1999:26

Scientific Basis for Swedish Occupational Standards XX

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ARBETE OCH HÄLSA VETENSKAPLIG SKRIFTSERIE
ISBN 91–7045–545–7 ISSN 0346–7821 http://www.niwl.se/ah/

Arbetslivsinstitutet
National Institute for Working Life

National Institute for Working Life

The National Institute for Working Life is Sweden's national centre for work life research, development and training.

The labour market, occupational safety and health, and work organisation are our main fields of activity. The creation and use of knowledge through learning, information and documentation are important to the Institute, as is international co-operation. The Institute is collaborating with interested parties in various development projects.

The areas in which the Institute is active include:

- labour market and labour law,
- work organisation,
- musculoskeletal disorders,
- chemical substances and allergens, noise and electromagnetic fields,
- the psychosocial problems and strain-related disorders in modern working life.

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http://www.niwl.se/ah/
Printed at CM Gruppen

Preface

The Criteria Group of the Swedish National Institute for Working Life (NIWL) has the task of gathering and evaluating data which can be used as a scientific basis for the proposal of occupational exposure limits given by the National Board of Occupational Safety and Health (NBOSH). In most cases a scientific basis is written on request from the NBOSH. The Criteria Group shall not propose a numerical occupational exposure limit value but, as far as possible, give a dose-response/dose-effect relationship and the critical effect of occupational exposure.

In searching of the literature several data bases are used, such as RTECS, Toxline, Medline, Cancerlit, Nioshtic and Riskline. Also information in existing criteria documents is used, e.g. documents from WHO, EU, US NIOSH, the Dutch Expert Committee for Occupational Standards (DECOS) and the Nordic Expert Group. In some cases criteria documents are produced within the Criteria Group, often in collaboration with DECOS or US NIOSH.

Evaluations are made of all relevant published original papers found in the searches. In some cases information from handbooks and reports from e.g. US NIOSH and US EPA is used. A draft consensus report is written by the secretariat or by a scientist appointed by the secretariat. The author of the draft is indicated under Contents. A qualified evaluation is made of the information in the references. In some cases the information can be omitted if some criteria are not fulfilled. In some cases such information is included in the report but with a comment why the data are not included in the evaluation. After discussion in the Criteria Group the drafts are approved and accepted as a consensus report from the group. They are sent to NBOSH.

This is the 20th volume which is published and it contains consensus reports approved by the Criteria Group during the period July 1998 to June 1999. Previously published consensus reports are listed in the Appendix (p 111).

Johan Högberg
Chairman

Johan Montelius
Secretary

The Criteria Group has the following membership (as of June, 1999)

Olav Axelson		Dept Environ Occup Medicine University Hospital, Linköping
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Arne Wennberg		International Secretariate NIWL
Olof Vesterberg		Respiratory health and Climate, NIWL

Contents

Consensus Report for Cyanamide

September 30, 1998

Physical and chemical data. Uses

CAS No.:	420-04-2
Synonyms:	amidocyanogen, carbimide, hydrogen cyanamide, carbodiimide
Formula:	CH_2N_2
Structure:	$H_2NC{=}N$
Molecular weight:	42.04
Melting point:	45 – 46 °C
Boiling point:	127 °C
Density:	1.28 g/ml
Flash point:	141 °C
Conversion factors:	1 ppm = 1.72 mg/m^3
	1 mg/m^3 = 0.58 ppm

Cyanamide at room temperature is a crystalline substance that absorbs moisture from the air and forms a damp solid or a solution. No odor threshold has been reported. Cyanamide is soluble in water (78 g/100 ml), alcohol and ether, but its solubility in benzene is low.

Cyanamide is used in chemical syntheses, in fertilizers, and as a biocide. It is also used in ore refining and in the wood processing and rubber industries. Cyanamide can also be formed by hydrolysis of calcium cyanamide, a substance which has similar uses. Cyanamide and its salts have also been used medicinally in treatment of alcoholics, since cyanamide inhibits aldehyde dehydrogenase. It is no longer registered as a medicine. No information on air concentrations was found in the literature.

Uptake, biotransformation, excretion

Cyanamide can be absorbed via the digestive tract and skin. It is metabolized to acetyl cyanamide, mostly in the liver, by acetyl-S-CoA-dependent N-acetyl-transferase (26). It may also be metabolized in a reaction induced by catalase (8). In one study, six volunteers were given cyanamide orally (0.25 mg/kg body weight): 40% of the dose was excreted in urine as acetylcyanamide during the next 48 hours, most of this within the first 12 hours. A 1-ml dose of a 1% cyanamide solution (0.25 mg/kg) was applied to skin (4 x 4 cm) and left for 6 hours: 7.7% of the dose was excreted in urine as acetylcyanamide (17).

Most cyanamide is excreted in urine. When animals were given ^{14}C-labeled cyanamide (8 mg/kg intraperitoneally for rats, 1.6 mg/kg intravenously and orally for dogs and rabbits) nearly all the radioactivity was detected in urine, and the primary metabolite was found to be N-acetylcyanamide (11, 26). In a study with rats and dogs, the highest plasma concentration was reached in the rats 30 minutes after oral administration of 4 mg/kg. After intravenous administration of 1.4 mg/kg, the half time in plasma was 30 to 61 minutes for both species (21). Indications of non-linear metabolism have been observed in rats under steady-state conditions (0.005 – 32 mg/kg i.p. at 45-minute intervals). Clearance and first passage metabolism were not constant between the doses, probably because the biotransformation capacity of the liver had been saturated (23).

Toxic effects

Human data

The effects of cyanamide on the liver have been studied in conjunction with cyanamide treatment of alcoholics. One study describes liver biopsies from 37 patients who had been treated with cyanamide for from 2 months to 7 years, with daily doses ranging from 45 to 180 mg. In addition to the liver changes seen with alcoholism, there were structural changes such as fibrosis and changes in connective tissue in all biopsies, as well as a particular type of inclusions in hepatocytes (Lafora-like inclusion bodies consisting of lipid vesicles, glycogen and traces of degenerated organelles) (19). Elevated blood levels of liver enzymes (ALAT, ASAT) induced by cyanamide were also detected in this study. The histological picture resembled that of cirrhosis, and the authors concluded that the longer the treatment, the greater the changes. Other studies have also shown that cyanamide used to treat alcoholism induces inclusion bodies in hepatic cells (2, 19, 30, 31, 32).

Two cases of skin sensitization from occupational contact with cyanamide have been described (5, 6). One man who was sensitized by working for 1.5 years with medicines that contained cyanamide had a positive reaction to the substance in a patch test (0.1% in water; 48 hours) (6). Sensitization to cyanamide was reported to be rare. The other case report describes sensitization in a chemist whose work included some contact with cyanamide. He had a positive reaction to a 0.01% cyanamide solution in a patch test (5). Seven cases of cyanamide-induced skin eruptions are reported in a study from 1977. The patients had been treated with oral doses of a 1% cyanamide solution, 7 ml daily for from 1 to 4 months (14). After 10 days to 3 months of the treatment, all of them had skin disorders: 6 had scaling dermatitis and one had lichen planus-like eruptions. The authors concluded that this type of reaction may have been a common but ignored problem. One case report describes granulocytopenia and skin sensitization in a man who was treated with 100 mg cyanamide during a 3-week period (1). The symptoms disappeared after the treatment was broken off.

There is a study on endocrine function (thyroidea, testes) in 21 persons who worked in a calcium cyanamide production plant and 9 controls (18). One of the reasons it was undertaken was that testicular atrophy had been observed in male rats experimentally exposed to cyanamide (28). N-Acetylcyanamide content in urine at the end of the workday was used as a measure of exposure, and clearly showed exposure in the 21 workers. There were no observed differences in endocrine function (testosterone, follicle-stimulating hormone, luteinizing hormone) between exposed persons and controls.

Animal data

The LD_{50} for rats has been calculated to be 125 mg/kg body weight for oral administration (3) and 56 mg/kg for intravenous administration (13). Cyanamide causes skin irritation (13) and severe eye irritation in rabbits (100 mg dropped in the eye) (7).

In several in vivo studies, cyanamide has been shown to inhibit alcohol dehydrogenase activity. Cyanamide treatment (2 mg/kg, i.p. 1 hour) of rats inhibited alcohol dehydrogenase and increased the toxicity of alcohol (24). Intraperitoneal doses of 0.35 mg/kg repeated at 45-minute intervals suppressed alcohol dehydrogenase activity completely (23). Elevated acetaldehyde levels following alcohol exposure were seen in rats pre-treated with cyanamide (0.7 mg/kg, p. o.) 45 minutes before the exposure. Elevated alcohol levels in blood have also been related to pre-treatment with cyanamide (10 mg/kg, p.o.) (12, 25).

Reduced body weight and elevated levels of monoamines in the brain were reported in rats after oral or intravenous administration of cyanamide (8 mg/kg, 20 weeks or more) (22). Catalase activity in various organs of rats has been shown to diminish at dose levels greater than 1.3 mg/kg (i.p., maximum after 1 hour) (9). Doses exceeding 10 mg/kg (i.p., 4 hours) increased the level of circulating ketone bodies in rats (10).

Mutagenicity

In a study with Salmonella typhimurium (strains TA98, TA100, TA1535, TA1537, TA1538) and E coli, cyanamide caused no increase in mutation frequency, either with or without metabolic activation (4). Cyanamide was not clastogenic in a micronucleus test with mice (16). Elevated frequencies of mitotic gene conversion and non-disjunction were seen in Aspergillus nidulans (29). No increase of DNA string breaks was seen in hepatocytes exposed to cyanamide in vitro (27).

Carcinogenicity

Cancer incidence and mortality were mapped in a cohort of 790 workers in a plant producing calcium carbide. No increase in cancer was seen among the 117 workers who had worked with cyanamide/dicyandiamide production for at least 18 months during 1953 – 1970 (15). Exposures are not reported. The National Cancer Institute in the United States has tested calcium cyanamide (which hydrolyzes to cyanamide)

for carcinogenic effect in a two-year study (20) with rats and mice. No carcinogenic effect was observed.

Teratogenicity

In a two-generation reproduction/fertility study, male rats were given cyanamide in oral doses of 2 to 25 mg/kg daily for 70 days before mating, and females for 15 days prior to or during gestation. The females in the highest dose group (25 mg/kg) had lower body weights, fewer corpora lutea, fewer implanted embryos and smaller litters. Males in the highest dose group had bilateral testicular atrophy and lower fertility. There were no observed effects on the F_1 generation. The NOEL in this study was 7 mg/kg (28).

Table 1. Effects of cyanamide on experimental animals.

Exposure mg/kg	Time	Species	Effect	Ref.
125 p.o.		Rat	LD_{50}	3
56 i.v.		Rat	LD_{50}	13
25 p.o.	daily for 70 days (prior to mating)	Rat (males)	Testicular atrophy, reduced fertility	28
25 p.o.	daily for 15 days (before or during gestation)	Rat (females)	Lower body weight, fewer corpora lutea and implanted embryos, smaller litters	28
10 i.p.	4 hours	Rat	Elevated levels of ketone bodies	10
8 p.o.	20 weeks	Rat	Lower body weight, elevated levels of monoamines in brain	22
2 i.p.	1 hour	Rat	Inhibited alcohol dehydrogenase activity, increased alcohol toxicity	24
1.3 i.p.	1 hour	Rat	Inhibited catalase activity	9
0.7 p.o.	45 minutes	Rat	Elevated acetaldehyde levels	12, 25
0.35 p.o.	45-minute intervals	Rat	Suppressed alcohol dehydrogenase activity	23

(p.o = oral; i.v. = intravenous; i.p. = intraperitoneal)

Dose-effect/dose-response relationships

There are no data on which to base a dose-effect or dose-response relationship for occupational exposure to cyanamide. The dose-response and dose-effect relationships observed in animal experiments are summarized in Table 1.

