.11, 02 July 2003

(61) BASF AG, Department of Product Safety, Laboratory of Ecology, Prüfung der biologischen Abbaubarkeit von 2-Methylhepten-2-on-6 im manometrischen Respirationstest, unpublished study, 95/0224/26/2, 28.04.1995 - 26.05.1995

(62) BASF AG, 2002, Product Safety, SRC BCFWIN v2.14, data assessment, 16 July 2002

(63) BASF AG, 2004, Product Safety, SRC BCFWIN v2.14, data assessment, 26 Jan 2004

(64) Brooke L.T., Call D.J., Geiger D.L. and C.E Northcott, 1984, Acute toxicities of organic chemicals to fathead minnows (Pimephales promelas), Center for Lake Superior Environmental Studies, University of Wisconsin-Superior

(65) Cash G.G. and R.G. Clements, 1996, Comparison of structure-activity relationships derived from two methods for estimating octanol-water partition coefficients, SAR QSAR Environ Res 5, 113-124

(66) Protic M. and A. Sabljic, 1989, Quantitative sructure-activity relationships of acute toxicity of commercial chemicals on fathead minnows: effect of molecular size, Aquat. Toxicol. 14, 47-64

(67) Veith G.D., Call D.J. and L.T. Brooke, 1983, Estimating the acute toxicity of narcotic industrial chemicals to fathead minnows, ASTM STP 802, American Society for Testing and Materials, Philadelphia, pp 90-97

(68) Veith G.D., Call D.J. and L.T. Brooke, 1983, Structure toxicity relationships for the fathead minnow, Pimephales promelas: Narcotic industral chemicals, Can. J. Fish. Aquat. Sci. 40(6), 743-748

(69) BASF AG, 2003, Calculation of recovery rates of 100 mg and 200 mg 6-methylhept-5-en-2-one from its volatility in an algae, daphnia and fish test, data assessment, 07 July 2003

(70) BASF AG, 2003, Test on the volatility of 6-Methylhept-5-en-2-one (CAS: 110-93-0) in an algae, daphnia and fish test, unpublished data, 00/0874-2, 07 July 2003

(71) BASF AG, Department of Toxicology, Bericht über die Prüfung der akuten Toxiziät an der Goldorfe (Leuciscus idus L., Goldvariante), unpublished report, 10F0602/885325, 06 June 1989

(72) Applegate V.C., Howell J.H. and A.E. Hall, 1957, Toxicity of

4,346 chemicals to larval lampreys and fishes, US Department of Interior, Special Scientific Report - Fisheries No. 207.

(73) BASF AG, Department of Product Safety, Laboratory of Ecology, Akute Toxizität für Daphnien n. DIN 38412 L 11: Methylheptenon, unpublished report, 01/89/1480, 15 Jan 1990

(74) BASF AG, Department of Product Safety, Laboratory of Ecology, Hemmung der Algenzellvermehrung nach DIN 38412 L9: Methylheptenon, 01/89/1480, 15 Dec. 1989

(75) Tallarida, R.J. and L.S. Jacob, 1979, The dose-response relation in pharmacology, Springer, pp 98-103

(76) ToxRatPro, Version 2.07, Statistical evaluation of a algal growth inhibition test, unpublished calculation, by Barbara Priesmann (BUA AG Dresden), 26 March 2003

(77) Curtis C, Lima A., Lozano S.J. and G.D. Veith, 1982, Evaluation of a bacterial bioluminescence bioassay as a method for predicting acute toxicity of organic chemicals to fish, ASTM STP 766, American for Testing and Materials, Philadelphia, pp 170-178

(78) BASF AG, Department of Product Safety, Laboratory of Ecology, Bacterial oxygen consumption test: 6-methylhept-5-en-2-one, unpublished summary-reprint of a test report, 10/0905/88, 11 Aug. 1988

(79) BASF AG, Department of Product Safety, Laboratory of Ecology, Prüfung der Atmungshemmung von Belebtschlamm durch 2-Methylhepten-2-on-6 im Kurzzeitatmungshemmtest, unpublished report, 95/0224/08/1, 08 May 1995

(80) Reichardt W., 1981, Influence of methylheptenone and related phytoplankton norcarotinoids on heterotrophic aquatic bacteria, Can. J. Microbiol. 27, 144-147

(81) B. Nandi, 1977, Effect of some volatile aldehydes, ketones, esters and terpenoids on growth and development of fungi associated with wheat grains in the field and in storage, J. Plant Diseas. Protect. 84, 114-128

(82) Schafer E.W., Bowles W.A.Jr. and J. Hurlbut, 1983, The acute oral toxicity, repellency, and hazard potential of 998 chemicals to one or more species of wild and domestic birds, Arch. Environ. Contam. Toxicol. 12, 355-382

(83) Wolken, W.A.M. and M.J. van der Werf, 2001, Geraniol biotransformation-pathway in spores of Penicillium digitatum, Appl. Microbiol. Biotechnol. 57, 731-737

(84) Demyttenaere, J.C.R. and L. De Pooter, 1996, Biotransformation of geraniol and nerol by spores of Penicillium italicum, Phytochemistry 41, 1079-1082

