



Recommendation from the Scientific Committee on Occupational Exposure Limits for cyclohexane

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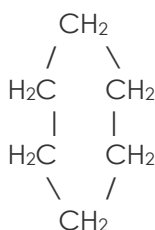


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8 hour TWA	:	200 ppm (700 mg/m ³)
STEL (15 mins)	:	-
Additional classification	:	-

Substance identification:

Cyclohexane



Synonyms	:	Hexahydrobenzene, Hexamethylene
EINECS N°	:	203-806-2
EEC N°	:	601-017-00-1
EU Classification	:	F; R11
CAS N°	:	110-82-7
MWt	:	84.18
Conversion factor (20°C, 101kPa)	:	3.50 mg/m ³ = 1 ppm



1 Occurrence/use

Cyclohexane is a colourless liquid with a melting point of 6.5°C and a boiling point of 80.7°C. It is highly flammable and has a pungent odour similar to that of petrol, with an odour threshold of approximately 50 ppm (175 mg/m³). The vapour pressure is 12.7 kPa at 20°C.

Cyclohexane occurs naturally in all crude oils in concentrations of 0.1 – 1.0%. It is manufactured in closed system by hydrogenation of benzene. Current EEC capacity for cyclohexane production is 835,000-925,000 tonnes per annum. The majority of cyclohexane is used for production of nylon, with lesser amounts used as a solvent and as a chemical intermediate. Occupational exposure to cyclohexane is in combination with other solvents. A mixture of solvents including n-hexane and cyclohexane, known as “commercial hexane”, is widely used as a solvent in shoe factories. Exposure levels of up to 360 ppm (1260 mg/m³) have been measured. Analysis is by gas chromatography or Drager tube and it should be noted that other solvents may interfere.

2 Health Significance

Data on the effects of pure cyclohexane are limited. Studies on “commercial hexane” are not adequate for setting an OEL for cyclohexane, as the observed effects may be due to n-hexane.

Cyclohexane is well absorbed in animal and humans by the lungs and readily exhaled in breath of all species studied. Metabolism involves mostly hydroxylated derivatives which are excreted after glucuronide conjugation in the urine. Urinary metabolites include cyclohexanol and cyclohexanone (ACGIH 1997).

A recent study in volunteers after 8 h periods of inhalation exposure at a concentration of 1010 mg/m³ (290 ppm) showed that 1,2 and 1,4 cyclohexandiol were the major metabolites, accounting for 23,4% and 11,3% respectively for the dose (Mraz et al. 1998).

Cyclohexane is of low acute toxicity by oral, inhalation or dermal routes. A short term inhalation (1 hour) concentration in rabbits of 29,190 ppm was lethal to all of the animals; for mice, the lowest lethal concentration exceeded 17,460 ppm (2 hour exposure). The oral LD₅₀ was found to be 6200 to 30,000 mg/kg body for the rat (DECOS 1990).

Mild skin irritation was observed in rabbits after single dermal doses (Jacobs and Martens 1987). After repeated application, the substance is significantly more irritating to the skin (Treon et al. 1943a). Cyclohexane can cause defatting and can accelerate the skin penetration rate of other agents (Greim 1996).

Cyclohexane was not found to be a skin sensitiser when tested by the modified Buhler method (Moore 1996).

In a recent study rats and mice were exposed to 0, 500, 2000 or 7000 ppm of cyclohexane vapour for 6/hr/day, 5 days/week for 90 days. Subgroups of rats and mice were further observed during a one month recovery period. During exposure to 2000 or 7000 ppm rats and mice had a diminished response or an absent response of a delivery of punctuate auditory alerting stimulus. Sedation occurred in rats exposed to 2000 ppm and 7000 ppm. Mice exposed at 2000 ppm exhibited sedation, while at 7000 ppm mice exhibited excitation behaviour. Mean relative liver weights for 7000 ppm rats and mice were significantly higher than their respective controls. However only male rats exposed to 7000



ppm had microscopic evidence of changes, comprising the adaptive response of hepatocellular hypertrophy. Male and female mice exposed to 7000 ppm showed increases in red blood cells, haemoglobin and hematocrit (Maley et al. in press). In this study a NOAEL of 500 ppm was established for mice and rats, with signs of nervous system suppression being seen at higher exposures. Notably there were no indications of pathological damage in the liver or kidneys (compare with Treon et al study below).

In earlier studies, exposure of rabbits to cyclohexane at 757 ppm (2650 mg/m³), for 6 h/day, 5 days/week, for 10 weeks, resulted in "barely demonstrable" microscopic changes in the liver and kidney (Treon et al., 1943b). Higher exposure levels, from 7170 ppm (25.1 g/m³), resulted in depression of the CNS. In another study a NOAEL of 2500 ppm (8750 mg/m³) in the rat for neurotoxic effects was established (Frontali et al 1981). In this study inhalation exposure to 1500 or 2500 ppm for 30 weeks did not induce pathological alterations of nerve tissue in rats, a characteristic effect of n-hexane exposure.