Conclusions

Cyanamide inhibits alcohol dehydrogenase when used medicinally. There are no data on which to base a critical effect for occupational exposure. Cyanamide can be skin sensitizing to humans and has been shown to irritate the eyes of rabbits.

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Consensus Report for Phosphorus Chlorides

September 30, 1998

This report treats phosphorus trichloride, phosphorus pentachloride and phosphoryl chloride.

Chemical and physical data. Uses

phosphorus trichloride

CAS No:	7719-12-2
Synonyms:	phosphorus(III)chloride, trichlorophosphine
Formula:	PCl_3
Molecular weight:	137.33
Boiling point:	76 °C
Melting point:	- 112°C
Vapor pressure:	12.7 kPa (20°C)
Conversion factors:	1 ppm = 5.70 mg/m^3 (20°C) 1 mg/m^3 = 0.175 ppm (20°C)

phosphorus pentachloride

CAS No.:	10026-13-8
Synonyms:	phosphorus(V)perchloride, pentachlorophosphorane
Formula:	PCl_5
Molecular weight:	208.24
Boiling point:	sublimates at 160°C *
Melting point:	167 °C (three-phase equilibrium) *
Conversion factors:	1 ppm = 8.64 mg/m^3 (20°C) 1 mg/m^3 = 0.116 ppm (20°C)

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* From Reference 14. Other sources give other boiling and melting points.

phosphoryl chloride

CAS No:	10025-87-3
Synonyms:	phosphoryl trichloride, trichlorophosphine oxide, phosphorus oxychloride, phosphorus oxytrichloride
Formula:	$POCl_3$
Molecular weight:	153.33
Boiling point:	105.5°C
Melting point:	1 °C
Vapor pressure:	3.6 kPa (20°C)
Conversion factors:	1 ppm = 6.36 mg/m^3 (20°C) 1 mg/m^3 = 0.157 ppm (20°C)

Phosphorus trichloride at room temperature is a clear liquid that steams in damp air: it hydrolyzes, emitting heat, to phosphorous acid and hydrochloric acid. Phosphorus pentachloride at room temperature is a steaming, yellowish or white to greenish-white solid. It hydrolyzes initially to hydrochloric acid and phosphoryl chloride, and in a second step the phosphoryl chloride (a clear, steaming liquid) hydrolyzes, producing heat, phosphoric acid and more hydrochloric acid. Phosphorus trichloride, phosphorus pentachloride and phosphoryl chloride all three have a sharp, penetrating odor (1, 2, 3, 12, 13, 17, 20).

Phosphorus trichloride is used mostly as an intermediate in the production of pesticides, surfactants, softeners, gasoline additives and pigments. Phosphorus pentachloride is used as a thickener. Phosphoryl chloride is used in the production of softeners and gasoline additives and also in the production of hydraulic fluids and fire retardants. All three substances are used as chlorinators and catalysts (1, 2, 3).

Uptake, biotransformation, excretion

No information was found in the literature.

Toxic effects

Human data

Vapor/dust (including hydrolysis products) of all three substances are irritating/corrosive to eyes and respiratory passages, but information on exposure levels is usually not given (5, 10, 11, 19, 22, 26, 27, 29). Phosphorus trichloride and phosphorus pentachloride are reported to be strongly irritating to mucous membranes (4, 29). Phosphoryl chloride has been reported to strongly affect both the upper and lower respiratory passages and to be more likely to have delayed effects on respiratory passages than phosphorus trichloride (10, 26, 29). Exposure to phosphorus trichloride and its hydrolysis products has also resulted in skin irritation (17, 27). Occupational exposure to phosphorus chlorides has been reported to etch the teeth (21), but the type of exposure was not described.

Effects other than local irritation/ulceration have also been reported to result from exposure to phosphorus chlorides. One study (27) reports nausea, vomiting, headache and transient elevation of lactate dehydrogenase levels in serum in several persons acutely exposed to phosphorus trichloride and its hydrolysis products. Another study (5) reports dizziness and severe headache in a person who had inhaled phosphorus pentachloride vapor for a few seconds. Brief exposure (in some cases only a few seconds) to phosphoryl chloride vapor has caused dizziness, nausea, vomiting and effects on the heart (5, 10, 11). In a few cases, enlarged liver, albuminuria and anemia have also been reported after exposure to phosphoryl chloride vapor (22), but it is not clear whether these effects were the result of the exposure.

Only a few studies report both exposure levels and the symptoms of exposed persons. One study (Dadej, 1962; reviewed in Reference 17) describes effects on some people who were exposed to phosphorus trichloride and its hydrolysis

products by an explosion. Three workers who were exposed for from a few seconds up to half a minute or so, and who died within 24 hours, had severe skin burns, ulcerated eyes, inflamed bronchi and pulmonary edema. Ulcerated eyes, respiratory passages and skin were also seen in one surviving worker who was exposed for several seconds. Concentrations during the first 120 seconds were roughly estimated to have been about 36,800 mg/m^3 phosphorus trichloride, 116,300 mg/m^3 hydrochloric acid, and 62,500 mg/m^3 phosphorous acid.

A report from NIOSH (25) states that, of 37 workers exposed to phosphorus trichloride and phosphoryl chloride, about 65% (24/37) suffered acute respiratory symptoms such as breathing difficulty or chest tightness at least once a month. Only 5% (1/22) of non-exposed persons reported these symptoms. Lung function tests, however, revealed no significant differences between the two groups. Air concentrations were measured with personal monitors for two days, and were below the detection limits for phosphorus trichloride and phosphoryl chloride in nearly all cases. There was one exposure to 5.7 mg/m^3 phosphorus trichloride (1 hour) and one to 4.2 mg/m^3 phosphoryl chloride (25 minutes). A significant difference between exposed and unexposed workers is also reported in a follow-up medical study made two years later (16). Half (13 of 26 persons) of the exposed workers had periods of acute breathing difficulty, tightness in the chest and breathlessness (5 of them regarded the symptoms as work-related), whereas none of the unexposed workers (11 persons) reported these symptoms.

Two studies from Italy (23, 24) contain exposure data and describe respectively effects on 23 workers exposed to phosphorus trichloride and 20 workers exposed to phosphorus oxychloride. Air concentrations varied considerably in both cases. They were reported to be 10 – 20 mg/m^3 most of the time, but could occasionally exceed 150 mg/m^3 (phosphorus trichloride) or 70 mg/m^3 (phosphorus oxychloride). The reports contain no information on hydrolysis products. Photophobia, stinging in eyes and throat, chest tightness, coughing and rapid breathing were reported in a few subjects within 2 to 6 hours of exposure, and some of them subsequently developed bronchitis. In other cases it took 4 or 5 days or up to 8 weeks for symptoms to appear, and then in the form of slight throat irritation, conjunctivitis, coughing, shortness of breath and asthmatic bronchitis. Emphysema was also mentioned. The studies have the form of multiple case reports, and there is no collation or analysis of the findings. The diagnoses are based only on clinical observations and x-rays: there is no mention of medical history or smoking habits, for example. All this gives rise to some uncertainty in assessing the results, and it is therefore impossible to draw any definite conclusions from these studies.

A sketchily reported Russian study (21) with volunteers gives irritation thresholds of 4 mg/m^3 for phosphorus trichloride, 10 mg/m^3 for phosphorus pentachloride and 1 mg/m^3 for phosphoryl chloride. Since no details on the results or the design of the experiment are given, however, the information can not be satisfactorily assessed.

Skin burns have been reported in persons splashed with phosphorus trichloride or phosphorus pentachloride (concentrations not given) (9, 19).

Animal data

The LC_{50} values for exposure to vapor/aerosol of phosphorus trichloride reported in different studies range from 226 to > 2582 mg/m^3. Reported LC_{50} values for phosphoryl chloride range from 71 to 330 mg/m^3 (18, 21, 28). The reported LC_{50} value for phosphorus pentachloride is 205 mg/m^3 (21). The LD_{50} values for oral administration to rats are 18 – 550 mg/kg for phosphorus trichloride, 600 mg/kg for phosphorus pentachloride, and 380 mg/kg for phosphoryl chloride (17, 18, 21). No LD_{50} information was found for skin application, but an LD_{Lo} of 1260 mg/kg has been reported for application of phosphorus trichloride to the skin of rabbits (24 hours) (18). Phosphorus trichloride applied to the skin of rabbits is reported to cause burns (one reference reports that the substance was undiluted) (17, 18). Phosphorus pentachloride (concentration not reported) and phosphoryl chloride (pure substance) have also been reported to cause burns when applied to the skin of rabbits (17, 18). Application of phosphorus trichloride (concentration not reported) or phosphoryl chloride (pure substance) in liquid form has also been shown to cause severe damage to the eyes of experimental animals (17, 18).

During 4-hour exposures made to determine the LC_{50} for phosphorus trichloride and phosphoryl chloride, experimental animals (rats and guinea pigs) showed agitation, indications of irritation, porphyrin secretion around the eyes and labored breathing (28). The exposure to phosphorus trichloride also caused severe erosion of the nostrils and paws as well as kidney damage (nephrosis). The exposure to phosphoryl chloride caused irritation in the trachea, bronchi and lungs. The deaths occurred within 10 days after exposure to phosphorus trichloride and within 48 hours after exposure to phosphoryl chloride. The study reports that about 40% of the phosphorus trichloride and 15% of the phosphoryl chloride were hydrolyzed (28).

Effects on eyes and respiratory passages were reported in a study in which rabbits and cats (one of each per group) were exposed by inhalation to concentrations of phosphorus trichloride ranging from 4 mg/m^3 to 3870 mg/m^3 for 3 to 10 hours (6). There was a large difference in sensitivity between the two species. Sneezing, coughing, salivation, nasal secretion and reduced respiratory rates were seen in the cats at exposure as low as 4 – 5 mg/m^3, and effects on the eyes were noted at air concentrations of 13 – 20 mg/m^3 or above. Histological examination (cats) revealed liquid in the lungs (13 – 20 mg/m^3). Exposed rabbits – two animals exposed to air concentrations of 13 – 20 mg/m^3 and 13 – 27 mg/m^3 respectively – became somewhat restless, had greatly reduced respiration rates, slight symptoms of irritation and/or slight nasal secretion (see Table 1).

A Russian study (21) reports that the threshold value for irritation of respiratory passages (rats) was 5 mg/m^3 for phosphorus trichloride, 8 mg/m^3 for phosphorus pentachloride and 1 mg/m^3 for phosphoryl chloride. The study also reports that the effect was more pronounced for phosphorus trichloride (clouding of the cornea, sores around the mouth and nose, pronounced irritation of respiratory passages) than for the other phosphorus chlorides. The same study reports that “dystrophic changes,” particularly in the liver, kidneys and nervous system, were observed after

single exposures to high (not reported) air concentrations of phosphorus chlorides, and that exposure to 10 mg/m^3 for 4 hours caused a reduction of pH in blood and urine. Pronounced morphological changes, most notably in respiratory passages, kidneys, liver, bone tissue (osteoporosis) and brain (degenerative changes in nerve cells), as well as cytogenetic effects (see below), were also observed after 4 months of exposure to 1.34 mg/m^3 phosphoryl chloride, and irritation of mucous membranes in airways and elevated kidney weights were noted at 0.48 mg/m^3. Since the report gives no details on the design of the experiment, controls etc., this information can not be evaluated.