(85) Demyttenaere, J. and N. De Kimpe, 2001, Biotransformation of terpenes by fungi - Study of the pathways involved, J. Mol. Catal. 11, 265-270

(86) Eldred, D.V. et al., Prediction of fathead minnow acute toxicity of organic compounds from molecular structure, Chem. Res. Toxicol., 12, 670-678, 1999

(87) BASF AG, Department of Product Safety, 22 March 2004

(88) BASF AG, Department of Toxicology, Ergebnis der gewerbetoxikologischen Vorprüfung, unpublished report (XXIII/401), 04 Sept. 1974

(89) Keating, J.W., Report to RIFM (1972) cited in Opdyke, Fd. Cosm. Toxicol., 13, 859, 1975

(90) Keating, J.W., Report to RIFM, 1972 cited in RIFM-FEMA database, Oct. 2002

(91) Compadre, C.M., Hussain, R.A., de Compadre, R.L.L., Pezzuto, J.M. and A.D. Kinghorn, 1987, The intensely sweet sesquiterpene hernandulcin: Isolation, synthesis, characterization, and preliminary safety evaluation, J. Agric. Food Chem., 35, 273-279, 1987

(92) Gigiena Truda i Professional'nye Zabolevaniya. Labor Hygiene and Occupational Diseases, 32 (88), 52-53, 1988 cited in RTECS, 2000/01

(93) Migukina, N.B., Lipanina, T.B., Maltzewa, N.M., Stepkina, L.N., Schumarina, N.B., Karpuchina, E.A. and C.B. Kajutina, 1988, To the validation of tentative safe level of methylheptenon exposure in the work zone air, Gigiena Truda i Professional'nye Zabolevaniya. Labor Hygiene and Occupational Diseases, 32 (88), 52-53, 1988

(94) Hoffmann-LaRoche, Acute toxicity, eye and skin irritation test of fragrance materials, unpublished results, 1967 cited in RIFM-FEMA data base, Oct. 2002

(95) Klecak, G., The Freund's complete adjuvant test and the Open Epicutaneous Test, Curr. Probl. Derm., vol. 14, 152-171, Karger, Basel, 1985

(96) Sharp, D.W., The sensitization potential of some perfume ingredients tested using a modified Draize procedure, Toxicology, 9, 262-271, 1978

(97) BASF AG, Product Safety, 6-Methylhept-5-en-2-one (Methylheptenon) Subchronic toxicity study in Wistar rats, administration by gavage for 3 months, unpublished report, project no. 51S0874/00114, 00/874, 05 Dec. 2002

(98) Florin, I., Rutberg, L., Curvall, M. and C.R. Enzell, 1979, Screening of tobacco smoke constituents for mutagenicity using the Ames' Test, Toxicology, 15, 219-232, 1979

(99) BASF AG, Product Safety, Report on Salmonella typhimurium / Escherichia coli reverse mutation assay (standard plate test and preincubation test) with 6-methylhept-5-en-2-one (methylheptenon), unpublished report, project no.

40M0874/004193, 00/874, 18 April 2002

(100) BASF AG, Product Safety, Cytogenetic study in vivo with 6-methylhept-5-en-2-one (methylheptenon) in the mouse micronucleus test after two intraperitoneal administrations, unpublished report, project no. 26M0874/004151, 00/0874-1, 10 July 2001

(101) BASF AG, Product Safety, 6-Methylhept-5-en-2-one (methylheptenon) - Prenatal developmental toxicity study in Wistar rats, oral administration (gavage), unpublished report, project no. 30R0874/00111, 00/874, 29 Oct. 2002

(102) Kligman, A., M.; Report to RIFM, 19 July, (1972) in: RIFM Monograph 13:859

(103) Curvall, M., Enzell, C.R. and B. Pettersson, An evaluation of the utility of four in vitro short term tests for predicting the cytotoxicity of individual compounds derived from tobacco smoke, Cell Biology and Toxicology, 1, 173-193, 1984

(104) Petersson, B. et al., Effects of tobacco smoke compounds on the ciliary activity of the embryo chicken trachea in vitro, Toxicology, 23, 41-55, 1982

(105) Thelestam, M. et al., Effect of tobacco smoke compounds on the plasma membrane of cultured human lung fibroblasts, Toxicology, 15, 203-217, 1980

(106) Pilotti, A. et al., Effects of tobacco and tobacco smoke constituents on cell multiplication in vitro, Toxicology, 5, 49-62, 1975

(107) Petterson, B. et al., Effects of tobacco smoke compounds on the noradrenaline induced oxidative metabolism in isolated brown fat cells, Toxicology, 18, 1-15, 1980

(108) Bearden, A.P. and Schultz T.W., Comparison of tetrahymena and pimephales toxicity based on mechanism of action. SAR and QSAR in Environmental Research, 9, 127-153, 1998

(109) Pinching, J. and Doving, K.B., Selective degeneration in the rat olfactory bulb following exposure to different odours, Brain Research, 82, 195-204, 1974

(110) RIFM (Research Institute for Fragrance Materials, USA) - FEMA (Flavor and Extract manufacturers' Association of the US) database, Oct. 2002