Cyclohexane was found not to be genotoxic either with or without a metabolic activation system in the Salmonella microbial assay or in in vitro UDS test with human lymphocytes (DECOS 1990). Negative results were also obtained in a mouse lymphoma test (TK locus) with and without metabolic activation (Greim 1996). Cyclohexane in a concentration of 0.5% in the diet of *Drosophila melanogaster* did not induce either autosomal recessive lethal mutations or sex-linked recessive lethal mutations (DECOS 1990). No structural chromosomal aberrations were found in male or female rats exposed to 97, 307 or 1042 ppm, 6 hours daily for 5 days (Greim 1996).

No standard studies of the carcinogenicity of cyclohexane have been performed, but there is no evidence to suggest that this substance has significant carcinogenic potential. Cyclohexane was a weak promoter of skin tumours initiated by carcinogenic polycyclic aromatic hydrocarbons (PAHs) in a mouse skin bioassays (Gupta and Mehrotra 1990). It is likely that the irritant effects of cyclohexane explain its promoting activity.

In a two-generation inhalation reproductive study rats were exposed throughout the study to 0, 500, 2000 or 7000 ppm cyclohexane. No adverse compound-related effects were observed at 500 ppm. At 2000 ppm the only adverse effect was diminished or no alerting response. At 7000 ppm reductions in mean body weight for P1 and F1 females and F1 males as well as reduced mean pup weight for F1 and F2 litters were observed (Kreckmann et al 1998a).

Assumed-pregnant rats were exposed to 0, 500, 2000 and 7000 ppm cyclohexane on days 7 to 16 of gestation. Dams of the 7000 ppm exposure group showed reductions in maternal body weight gain and diminished or absent alerting response. No other compound-related effect was observed.

Assumed-pregnant rabbits were exposed to 0, 500, 2000 or 7000 ppm cyclohexane on days 6 to 18 of gestation. The animals were sacrificed on day 29. No maternal or developmental toxicity were observed in this study (Kreckmann et al. 1998b). But it was shown for the metabolite cyclohexanol that oral administration per gavage of 25 mg/kg body weight, daily for 40 days causes in rabbits spermatotoxic effects (Dixit et al. 1980). In mice spermatotoxic effects were seen after subcutaneous injection of 15 mg/kg body weight (Tyagi et al. 1979).

In contrast no adverse effects on fertility of male rats were detected after inhalative exposure to cyclohexanone, another metabolite of cyclohexane, for several months, 6 hours daily, in concentration of 500 ppm. No histological alteration in the sexual organ were seen after 4 months' exposure to a cyclohexanone concentration of 1000 ppm. At



this concentration the rats were exposed to cyclohexanol concentrations of at least 120-180 mg/kg body weight (Greim 1996). It is probable that application by bolus or i.p. produces peak concentrations that cannot be achieved in the case of inhalation, and that consequently no spermatotoxic effects were seen after inhalation.

In humans, an experimental study has been reported in which 12 healthy male volunteers were exposed using a double blind, two-way cross-over design to 250 ppm for 4 hours. As control condition an exposure of the same subjects to 25 ppm was established. The two test conditions were spaced 7 days apart. Cognitive functioning was assessed using selected tests from the Neurobehavioural Evaluation System (NES). In addition a computer-administered questionnaire designed to assess changes in mood and affect was also included. Measurements were normally carried out before, during (twice) and following exposure. There were no significant effects of inhalatory exposure to 250 ppm cyclohexane on any of the 20 variables measured. An analysis of self-reported symptoms showed that 7 out of 12 volunteers reported headache at 250 ppm cyclohexane, compared with one person out of 12 at 25 ppm (Hoogendijk and Emmen 1998). It cannot be ruled out that the persons that reported having headache may have felt slightly unwell. But this finding can not be regarded as a consistently and significantly adverse effect.

Exposure to cyclohexane (geometric mean 27 ppm and highest concentration 274 ppm; reference period was not given, but supposedly it is an 8h TWA) did not induce in the exposed workers any significant rise in the prevalence of subjective symptoms or in haematological and serum biochemical parameters of liver and kidney functions (Yasugi et al. 1994).

A study on 18 workers exposed to a glue containing 75.6% cyclohexane, 12% toluene and 0.9% n-hexane showed that concentrations of airborne cyclohexane ranging from 5 to 211 ppm (reference period was not given but presumably it is an 8h TWA) did not have any adverse effects on the peripheral nervous system. No differences were found in nerve conduction velocities between workers exposed to cyclohexane and age and sex matched controls (Yuasa et al. 1996).

3 Recommendation

The data on behavioural effects of cyclohexane in human volunteers suggest that headache can occur at 250 ppm. Exposure of 4 hours to 250 ppm cyclohexane caused headache complaints in 7 out of the 12 exposed volunteers. This finding however is not regarded as a consistently and significantly adverse effect, in fact on the same volunteers no significant effect of exposure was found on cognitive performance. (Hoogendijk and Emmen 1998). In addition animal data established a NOAEL of 500ppm and a LOAEL of 2000 ppm for narcotic effects in rats and mice (Malley et al. in press). The human data suggest that the NOAEL is about 250 ppm, taking this into account a recommended 8 hour TWA of 200 ppm can be established.



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