Mutagenicity, carcinogenicity, reproduction toxicity

No mutagenic effects were observed when phosphorus trichloride was tested on bacteria in vitro (15). A Russian study (21), which can not be adequately assessed (see above), reports mutagenic and cytostatic effects in rats (bone marrow) after chronic exposure to 1.34 mg/m^3 phosphoryl chloride but no significant changes after exposure to 0.48 mg/m^3. Phosphoryl chloride (air concentration not given) was also reported to affect the motility of sperm but to have no effect on spermatogenesis.

Dose-effect/dose-response relationships

There are few reliable measurements of air concentrations of these phosphorus chlorides in work environments. Two Italian studies report symptoms of eye irritation with exposure to phosphorus trichloride and airway irritation with exposure to phosphoryl chloride. Air concentrations varied, but in both cases were reported to be around 10 – 20 mg/m^3 most of the time.

Effects on experimental animals exposed by inhalation to phosphorus trichloride are summarized in Table 1.

Dose-dependent effects on respiratory passages were observed in cats at air concentrations of 4 – 5 mg/m^3 phosphorus trichloride or higher, and dose-dependent effects on eyes at air concentrations of 13 – 20 mg/m^3 or higher.

Conclusions

The critical effect of exposure to phosphorus trichloride, phosphorus pentachloride and phosphoryl chloride is irritation of respiratory passages. Due to their chemical characteristics, these three substances can also irritate/ulcerate eyes and skin.

Table 1. Effects on experimental animals exposed by inhalation to phosphorus trichloride.

Exposure (mg/m^3)	Species	Effects	Ref.
930 – 1070 4 hours	cat, rabbit (one of each)	Cat: agitation, salivation, red noses, dyspnea, corneal and nasal ulceration, pleurisy, reddened trachea, death after 36 hours. Rabbit: sneezing, agitation, conjunctivitis, secretions from eyes and nose.	6
530 – 1090 6.5 hours	cat (one animal)	Salivation, dyspnea, death after 390 minutes, corneal ulceration, emphysema, swollen epiglottis.	6
586 4 hours	rat	LC_{50}	28
330 6 hours	cat, rabbit (one of each)	Cat: agitation, coughing, sneezing, salivation, conjunctivitis, rhinitis, dyspnea, red nose, ulcerated cornea and nose, emphysema, swollen epiglottis. Rabbit: agitation, nasal secretion, rhinitis, reduced respiratory rate, dyspnea, red nose, ulcerated cornea.	6
282 4 hours	guinea pig	LC_{50}	28
226	rodents	LC_{50}	21
40 – 90 7 hours	cat, rabbit (one of each)	Cat: sneezing, secretion, cough, dyspnea, conjunctivitis, red nose. Rabbit: marked decline in respiratory rate, but few other symptoms.	6
13 – 27 6 hours	cat, rabbit (one of each)	Cat: salivation, dyspnea. Rabbit: slight irritation, nasal secretion, reduced respiratory rate.	6
13 – 20 6 hours	cat, rabbit (one of each)	Cat: salivation, nasal secretion, cough, dyspnea, conjunctivitis, liquid accumulation in lungs. Rabbit: slight restlessness, marked drop in respiratory rate.	6
4 – 5 3 hours	cat (one animal)	Sneezing, coughing, salivation, nasal secretion, reduced respiratory rate.	6

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Consensus Report for Glutaraldehyde

September 30, 1998

This report is based primarily on a criteria document produced jointly by the Nordic Expert Group and the Dutch Expert Committee (3).

Chemical and physical data. Uses

CAS No.:	111-30-8
Name:	glutaraldehyde
Synonyms:	glutaral, pentanedial, glutardialdehyde, 1,5-pentanedial
Formula:	$CHO-(CH_2)_3-CHO$
Molecular weight:	100.13
Boiling point:	188 °C
Freezing point:	- 14 °C
Vapor pressure:	0.00016 kPa (20% solution) 0.002 kPa (50% solution)
Saturation concentration:	6.6 mg/m^3 (1.6 ppm) (20% solution) 82 mg/m^3 (20 ppm) (50% solution)
Distribution coefficient:	log $P_{o/w}$ = 0.01
Conversion factors:	1 mg/m^3 = 0.25 ppm 1 ppm = 4.0 mg/m^3

Glutaraldehyde at room temperature is a colorless, oily liquid with a sharp odor. The reported odor threshold is 0.04 ppm (2, 3). Glutaraldehyde is soluble in water, ethanol, benzene, ether and other organic solvents. It can react violently with strong oxidants. An aqueous solution of glutaraldehyde has a pH of 3 – 4.

Glutaraldehyde is marketed in aqueous solutions of 1%, 2%, 25% or 50%. The solutions may contain alkalis added to raise their pH to 7.5 – 8.5 (activated solutions). Glutaraldehyde has a wide range of uses: as a disinfectant and sterilizer in hospitals, in embalming, as a fixative in electron microscopy, as a slimicide in the paper industry etc.

Glutaraldehyde was monitored in hospitals in England: concentrations ranged from 0.003 to 0.17 mg/m^3 (19). In Denmark, concentrations of 0.25 to 0.5 mg/m^3 were measured in surgery wards (28). In a Swedish study, the highest concentration – 0.57 mg/m^3 – was associated with sterilization of gastroscopes. The average of 16 measurements made around this task was 0.05 mg/m^3 (25).

Uptake, biotransformation, excretion

In an in vitro study, skin from rats, mice, rabbits, guinea pigs and humans was exposed to a 1,5-^{14}C-labeled glutaraldehyde solution for 6 hours. Concentrations were 0.75% or 7.5%. Between 0.5 and 0.7% of the solution was absorbed through/into the skin (11). Human stratum corneum and epidermis were exposed in vitro to 450 µl of a 10% glutaraldehyde solution for 1 hour. Penetration through the stratum corneum ranged from 3.3% (skin from the back) to 12% (skin from the stomach), with large individual variations. Penetration through epidermis was about 4% of the applied amount (27). When glutaraldehyde (0.75 or 7.5%) was left on the skin for 24 hours, the amount absorbed was calculated to be 4 – 9% for rats, and 33 – 53% for rabbits (3, 23).

Biotransformation of glutaraldehyde involves oxidation, decarboxylation and hydroxylation. Oxidation to glutaric acid, bonding to coenzyme A and breakdown to acetate yields the end product carbon dioxide. In hepatic and renal tissue of rats in vitro, glutaraldehyde is oxidated (probably in the mitochondria) to CO_2 (26). Doses of 0.2 ml of a 0.075 or 0.75% solution of ^{14}C-labeled gutaraldehyde were injected into the caudal vein of rats, and up to 80% of the radioactivity was found in CO_2 during the next 4 hours. Within 3 days 90% of the radioactivity had left the body (3, 23).

Toxic effects

Human data

Glutaraldehyde solutions can cause skin irritation, the severity of which depends on the strength of the solution and the duration of the contact. Inhalation of low levels of glutaraldehyde – less than 0.8 mg/m^3 – has been reported to cause irritation of nose and throat as well as nausea and headache (4). A special effect is the bleeding in mucous membranes of the intestine that may be caused by endoscopes sterilized in glutaraldehyde (8).

Of 167 nurses in endoscopy units, 65% had complaints of eye irritation, skin irritation, headaches, coughing and nasal congestion. In those cases in which measurements of glutaraldehyde concentrations are reported, they are below 0.2 ppm (0.8 mg/m^3) (5). There are several reported cases of sensitization caused by glutaraldehyde (3). Repeated or prolonged contact with glutaraldehyde or disinfectants containing glutaraldehyde has caused dryness, redness, eczema, cracking and sensitization of the skin (3). In a multi-center study of patients patch-tested at dermatology clinics in Germany over a 5-year period (1990 – 1994), it is reported that the number of patients sensitized to glutaraldehyde increased markedly during the study period (29). In a follow-up report (30) covering the years 1992 – 1995, 1194 women working in health care were tested: 10% had a positive response, compared with 2.6% of about 4000 patients who did not work in health care. Dental nurses were found to have the highest risk of skin sensitization (30).

Skin tests were given to 109 volunteers, using a 0.5% glutaraldehyde solution for both induction and provocation. One of the 109 subjects had a clear reaction, and 16

developed mild local erythema (redness) (1). In another study with 102 persons, 0.1% glutaraldehyde in vaseline was used for induction and 0.5 % in vaseline for provocation. No sensitization was observed. When the induction dose was 5.0%, 7 of 30 persons became sensitized to glutaraldehyde (21).

There are several case reports of severe asthma attacks suffered by asthmatics exposed to glutaraldehyde (3, 6, 7, 14, 32). There are also descriptions of six cases of glutaraldehyde-induced asthma in non-asthmatics, four of whom were not atopic (14).

Animal data

An alkaline 2% glutaraldehyde solution applied to the skin of rabbits caused "moderate" skin irritation. When a 24% glutaraldehyde solution was applied to rabbit skin it caused edema, followed by necrosis and scarring (3, 34). A drop of 2% acid glutaraldehyde solution placed in the conjunctival sac of rabbit eyes caused severe damage to the conjunctiva (edema and inflammation). A 2% alkaline solution applied to the eyes of rabbits caused opacity of the cornea and irritation of the iris, and was judged to be severely irritating to the eyes (3, 24).

Glutaraldehyde in gas form caused eye irritation at a concentration of 0.2 ppm. Mice were exposed to concentrations ranging from 1.6 to 36.7 ppm and the RD_{50} (a measure of airway irritation) was calculated to be 13.9 ppm. The LC_{50} for glutaraldehyde was estimated to be 24 – 40 ppm (3).

Solutions of 0.3, 1.0 and 3.0% glutaraldehyde were tested in a skin sensitization study with guinea pigs. A 10% solution was used as provocation. Each group consisted of six animals. There was no difference between the lowest dose group (0.3% solution) and controls (index 0.4). In the group receiving the 1.0% solution the index was 1.1 and in the high-dose group 2.7. In a positive control group that received DNFB (1-fluoro-2,4-dinitrobenzene) the index was 5.9. The maximum non-irritating concentration was reported to be 3%, since the 10% solution produced some irritation. The result was the same when the same test, using the same concentrations, was given to mice, but here the lowest dose group was also significantly different from the vehicle-control group (33).

A modified Magnusson-Kligman test was given to 30 guinea pigs using a 10% solution of glutaraldehyde, and 72% of the animals were sensitized. Glutaraldehyde was concluded to be a potent allergen. Cross-sensitization was shown between glyoxal, formaldehyde and glutaraldehyde (10). In another type of test, the mouse ear swelling test, a 1% solution was used for induction and a 10% solution for provocation, and 67% of the animals were sensitized (12). With a local lymph node assay, it was shown that glutaraldehyde had greater potential than formaldehyde for inducing skin sensitization (18). Sensitization of respiratory passages was tested with guinea pigs, using 13.9 ppm as induction and 4.4 ppm as provocation. No indications of sensitization were observed (3).

Rats were given 0, 10, 20 or 40 mM glutaraldehyde by intranasal instillation. No damage was observed at 0 and 10 mM. The two higher doses caused inflammation, hyperplasia and squamous metaplasia in epithelium, and increased cell prolifera-

tion. The damage resembled that observed in rats after inhalation of carcinogenic concentrations of formaldehyde (31).

In a two-week study, groups of rats and mice (5 of each sex per group) were exposed to 0, 0.16, 0.5, 1.6, 5 or 16 ppm glutaraldehyde for 6 hours/day, 5 days/week. All the rats in the two highest dose groups and all the mice in the three highest dose groups died during the exposure period. Deaths were caused by respiratory arrest. Rats exposed to 1.6 ppm grew more slowly, and all of them had necroses in nasal epithelium. Two males and all the females in this group also had squamous metaplasia. At 0.5 ppm there was nasal hyperplasia in three males and squamous metaplasia in two males and one female (26). In a 13-week follow-up study, groups of rats and mice (10 animals of each sex per group) were exposed to 0, 62.5, 125, 250, 500 or 1000 ppb glutaraldehyde. Slower growth was noted in the males in the highest dose group, in the females in the two highest groups, and in the mice in the four highest groups. For rats, the NOAEL for damage to respiratory passages was determined be 125 ppb, whereas inflammation was observed in the noses of the mice at 62.5 ppb (15, 26).

Mice were exposed to 0.3, 1.0 or 2.6 ppm glutaraldehyde, 6 hours/day for up to two weeks, and histopathological damage to respiratory epithelium was observed in all exposure groups. Inhalation of 1.0 ppm for 14 days caused an elevated incidence of squamous metaplasia and necrosis in nasal epithelium. No damage was observed in the lungs (37).

Mutagenicity, carcinogenicity, teratogenicity

Glutaraldehyde has been shown to be genotoxic in vitro, and to induce mutations in both bacteria and mammalian cells. It has also caused sister chromatid exchanges and chromosome aberrations in mammalian cells in vitro. However, glutaraldehyde yielded negative results when it was tested in vivo, in both the micronucleus test and a test for chromosome aberrations in bone marrow (3, 13, 16, 22, 26, 36).

No elevation in the incidence of malignant tumors was observed in a mortality study of 186 occupationally exposed workers in a factory producing glutaraldehyde. There were 4 deaths due to cancer (6.1 expected), one each lymphosarcoma, stomach, lung and brain (35).

A cancer study with rats and mice is being made by the NTP, and results have not yet been reported.

Spontaneous abortions and birth defects were studied among hospital personnel exposed to glutaraldehyde disinfectants. No elevation in risk was observed (17).

In a study with mice, the animals were given a 2% glutaraldehyde solution by gavage on days 6 – 15 of gestation. Doses were 16, 20, 24, 40, 50 or 100 mg/kg body weight. The animals were sacrificed on day 18. Fetal weights in the lowest dose group were lower than those in controls. In the highest dose group there was a marked increase of deformities. The deformities were thus seen only at doses that were highly toxic to the mothers (20).

In a similar study, rats were given glutaraldehyde by gavage in doses of 25, 50, or 100 mg/kg body weight on days 6 – 15 of gestation. Maternal toxicity was seen in the highest dose group but morphological examinations of the fetuses revealed no teratogenic effects (9).

Dose-response/dose-effect relationships

Available data on human exposures do not provide a sufficient basis for estimates of a dose-response or dose-effect relationship. Data from inhalation studies with rats and mice are summarized in Tables 1 (rats) and 2 (mice).

Conclusions

There are little data that can be used as a scientific basis for an occupational exposure limit for glutaraldehyde. The critical effect is irritation of eyes and mucous membranes, which can occur at exposure levels below 0.2 ppm. Exposure to 0.0625 ppm (the lowest tested dose) can cause inflammatory changes in the nasal mucosa of mice.

Glutaraldehyde is definitely sensitizing to skin. It exacerbates asthma in asthmatics and may cause asthma in non-asthmatics.

Table 1. Effects noted in rats exposed to glutaraldehyde by inhalation

Exposure ppm	Exposure time	Effect	Ref.
24 - 120	4 hours	LC_{50}	3
1.6	6 h/d, 5 d/w, 2 weeks	Retarded growth	26
1.0	6 h/d, 5 d/w, 13 weeks	Lower weight gain	26
0.5	6 h/d, 5 d/w, 13 weeks	Squamous metaplasia in nose	26
0.25	6 h/d, 5 d/w, 13 weeks	Inflammation in nose	26
0.125	6 h/d, 5 d/w, 13 weeks	NOAEL for damage to respiratory passages	26

Table 2. Effects noted in mice exposed to glutaraldehyde by inhalation

Exposure ppm	Exposure time	Effect	Ref.
2.6	15 minutes	RD_{50}	37
1.6	6 h/d, 5 d/w, 2 weeks	10/10 animals died	26
1.0	6 h/d, 5 d/w, 13 weeks	20/20 animals died	26
1.0	14 days	Squamous metaplasia, epithelial necrosis	37
0.3	4 days	Damage to epithelium in respiratory passages	37
0.25	6 h/d, 5 d/w, 13 weeks	Retarded growth	26
0.125	6 h/d, 5 d/w, 13 weeks	Retarded growth	26
0.0625	6 h/d, 5 d/w, 13 weeks	Inflammation in nose	26

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Consensus Report for Methyl Tert-Butyl Ether

September 30, 1998

This report is an update of the Consensus Report of November 26, 1987 (38).

Chemical and physical characteristics.* Uses

CAS No.:	1634-04-4
Synonyms:	methyl *tertiary* butyl ether, methyl t-butyl ether, 2-methoxy-2-methyl propane, *tert.*-butyl methylether, methyl-1,1-dimethylether, MTBE
Formula:	CH_3-O-C$(CH_3)_3$
Molecular weight:	88.15
Density:	0.7404 (20 °C)
Boiling point:	55.2 °C
Vapor pressure:	32.67 kPa (245 mm Hg) (25 °C)
Autoignition temperature:	224 °C
Distribution coefficient:	log $P_{octanol/water}$ = 1.04 (25 °C)
Solubility:	4.8 g/100 g water
Saturation concentration:	320,000 ppm (25 °C)
Conversion factors:	1 ppm = 3.60 mg/m^3 (20 °C, 101.3 kPa) 1 mg/m^3 = 0.278 ppm (20 °C, 10l.3 kPa)

* from References 16, 18 and 33

Methyl tert-butyl ether (MTBE) is an aliphatic, branched ether. At room temperature it is a clear, flammable liquid with a characteristic odor and a low odor threshold (0.05 – 0.2 ppm) (28, 57). Peroxide formation on exposure to ultraviolet light is lower for MTBE than for linear ethers (25, 43).

MTBE is produced from methanol and isobutene, and on a very large scale (1). World production in 1994 was 20.6 million metric tons (24). Sweden produced 36,500 tons and imported 33,000 tons in 1996 (59).

Nearly all MTBE is used as an additive (oxygenator) in unleaded gasoline. MTBE raises the octane of gasoline and improves combustion, thus reducing emissions of carbon monoxide, benzene etc. (28, 67). MTBE is also used in chromatography as an eluent (37, 50) and in medicine to dissolve gallstones *in situ* (31, 34).

Since MTBE is extremely volatile, most exposure occurs via inhalation, and particularly in conjunction with production and distribution. A study from Finland

reports MTBE exposures of 0.8 to 63 ppm (10 – 40 minutes) for tank truck drivers delivering gasoline (27). A summary of exposure measurements made in the United States (28) gives MTBE exposures for distribution of pure MTBE (peaks of 14 – 1000 ppm) and MTBE in gasoline (peaks of 2 – 100 ppm for < 30 minutes), for filling station personnel (0.3 – 6 ppm; peaks of > 10 ppm for 1 – 2 minutes; 6 to 8-hour median values 0.1 – 1 ppm) and for professional drivers and garage mechanics (< 1 ppm, 4 hours). The general public is exposed mostly while putting gasoline in their cars (3 – 10 ppm, 2 minutes) and driving (0.002 – 0.02 ppm per hour) (36). Drinking water may be a further source of exposure: in some parts of the United States low concentrations of MTBE (ng/liter) have been detected in groundwater following leakage from gasoline storage tanks (28).

It has been estimated that uptake of MTBE by people who are occupationally exposed to gasoline or MTBE in air is 0.1 to 1.0 mg/kg body weight/day, and that uptake for people not occupationally exposed (uptake from the general environment) is 0.0004 to 0.006 mg/kg body weight/day (14). These estimates are based on a collation of analyses and occupational exposure data collected in the United States.

Uptake, biotransformation, distribution, excretion

Uptake

In studies with volunteers, uptake of MTBE has been reported to be 32 to 42% of amount inhaled with exposures to concentrations ranging from 5 ppm to 75 ppm for two to four hours of rest or light physical labor (0 – 50 W) (52, 55). There are no quantitative data on human uptake via skin or digestive tract.

In rats, uptake from the digestive tract is rapid and complete, whereas skin uptake is limited (44). Absorption via the lungs is also rapid, and MTBE concentrations in blood reach a plateau about 2 hours after the start of exposure to low levels (400 ppm) as well as high ones (8000 ppm).

Biotransformation

MTBE is metabolized by oxidative dealkylation to tert-butyl alcohol (TBA) and formaldehyde. TBA and MTBE have been detected in human blood and urine (17, 46, 52, 55, 57). In addition, α-hydroxyisobutyric acid and 2-methyl-1,2-propanediol have been identified in the urine of persons exposed by inhalation to 50 ppm 1,2-^{13}C-labeled MTBE for two hours (n = 4) (53) and after oral intake of 5 mg/kg ^{13}C-labeled TBA (n = 1) (7).

Rat liver microsomes biotransform MTBE to TBA (13) and the TBA to formaldehyde (20). TBA was found in the blood of rats exposed to ^{14}C-labeled MTBE (44). Four other metabolites have been found in urine, and two of them have been identified as α-hydroxyisobutyric acid (70% of total excreted radioactivity) and 2-methyl-1,2-propanediol (14%). The three main metabolites found in the urine of rats after 6 hours of exposure to 2000 ppm 2-^{13}C-labeled MTBE were α-hydroxyisobutyric acid, 2-methyl-1,2-propanediol and an unidentified conjugate of TBA (7).

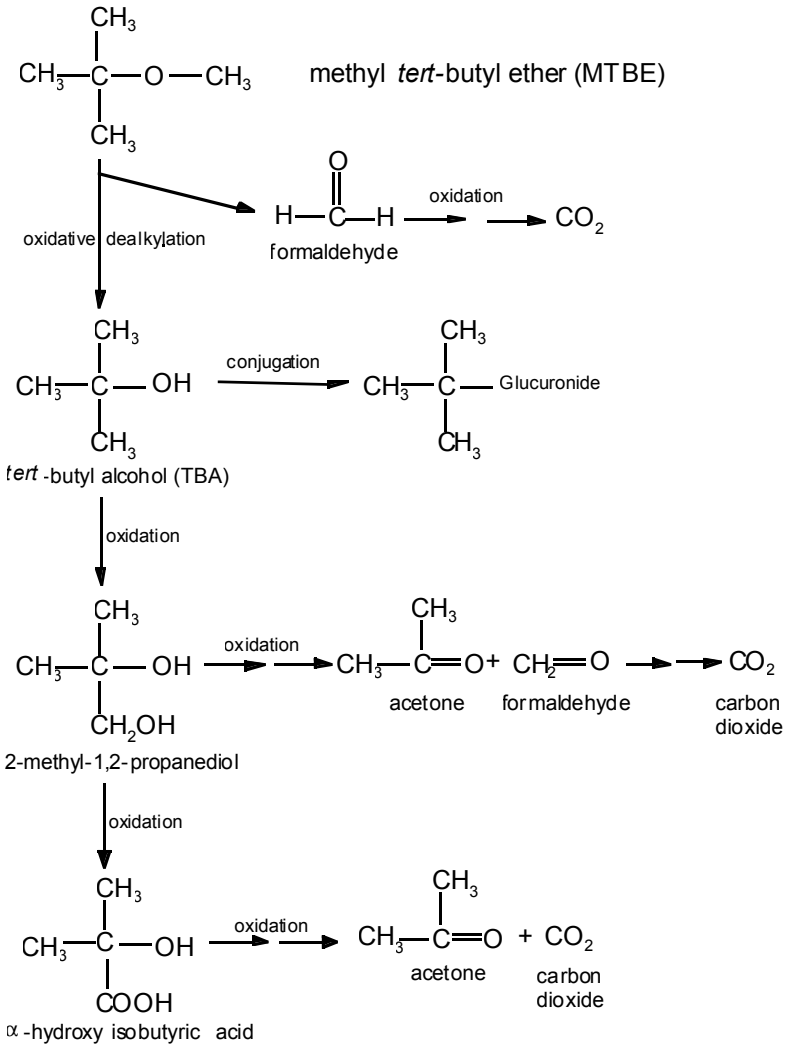

Fig 1. Proposed metabolism pathways for MTBE (rats) (28).

Low concentrations of TBA, TBA conjugate (possibly a glucuronide) and acetone were also found. Rats exposed to ^{14}C-labeled TBA excreted the isotope in acetone and carbon dioxide (5). Figure 1 shows the proposed pathways for metabolism of MTBE in rats.

MTBE activates UDP-glucuronosyltransferase and the cytochrome P-450 system (isoenzymes 2B1, 2A6 and 2E1) in liver microsomes from both rats and humans (13, 30, 63, 66). Hong et al. (29) measured the metabolic activity (formation of TBA) in microsomes in various organs in male rats and found much higher activity in nasal mucosa than in liver.

Distribution

The olive oil/blood distribution coefficient for MTBE is 7 to 10, which indicates that the substance, like many other solvents, has a greater affinity for fatty tissue than for blood (12, 51). For TBA, distribution coefficients are 0.27 for oil/water and

0.36 for oil/blood, indicating that in the body this metabolite tends to stay in body fluids.

When volunteers were exposed to MTBE in a closed chamber (5 – 75 ppm, 2 – 4 hours, 0 – 50 W), concentrations of MTBE in blood rose rapidly at first and leveled out toward the end of the exposure period (17, 52, 55, 57). As soon as exposure was terminated blood levels of MTBE began to drop rapidly, and two to three hours later concentrations were down to about 1/10 of maximum levels. In contrast, the concentration of TBA in blood increased slowly and reached a plateau after exposure was ended (52, 55, 57). TBA in blood began to drop two to four hours after termination of exposure, and then dropped more slowly than MTBE: 24 hours after the exposure blood levels of TBA were about a third of peak levels. Concentrations of MTBE and TBA in blood were proportional to exposure levels, which suggests linear kinetics, i.e. metabolism is not saturated at the tested concentrations (up to 75 ppm for humans) (52, 55).

MTBE and TBA in the blood of rats also exhibited linear kinetics after inhalation of 50, 100 or 300 ppm MTBE for 2 weeks (63).

Excretion

In the 24 hours following the volunteers' exposure in the chamber, 32 to 58% of the MTBE taken up was eliminated unchanged in exhaled air (52, 55) and about 1% as MTBE or TBA in urine (15, 52, 55, 57).

Excretion of ^{14}C-labeled MTBE by rats was rapid and independent of sex or method of administration. Most of the radioactivity was excreted within 3 hours via lungs and within 24 hours via urine (44). After intravenous administration of ^{14}C-labeled MTBE (40 mg/kg body weight), 60% of the radioactivity was eliminated via lungs, 35% in urine and 2% in feces, and 0.4% remained in tissues. With increasing doses of MTBE the proportion in urine decreased and the proportion in exhaled air increased (this was seen primarily with inhalation exposures, but also with oral doses). The shift in excretion pathways suggests that one or more of the stages in metabolism of the substance tends to become saturated at high exposures (8000 ppm and 400 mg/kg).

In another study, mice were give MTBE by intraperitoneal injection (50, 100 or 500 mg/kg), and 23 to 69% of the dose was eliminated unchanged in exhaled air – most of this (90%) within three hours (70).

Toxic effects

Human data

Healthy volunteers were exposed to MTBE under controlled conditions in a chamber: levels were chosen to approximate non-occupational exposure while putting gasoline in a vehicle (0 – 1.7 ppm) (17, 57) and occupational exposure (0 – 75 ppm) (52, 54, 55, 60).

Prah et al (57) exposed healthy men and women (n = 37) to pure air or to 1.4 ppm MTBE for 1 hour (resting). Before and after the exposures, the subjects filled in questionnaires and were given objective tests: eye exams (redness, "tear

film breakup time") and analysis for inflammation markers in nasal lavage fluid and tears. The only significant result observed was a difference in the assessments of air quality: the women reported that the air smelled better under control conditions than during the MTBE exposure.

A corresponding study was made by Cain et al (17): 43 healthy volunteers were exposed to pure air or to air containing 1.7 ppm MTBE or 7.1 ppm VOC (a mixture of 17 organic chemicals) for 1 hour (resting). They made subjective assessments (eye or throat irritation, headache, feelings of grogginess or light-headedness, air quality etc.) and took a computerized performance test, and objective examinations (described above for the previous study) were made before and after the exposures. No acute effects could be related to the MTBE exposure, although the subjects did notice the difference in the odors.

In a Swedish study, ten healthy men were exposed to 5, 25 or 50 ppm MTBE for 2 hours (light physical work; 50 W) (54). Subjective assessments (odor, breathing difficulty, headache, fatigue, nausea, dizziness, grogginess or irritation of eyes, nose or throat) and objective examinations of the eyes (redness, "tear film breakup time", damage to conjunctival epithelia, blinking frequency) and nasal mucosa (nasal congestion and inflammation markers in nasal lavage fluid) were made before and after the exposures. Estimates of the odor were significantly higher when the subjects first entered the chamber, but declined as exposure progressed. A slight nasal swelling was noted after the exposures, but the effect was not dose-related and probably not related to the exposure to MTBE.

In a Finnish study, 13 subjects were exposed to 0, 25 or 75 ppm MTBE for 4 hours (resting) (60). Symptoms and other effects were assessed, and reaction times and balance were tested during the exposures (after 1 and 3 hours) and 1 hour afterward. The frequency of symptoms increased with level and duration of exposure. At the highest level (75 ppm) and after 3 hours of exposure, there was a significant increase in reports of minor symptoms such as grogginess, and also to a lesser extent irritation of mucous membranes. Most of the symptoms had disappeared when assessments were made one hour after exposure was terminated. Six of the 13 persons reported MTBE-related symptoms. No effects related to the exposures were noted in the tests of balance and reaction time.

In the United States, several field studies and epidemiological studies have been made in which subjective assessments of effects were compared with exposures (4, 26, 28, 45, 46, 68). In Fairbanks, Alaska, 18 persons exposed to gasoline were given blood tests and filled in questionnaires: the reported frequencies of the following symptoms: headache, eye irritation, irritation of nose and throat, coughing, nausea, dizziness, disorientation, ranged from 33 to 72% (46). Measured MTBE levels were 0.02 μM in blood and 0.1 ppm in air (8-hour average). A follow-up study was made three months later, when MTBE was not added to gasoline. At that time the frequencies of the symptoms ranged from 0 to 7% among those exposed to the gasoline (n = 28). Measured MTBE levels on that occasion were 0.003 μM in blood and 0.04 ppm in air (8-hour average). Other studies (4, 26, 28, 45, 46, 48) have found no connection between MTBE exposure and

subjective assessments of acute symptoms. The Fairbanks study has been criticized on the grounds that a number of external factors – odor, increase in gasoline price, winter climate, media coverage etc. – may have influenced the persons reporting the symptoms (24, 28, 67).

Vojdani et al (69) compared 32 unexposed subjects with 60 persons who had been exposed to water containing MTBE (0.0036 – 0.27 μg/liter) and benzene (0.00064 – 0.045 μg/liter) for 5 to 8 years (the analysis method is not described). The proportion of apoptosis (programmed cell death) observed in the lymphocytes of exposed persons (in vitro) was significantly higher than that in the controls. Cell cycle progressions were determined, and the persons who had elevated apoptosis had more cells in the DNA synthesis or mitosis phase than in the resting phase (compared with controls). It is not clear how this applies to risk assessment, but according to the authors these cellular deviations may be due to exposure to MTBE or benzene or their metabolites or to a synergistic effect of MTBE and benzene.

MTBE is sometimes used to dissolve gallstones. It is introduced into the gallbladder via a catheter, and the procedure is repeated as often as necessary. Acute effects – nausea, vomiting, drowsiness, mild inflammation of intestinal mucosa, etc. – have been reported. Accidental leakage has occurred during a few treatments, and has resulted in lethargy, hemolysis, low blood pressure, kidney failure and stomach ulcers (31, 34, 56, 65).

Animal data

The acute toxicity of MTBE is low to moderate. For mice, the LD_{50} is 4000 mg/kg (37) and the LC_{50} for inhalation exposure (15 minutes) has been determined to be 39,000 ppm (40).

Rats were given oral doses of MTBE daily for 14 days (357, 714, 1071 or 1428 mg/kg body weight) or 90 days (100, 300, 900 or 1200 mg/kg) (61). Anesthesia was observed at doses of 1200 mg/kg and above, and lasted for 2 hours, but then disappeared entirely. Diarrhea was common in all treatment groups, but there were no deaths. Animals in the 14-day study had reduced lung weights and the females had lower urea and creatinine levels in their blood, whereas cholesterol was elevated in both sexes. In the 90-day study there was an increase of cholesterol in blood, but blood urea (females) and creatinine (males) dropped. Organs were unchanged, except for accumulation of α_{2u}-globulin in the kidneys of males in the high-dose group (1200 mg/kg), which is a known and specific effect in male rats.

In a long-term study, rats were exposed for 24 months and mice for 18 months to 0, 400, 3000 or 8000 ppm MTBE (6 hours/day, 5 days/week) (11). Toxicity was observed at the two higher dose levels. At 8000 ppm there were clinical indications of effects on the central nervous system (eyelid tics, reduced activity, ataxia and deterioration of reflex movements). The rats showed these effects for up to a week after the start of exposure, but in the mice the effects continued throughout the study. In addition, there were changes in body weights and organ weights in both species, and the males had shorter life spans. No exposure-related hematologic

changes were observed, but there were lowered corticosterone levels in the male rats exposed to 8000 ppm. Liver and kidney weights were higher in rats exposed to 3000 and 8000 ppm, but this was not accompanied by any histopathological changes.

The occurrence of neurotoxicity was examined in rats after exposure to MTBE concentrations of 0, 800, 4000 or 8000 ppm for 6 hours or 13 weeks (23). In the 6-hour study, there were indications of an acute reversible effect on the central nervous system (ataxia, changes in respiratory rate and movement patterns, and loss of gripping strength in hind legs) for up to an hour after the exposure to 8000 ppm and to a lesser extent after the 4000 ppm exposure as well. In the 13-week study, examinations were made 42 to 50 hours after the last exposure day and no effects on the nervous system were observed.

In a 13-week inhalation study (0, 800, 4000 or 8000 ppm) increases in liver, adrenal and kidney weights were noted in rats (both sexes) at the two higher exposure levels (35). At the highest level the rats had lower body weights and poor coordination (the first 4 weeks), and the males also had slight histopathological changes in spleen, kidneys and lymph nodes.

In another inhalation study (0, 400, 1500 or 3000 ppm MTBE, 6 hours/day, 10 days) male rats in the highest exposure group had elevated concentrations of α_{2u} -globulin (58). Necrosis and protein droplet accumulation were observed in renal collecting ducts at 1500 ppm.

Mice were exposed to MTBE concentrations of 83, 280, 830, 2800 or 8300 ppm for 1 hour (64). In all but the highest dose group, respiratory rates dropped initially but normalized after five to ten minutes of exposure. At 8300 ppm the respiratory rate was lower during the entire exposure and did not return to normal until 15 minutes after the exposure was ended. The authors attribute this to both effects on the respiratory passages and ”sensory irritation” (3). Analysis of cells in bronchoalveolar lavage fluid showed no changes in this group.

To examine the question of whether MTBE causes tissue damage when used to treat gallstones, MTBE was injected (2 ml/kg body weight) into etherized rats, either through the vena cava to the central circulatory system (n = 13), through a peripheral vein (n = 10), or into the hepatic parenchyma (n = 22) (2). The study demonstrated that MTBE is locally cytotoxic to tissues and causes severe and often fatal lung damage when injected into the vena cava.

Mutagenicity, carcinogenicity, teratogenicity

Animal data

In a genotoxicity study, rats were exposed to 800, 4000 or 8000 ppm MTBE 6 hours/day for 5 days and mice to 400, 3000 or 8000 ppm MTBE 6 hours/day for 2 days (41). In another study, mice were given injections of MTBE in single doses of 0.25, 0.5, 1, 1.5 or 1.75 g/kg body weight (32). Samples of bone marrow were taken from the femurs 6, 24 or 48 hours after the exposures (41) or 24 hours after the injections (32). No changes were noted in either chromosome aberrations in

bone marrow of the rats or number of micronuclei in the mice. MTBE did not induce DNA repair in liver cells of mice (41) or rats (exposures not given) (21). No mutagenicity was observed in the sex-linked recessive lethal test with *Drosophila* after administration of 0.01 – 0.3% MTBE in food (41).

MTBE was negative (i.e. no point mutations) in Ames tests (21, 32). It caused no gene mutations in V79 cells from Chinese hamsters, either with or without the addition of liver fraction (S9 mix); however, the survival of the cells was lower in the presence of the S9 mix (21). In one of the studies (32) some toxicity was observed at the highest dose (7400 μg MTBE).

The study above was repeated (21), and this time the mutagenicity of MTBE was tested with the addition of formaldehyde dehydrogenase and its cofactor NAD^+ (39). A linear increase of mutation frequency and a reduction of cell proliferation were observed without the enzyme system, but when the system was added the effect was reduced. The authors regard this as an indication that the metabolite formaldehyde may affect the mutagenicity of MTBE.

In an inhalation study, rats were exposed for 24 months and mice for 18 months to 0, 400, 3000 or 8000 ppm MTBE (6 hours/day, 5 days/week). The two higher concentrations had toxic effects (11). At 8000 ppm there was an increase in the number of hepatocellular adenomas in the female mice. This observation was followed up with another study, with exposures for 5 or 28 days at the same levels, and a significant increase of cell proliferation in the livers of female mice was seen after 5 days of exposure, but not after 28 days. The authors regard this as an indication that MTBE induces mitogenesis. Male rats were killed before the end of the study (8000 ppm – week 82; 3000 ppm – week 97) because so many of them were dying of advanced nephrosis. At the end of the study an elevated prevalence of chronic nephropathy was observed in male rats in all exposure groups and in females in the two higher groups. The nephropathy was associated with secondary organ damage to cell growth in the parathyroid and a mineralization of tissues. Kidney tumors (renal tubular cell tumors) were seen in males in the two higher exposure groups. The authors (11) suggest that this may be associated with the accumulation of a protein in the epithelium of the renal tubules (observed after as little as 4 weeks of exposure) and may be an effect similar to or analogous to α_{2u}-globulin (42). The high mortality of both mice and rats indicates, according to the authors, that at the highest exposure level (8000 ppm) the maximum tolerable dose had been exceeded.

In a long-term study, rats were given MTBE in oral doses of 0, 250 or 1000 mg/kg body weight, 4 days/week for 104 weeks (6). Survival after 80 weeks of exposure was higher in the high-dose group than in the controls or the low-dose group (males), though there were more testicular (Leydig cell) tumors in this group (controls 8%, low-dose group 8%, high-dose group 34%, calculated from the number of rats still living week 96, when the first tumor was discovered). Leydig cell tumors are quite common in control material (42), which suggests that the reported increase (6) may be random. For females there was a dose-related increase in mortality from week 32 onward. Increases of leukemia and lymphoma were seen

in females (combined leukemia and lymphoma: controls 3%, low-dose group 12%, high-dose group 25%, calculated from the number of living rats week 56, when the first leukemia was discovered). The authors point out that some fluctuation was expected, since up to 10% of their historical controls (females) had developed neoplasias, but the increase in the high-dose group was significant in comparison with these earlier controls as well. Despite the fact that the combined incidence of these two tumor types (leukemia and lymphoma) was significantly elevated, the results for the neoplasias were not presented separately (42).

No DNA–protein cross-links or RNA-formaldehyde adducts were observed in mouse liver cells in vitro after incubation with MTBE (19). However, incubation with formaldehyde yielded a concentration-dependent increase in the number of cross-links and adducts. According to the authors, this shows that the formation of formaldehyde from MTBE is slower than other endogenic metabolism, and indicates that metabolism of MTBE to formaldehyde does not make a critical contribution to MTBE's carcinogenicity in mice.

No treatment-related teratogenicity was observed in rabbits exposed to MTBE, up to 8000 ppm, for 6 hours/day on days 6 to 18 of gestation (9).

Mice exposed to 4000 or 8000 ppm MTBE for 6 hours/day on days 6 to 15 of gestation showed reduced activity, suppressed reflexes and changes in gestation; the effects were not seen at exposure to 0 or 1000 ppm (9).

Mice and rats were exposed to MTBE for 6 hours/day on days 6 to 15 of gestation (0, 250, 1000 or 2500 ppm): no toxic effects were observed in the mothers or in the fetuses, or on reproduction (22).

No reproduction toxicity was observed in two generations of rats after 10 weeks of exposure to MTBE in concentrations of 0, 400, 3000 or 8000 ppm (8).

In a one-generation study, rats were exposed to 300, 1300 or 3400 ppm MTBE – males for 12 weeks before mating, and females for 3 weeks (10). Exposures continued during gestation and nursing, and two litters were born. No effects on reproduction were observed. The only observed effect was a widened renal pelvis in the females exposed to 300 and 3400 ppm.

Moser et al (49) exposed female mice to air containing 7800 ppm MTBE or 2000 ppm gasoline (containing no MTBE) for 3 or 21 days (6 hours/day, 5 days/week). Elevated liver weights and reduced uterus weights were observed in all exposure groups. When mice were given the substances by gavage (1800 mg/kg MTBE or gasoline, 3 days) the metabolism of estrogen in isolated hepatocytes increased. The authors suggest that the increase of estrogen metabolism in the liver and reduction of uterus weight may indicate an endocrine modulation in both MTBE-induced and gasoline-induced liver carcinogenesis. In a follow-up study it was observed that exposure to MTBE (8000 ppm, 3 or 21 days, 4 or 8 months, female mice) elicits a response in some cells and tissues of the endocrine system, and that these effects are not activated through the estrogen receptor (47).

Female mice 12 days of age were initiated with either *N*-nitrosodiethylamine (7.1 ml DEN/kg body weight) or a saline solution, and exposed to 0 or 8000 ppm MTBE for 16 or 32 weeks (48). Elevated liver weights and increased microsomal

cytochrome P-450 activity in liver were seen, but no toxicity was observed. The MTBE exposure did not increase the size or volume fraction of liver foci compared with DEN-initiated controls. According to the authors, the absence of tumor formation in the DEN-initiated female mice exposed to MTBE was unexpected, and indicates that MTBE does not produce liver tumors by the same mechanism as gasoline.The genotoxicity, mutagenicity and carcinogenicity of MTBE in experimental animals have also been assessed with a predictive (structure-activity) computer program, which gave no indications of genotoxic, mutagenic or carcinogenic activity (62, 71).

Dose-effect/dose-response relationships

Human data

No clear dose-effect relationship has been found in epidemiological studies. In experimental studies, no effect has been observed at concentrations below 50 ppm (questionnaires and objective measures of eye and nose irritation) (17, 54, 57). After 3 hours of exposure to 75 ppm (n = 13) there was a significant increase of minor symptoms such as grogginess and irritated mucous membranes (60).

Animal data

The estimated NOAEL for rats is 1000 ppm, and the LOAEL is 1500 ppm, since effects on kidneys are seen at that level. Mice had reduced respiratory rates for the first 5 to 10 minutes of exposure to 83 ppm, and this level was therefore taken to be the LOAEL for mice. Dose-effect relationships observed in inhalation studies with animals are summarized in Table 1.

Conclusions

The critical effect of occupational exposure to MTBE is judged to be irritation of mucous membranes.

In a study with volunteers, grogginess and irritation of mucous membranes were reported at exposure to 75 ppm. In another study, no irritation or other effect was seen at 50 ppm.

Liver tumors have been observed in female mice, and renal and testicular tumors in male rats, after exposure to high concentrations of MTBE.

Table 1. Dose-effect relationships observed in experimental animals exposed to MTBE by inhalation (6 hours/day, 5 days/week)

Species, exposure	Observed effects	Ref.
Rabbit		
1000 ppm days 6-18 of gestation	No observed effects.	9
4000 ppm days 6-18 of gestation	Lower body weights and food consumption.	9
8000 ppm days 6-18 of gestation	Lower body weights and relative liver weights, lower food consumption.	9
Rat		
250 ppm days 6-15 of gestation	No observed effects.	22
400 ppm 10 days or 4 weeks	No observed effects.	11, 58
400 ppm 10 weeks or 24 months	No observed effects	8, 11
800 ppm 6 hours or 13 weeks	No observed neurotoxic effects	23
800 ppm 13 weeks	No observed effects	35
1000 ppm days 6-15 of gestation	No observed effects	22
1500 ppm 10 days	Necrosis and protein droplet accumulation in renal collecting ducts (males).	58
2500 ppm days 6-15 of gestation	No observed effects.	22
3000 ppm 10 days	Necrosis and protein droplet accumulation in renal collecting ducts, increased α_{2u}-globulin concentration in kidneys (males).	58
3000 ppm 4 weeks	Cell proliferation in kidneys on days 5 and 28 (males)	11
3000 ppm 10 weeks	F_1: reduced activity, depressed reflexes, elevated liver weights; no histopathological changes.	8
3000 ppm 24 months	Renal tumors (males); elevated kidney and liver weights (both sexes)	11
4000 ppm 6 hours	CNS effects (ataxia, changes in respiratory rate and movement patterns, reduced grip strength in hind legs) for up to 1 hour after exposure	23
2500 ppm days 6-15 of gestation	No observed effects	22
3000 ppm 10 days	Necrosis and protein droplet accumulation in renal collecting ducts, increased α_{2u}-globulin concentration in kidneys (males)	58
3000 ppm 4 weeks	Cell proliferation in kidneys on days 5 and 28 (males)	11
3000 ppm 10 weeks	F_1: reduced activity, depressed reflexes, elevated liver weights; no histopathological changes.	8
3000 ppm 24 months	Renal tumors (males) and elevated kidney and liver weights (both sexes).	11

Table 1. Continued

Species, exposure	Observed effects	Ref.
4000 ppm 6 hours	CNS effects (ataxia, changes in respiratory rate and movement patterns, reduced grip strength in hind legs) for up to 1 hour after exposure.	23
4000 ppm 13 weeks	No observed neurotoxic effects.	23
4000 ppm 13 weeks	Elevated liver, adrenal and kidney weights	35
8000 ppm 6 hours	CNS effects (ataxia, changes in respiratory rate and movement patterns, reduced grip strength in hind legs) for up to 1 hour after exposure.	23
8000 ppm 13 weeks	No observed neurotoxic effects.	23
8000 ppm 10 weeks	F_1: lower body weights, reduced activity, depressed reflexes, ataxia and elevated liver weights, but no histopathological findings. F_2: elevated mortality 4 days after birth	8
8000 ppm 13 weeks	Lower body weights, poor coordination (first 4 weeks); elevated liver, adrenal and kidney weights. Slight histopathological changes in spleen, kidneys and lymph nodes (males).	35
8000 ppm 4 weeks	Cell proliferation in kidneys on day 28 (males). Weight loss.	11
8000 ppm 24 months	Renal tumors, elevated mortality and reduced body weights (males). CNS effects during first week of exposure. Elevated kidney and liver weights (both sexes).	11
Mouse		
83 ppm 1 hour	Temporary reduction in respiratory rate (13%).	64
250 ppm days 6-15 of gestation	No observed effects.	22
280 ppm 1 hour	Temporary reduction in respiratory rate (17%).	64
400 ppm 4 weeks or 18 months	No observed effects.	11
830 ppm 1 hour	Temporary reduction in respiratory rate (28%).	64
1000 ppm days 6-15 of gestation	No observed effects.	9
2500 ppm days 6-15 of gestation	No observed effects.	22
2800 ppm 1 hour	Temporary reduction in respiratory rate (35%).	64
3000 ppm 4 weeks	No observed effects.	11
3000 ppm 18 months	Elevated kidney weights (males) and liver weights, reduced brain weights.	11
4000 ppm days 6-15 of gestation	Reduced activity, suppressed reflexes. Effects on gestation (fetal body weight/litter, skeletal anomalies).	9

Table 1. Continued

Species, exposure	Observed effects	Ref.
8000 ppm days 6-15 of gestation	Reduced body weights, activity and food consumption; suppressed reflexes. Effects on gestation (post-implantation damage, fewer males born, fetal body weight/litter, skeletal anomalies).	9
8000 ppm 4 weeks	Cell proliferation in livers (females) on day 5, disappeared by day 28.	11
8000 ppm 18 months	Hepatic adenomas (females); elevated kidney weights (males), elevated mortality (males). Elevated liver weight, reduced body, brain and spleen weights, CNS effects during first week of exposure (both sexes).	11
8300 ppm 1 hour	Reduction in respiratory rate (52%); normal again 15 minutes after termination of exposure.	64

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Consensus Report for Dimethyl Adipate, Dimethyl Glutarate and Dimethyl Succinate

December 9, 1998

Chemical and physical data. Uses

dimethyl adipate

CAS No.:	627-93-0
Synonyms:	dimethyl hexanedioate, methyl adipate
Formula:	$C_8H_{14}O_4$
Structure:	CH_3-O-CO-CH_2-CH_2-CH_2-CH_2-CO-O-CH_3
Molecular weight:	174.22
Boiling point:	231 °C
Melting point:	10.3 °C
Vapor pressure:	0.0016 kPa (20 °C)
Conversion factors:	1 ppm = 7.23 mg/m^3 (20 °C)
	1 mg/m^3 = 0.138 ppm (20 °C)

dimethyl glutarate

CAS No.:	1119-40-0
Synonyms:	dimethyl pentanedioate, methyl glutarate
Formula:	$C_7H_{12}O_4$
Structure:	CH_3-O-CO-CH_2-CH_2-CH_2-CO-O-CH_3
Molecular weight:	160.17
Boiling point:	214 °C
Melting point:	- 42.5 °C
Vapor pressure:	0.0061 kPa (20 °C)
Conversion factors:	1 ppm = 6.65 mg/m^3 (20 °C)
	1 mg/m^3 = 0.150 ppm (20 °C)

dimethyl succinate

CAS No.:	106-65-0
Synonyms:	dimethyl butanedioate, methyl succinate
Formula:	$C_6H_{10}O_4$
Structure:	CH_3-O-CO-CH_2-CH_2-CO-O-CH_3
Molecular weight:	146.14
Boiling point:	196.4 °C
Melting point:	19 °C
Vapor pressure:	0.0167 kPa (20 °C)
Conversion factors:	1 ppm = 6.06 mg/m^3 (20 °C)
	1 mg/m^3 = 0.165 ppm (20 °C)

Dimethyl adipate (DMA), dimethyl glutarate (DMG) and dimethyl succinate (DMS) usually occur together. The mixtures are a clear liquid (1). DMA, DMG and DMS are soluble in substances such as ethanol and ether (13). Solubility in water has been reported to be 29.9 g/l (20 °C) for DMA and 131 g/l (25 °C) for DMS (7). Product information leaflets state that the water solubility of mixtures containing > 55% DMG, >15% DMS and > 10% DMA is 5 to 5.5% by weight (20 °C), and that the vapor pressure is 0.013 kPa.

Mixtures of DMA, DMG and DMS are used as solvents. The mixtures are used industrially in paints and coatings, and also for paint removal and for cleaning polyurethane foam and unsaturated polyester resin (22).

DMS is also used in the cosmetics and food processing industries. It occurs, for example, as a flavoring in ice cream, candy, bakery products and drinks (2).

Uptake, biotransformation, excretion

There is little information in the literature on oral uptake of either DMA, DMG or DMS. In one study with rats (16), it was shown that DMS was rapidly absorbed in the digestive tract. There are no quantitative data on skin uptake for any of the three esters.

Nearly all of inhaled DMA, DMG and DMS is deposited in the upper respiratory passages. In one study (19), rats were exposed (one-way air flow) for 40 minutes to 50 –100 mg/m^3 DMA, DMG and DMS vapors, either separately or mixed, or to 250 – 320 mg/m^3 DMG vapor. It was found that deposition in the upper respiratory passages exceeded 97% in all cases – much the same for all three substances, both sexes, and all air concentrations tested.

DMA, DMG and DMS are metabolized by hydrolysis and can form monomethyl esters, dicarboxylic acids and methanol; this has been demonstrated in vitro with homogenates, nasal explants and isolated cells (5, 19, 22, 29, 30). In rats, the esters are efficiently hydrolyzed by carboxylesterase, mostly in the airways (especially in olfactory epithelium) and liver (4, 5, 19, 22, 30). There is some difference between males and females. In a comparative study, the efficiency of hydrolysis to monomethyl esters was tested with low (“subsaturating”) substrate concentrations of DMA, DMG and DMS in homogenates of olfactory epithelium from female or male rats. Hydrolysis was equally effective in homogenates from both sexes when DMG was used as substrate, whereas DMA was hydrolyzed more effectively by the olfactory epithelium homogenate from females and DMS by that from males. A structure-activity relationship was also observed in the hydrolysis reactions: DMA > DMG > DMS (5).

The activity of carboxylesterase in olfactory epithelium from humans (hydrolysis of DBE in 3/6 samples) was reported in an abstract to be 100 to 1000 times lower than that of rats (9). (DBE = dibasic esters, i.e. DMA, DMG and DMS.)

Glutaric acid (a metabolite of DMG) is one of the body’s natural components. It is formed in metabolism of the amino acid lysine (8). Malic acid (a metabolite of DMS) also occurs naturally in the body’s general metabolism. It is a component, for

example, of the energy-producing Krebs cycle, which yields the end products carbon dioxide and water (8, 14, 29). Increased synthesis of proinsulin and increased insulin secretion have been shown to follow administration of DMS, and are ascribed primarily to an influx of malic acid to the Krebs cycle in the pancreatic islets (isles of Langerhans) (15, 17, 18, 30).

Toxic effects

Human data

Product information leaflets from suppliers/manufacturers report that temporary effects on vision (foggy vision) may occur after exposure to mixtures of DMG, DMS and DMA. No air concentrations are given, but it is stated that the problem had occurred at exposure to "high" concentrations of vapor or after direct contact with the eyes (product mixtures containing > 55% DMG, > 15% DMS and > 10% DMA). The product information leaflets also warn that inhalation of "high" concentrations of the vapors can irritate respiratory passages.

Animal data

The LD_{50} for rats given DMA by intraperitoneal injection is reported in one study (21) to be 1.9 g/kg.

Mice were exposed by inhalation to various concentrations of DMA, DMG and DMS, either separately or in mixtures, and their respiratory rates were measured (mixture 1: 63% DMG, 25% DMS, 12% DMA; mixture 2: 57% DMG, 23% DMS, 20% DMA). The data were used to calculate the concentration that caused a 50% decrease in respiratory rate (RD_{50}), which is a measure of the irritation potential of a substance. The RD_{50} was reported to be 890 mg/m^3 for DMG, 910 mg/m^3 for DMA, 1600 mg/m^3 for DMS, 590 mg/m^3 for mixture 1 and 610 mg/m^3 for mixture 2. DMS appeared to be the least potent of the three esters but was reported to have the steepest dose-response curve (20).

In an inhalation study (12), male rats were exposed to a vapor-aerosol DBE mixture (90% aerosol) generated from a liquid that was 66% DMG, 17% DMS and 17% DMA. The animals were exposed for 4 hours to an average aerosol concentration of 5900 mg/m^3 and sacrificed at intervals of 1 to 42 days after the exposure. Damage (degeneration, inflammation, necrosis), which was largely reversible within 6 weeks, was seen in nasal mucosa exposed to air flow. Severe necrosis was noted in olfactory epithelial cells in the anterior portion of the nasal cavity, for example (these cells did not recover within 6 weeks), whereas the damage further back was lighter and more limited. The cells that were most sensitive and affected first were the sustenacular cells (which support the olfactory epithelium).

In another inhalation study, male and female rats were exposed to DBE vapors generated from a liquid mixture of 67% DMG, 17% DMS and 17% DMA, 6 hours/day, 5 days/week for up to 13 weeks. Concentrations were 390, 76 or 20 mg/m^3. After seven weeks, animals in the two higher dose groups (both sexes) showed dose-dependent degeneration (minimal to mild) of olfactory epithelium in the nasal cavities. After 13 weeks of exposure, minimal to moderate degenerative changes

were observed in the nasal cavities (olfactory epithelium) of females at all dose levels and males at the two higher dose levels. The damage was characterized by initial cell death and loss of sustenacular and sensory cells. After an exposure-free period of 6 weeks there were indications that the damaged tissue was regenerating. At the highest dose level there were negative effects on weight gain and lower liver weights in the females (reversible changes) (10).

In a reproduction study (1), female rats were exposed for 6 hours/day during gestation to 990 mg/m^3 vapor/aerosol (about 40/60), or to 380 or 150 mg/m^3 vapor of a DBE mixture (65% DMG, 18% DMS, 17% DMA). Somewhat reduced absolute liver weights were observed in all dose groups (trend, not significant). Animals in the two higher dose groups had reduced food intake during the first 6 days and reduced growth. In another reproduction study, male and female rats (parent generation) were exposed 6 hours/day before, during and after gestation (a total of about 22 weeks) to 1000 mg/m^3 vapor/aerosol or 400 or 160 mg/m^3 vapor generated from an identical DBE mixture (65% DMG, 18% DMS, 17% DMA) (11). From week 7 onward, both males and females in the highest dose group gained weight more slowly. When the animals were killed, however, significantly lower body weights were noted only in females in the highest dose group. Some organ weights were also significantly different in the high-dose group, especially among the females. Liver weights were significantly lower in males (relative liver weights only) and females in both the high and medium dose groups. Histopathological examination of nasal tissue revealed squamous cell metaplasias, especially in olfactory epithelia. Their prevalence and severity increased with dose, and females were more sensitive than males.

A dose-dependent cytotoxicity, expressed as increased leakage of acidic phosphatases, was observed when olfactory and respiratory epithelia from female rats were incubated with DMA, DMG or DMS (10 – 100 mM). All three substances were observed to have significant effects on the olfactory epithelium at concentrations of 25 mM and above, whereas effects on respiratory epithelium were not seen below 50 mM. The toxicity of DMA, DMG and DMS was shown to be dependent on a carboxylesterase-mediated activation. Liberation of acidic phosphatases was reduced when the animals were pre-treated with a carboxylesterase inhibitor (22). No significant elevation of acidic phosphatases was observed when nasal explants from rats were incubated with the metabolite methanol (100 mM) (22), but the monomethyl esters and dicarboxylic acids produced by metabolism of DBE were reported in another in vitro study (23) to stimulate liberation of acidic phosphatases (in tests with 25 or 50 mM). A ranking of the monoesters for cytotoxic potency yielded the result monomethyl adipate > monomethyl glutarate > monomethyl succinate (23).

In a subsequent study (24), olfactory and respiratory epithelia from female rats were examined under optical and electron microscopes after incubation with 10, 25 or 50 mM DMA. At the lowest dose level mild degenerative changes were noted in the respiratory epithelium and more pronounced degenerative changes in the sustenacular cells in olfactory epithelium. The two higher dose levels caused severe

necrotic changes in both respiratory and olfactory epithelia. Respiratory epithelium from animals that had been pre-treated with a carboxylesterase inhibitor showed less severe damage after exposure to 50 mM DMA, but the changes in olfactory epithelium from treated and untreated animals were the same.

A report from the chemical industry (25) describes several laboratory experiments with a mixture of 63% DMG, 20% DMS and 17% DMA. Rabbits were exposed via inhalation (about 15 or 60 ppm vapor for 4 hours), skin (50 or 200 µl) or eyes (10 µl) and effects on eyes were examined. In one of the inhalation experiments (15 ppm, 60 ppm) there was a dose-dependent increase in the incidence of slight conjunctival irritation (slight chemosis: mild reddening). In another inhalation experiment (60 ppm) there was also a single case of moderate irritation of the iris with very slight corneal clouding. Putting the mixture directly into the eyes of the rabbits produced indications of slightly stronger irritation (including slight cloudiness of the cornea). In addition to these effects on the eyes, rabbits exposed to 60 ppm in one of these inhalation experiments showed a slight but significant ($p < 0.05$) increase in the depth of the anterior chamber of the eye 4 hours after exposure, but not on the following day. A slight but significant increase in depth of the anterior chamber was also observed 2 hours after treatment in the animals that had the mixture applied to their skins (200 µl).

When fasting rats were given oral doses of 294 mg DMS (1,4-^{14}C labeled) there was a rapid rise in insulin and a drop in glucose concentration in plasma (16). Another study reports a marked increase of plasma insulin within 2 minutes, but no significant effect on blood glucose, when fasting rats were given DMS intravenously (146 mg/kg body weight) (26). Increased liberation of insulin has also been observed in several in vitro studies when pancreatic islets from rats were incubated with 10 mM DMS (3, 6, 15, 17).

Mutagenicity, carcinogenicity, effects on reproduction

DMS, either with or without addition of metabolizing systems, has shown no mutagenic activity in studies with several strains of Salmonella typhimurium (2, 28). It is reported in an abstract (27) that mutagenicity/chromosome damage did not occur in tests using a mixture of DMG, DMS and DMA, either in bacteria in vitro or with inhalation exposure of mice (micronucleus test). However, it was also reported that, when high concentrations of the mixture were used, chromosome aberrations could be demonstrated in human lymphocytes in vitro – especially lymphocytes from women (significant at concentrations of 3.3 mg/ml and above) (27).

In a reproduction study (1), pregnant rats were exposed to a DBE mixture (65% DMG, 18% DMS, 17% DMA) 6 hours/day on days 7 to 16 of gestation. Concentrations were 990 mg/m^3 (vapor/aerosol, about 40/60), 380 or 150 mg/m^3 (vapor). No exposure-related effects on reproduction were observed (weight, malformations, number of corpora lutea, implantations, resorptions, living young). In another reproduction study (11), male and female rats were exposed to the same mixture (65% DMG, 18% DMS, 17% DMA) before, during and after gestation, a

total of about 22 weeks. Concentrations were 1000 mg/m^3 (vapor/aerosol), 400 or 160 mg/m^3 (vapor). Animals in the parent generation were exposed 6 hours/day, 5 days/week before the breeding period (14 weeks), and thereafter 6 hours/day, 7 days/week during breeding, gestation (days 1 – 19) and nursing (days 4 – 21). Body weights were significantly lower in pups in the high-dose group ($p < 0.05$) at birth and at 21 days of age, and the mothers in this dose group also had lower body weights. No other exposure-related effects (visible aberrations, deviations in organ weights) were observed in fetuses/pups in any group, nor were there any observed effects on other studied reproduction parameters (including fertility, length of gestation, number of living fetuses, litter size, milk production).

In a study (21) in which DMA was given to rats by intraperitoneal injection on days 5, 10 and 15 of gestation (64, 192, 384 or 640 mg/kg) there was a significant increase of aberrations (including hemangiomas, skeletal anomalies) at the two highest dose levels. No such effects were noted at the lowest dose level. This study contains no information regarding effects on the mothers.

Table 1. Effects on experimental animals exposed by inhalation to DMA, DMG and DMS, separately or in mixtures.

Exposure	Species	Effect	Ref.
5900 mg/m^3, 4 hours 66% DMG, 17% DMS, 17% DMA	Rat	Damage to various kinds of cells in nasal mucosa	12
1600 mg/m^3 DMS	Mouse	50% reduction in respiratory rate	20
1000 mg/m^3 6 h/d, 5-7 d/w, 22 weeks before, during and after gestation 65% DMG, 18% DMS, 17% DMA	Rat	Somewhat lower weight gain, increased relative lung and brain weights, lower absolute spleen weight, lower absolute and relative liver weight, minimal to moderate squamous cell metaplasia in olfactory and respiratory epithelia. Pups: lower body weight.	11
990 mg/m^3 6 h/d, days 7-16 of gestation 65% DMG, 18% DMS, 17% DMA	Rat	Somewhat lower absolute liver weight, reduced growth Fetuses: no exposure-related effects	1
910 mg/m^3 DMA	Mouse	50% decrease in respiratory rate	20
890 mg/m^3 DMG	Mouse	50% decrease in respiratory rate	20
610 mg/m^3 57% DMG, 23% DMS, 20% DMA	Mouse	50% decrease in respiratory rate	20

Table 1. Continued

Exposure	Species	Effect	Ref.
590 mg/m^3 63% DMG, 25% DMS, 12% DMA	Mouse	50% decrease in respiratory rate	20
400 mg/m^3 5-7 d/w, 22 weeks before, during and after gestation 65% DMG, 18% DMS, 17% DMA	Rat	Lower relative and absolute liver weights, minimal to moderate squamous cell metaplasia in olfactory epithelium	11
390 mg/m^3 6 h/d, 5 d/w, up to 13 weeks 67% DMG, 17% DMS, 17% DMA	Rat	Minimal to moderate degeneration of olfactory epithelium, lower weight gain, lower liver weight	10
380 mg/m^3, 6 h/d, days 7-16 of gestation 65% DMG, 18% DMS, 17% DMA	Rat	Somewhat lower absolute liver weight, lower growth Pups: no exposure-related effects	1
160 mg/m^3 6 h/d, 5-7 d/w, 22 weeks before, during and after gestation 65% DMG, 18% DMS, 17% DMA	Rat	Minimal to mild squamous cell metaplasia in olfactory epithelium	11
76 mg/m^3 6 h/d, 5 d/w, up to 13 weeks 67% DMG 17% DMS, 17% DMA	Rat	Minimal to mild degeneration of olfactory epithelium	10
20 mg/m^3 6 h/d, 5 d/w, up to 13 weeks 67% DMG, 17% DMS, 17% DMA	Rat	Minimal degeneration of olfactory epithelium	10

Dose-effect/dose-response relationships

There are no data from which to derive a dose-effect or dose-response relationship for humans.

Effects on experimental animals exposed to DMA, DMG and/or DMS are summarized in Table 1. Dose-dependent effects on olfactory epithelium have been observed in rats exposed to a mixture of DMA, DMG and DMS in air concentrations of 20 mg/m^3 and higher.

Conclusions

There are no data from which to determine a critical effect of occupational exposure to DMA, DMG and DMS. Judging from animal experiments, degeneration of olfactory epithelium is the critical effect of exposure to mixtures of the three substances.

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Consensus Report for 1,1,1-Trifluoroethane and 1,1,1,2,2-Pentafluoroethane

February 24, 1999

Chemical and physical data. Uses

1,1,1-trifluoroethane

CAS No.:	420-46-2
Synonyms:	methylfluoroform, HFC143a, FC143a, R143a
Formula:	CH_3CF_3
Molecular weight:	84.04
Boiling point:	- 47.5 °C
Melting point:	- 111.3 °C
Vapor pressure:	1267 kPa (25 °C)
Conversion factors.	1 ppm = 3.49 mg/m^3 (20 °C) 1 mg/m^3 = 0.287 ppm (20 °C)

1,1,1,2,2-pentafluoroethane

CAS No.:	354-33-6
Synonyms:	pentafluoroethane, HFC125, FC125, HFA125, R125
Formula:	CHF_2CF_3
Molecular weight:	120.02
Boiling point:	- 48.5 °C
Melting point:	- 103 °C
Vapor pressure:	1381 kPa (25 °C)
Conversion factors:	1 ppm = 4.98 mg/m^3 (20 °C) 1 mg/m^3 = 0.20 ppm (20 °C)

1,1,1-Trifluoroethane at room temperature is a gas. It is combustible at air concentrations of about 70,000 ppm or higher (2). It is reported to be soluble in ethanol and chloroform (7). 1,1,1,2,2-Pentafluoroethane at room temperature is a colorless, non-combustible gas with low solubility in water (0.97 g/liter) (4, 6). Both substances are used in coolants/refrigerants. 1,1,1,2,2-Pentafluoroethane is also used in fire extinguishers. In 1977, 77 tons of 1,1,1-trifluoroethane (in 5 products) and 104 tons of 1,1,1,2,2-pentafluoroethane (in 16 products) were used in Sweden (product register, National Chemicals Inspectorate).

Uptake, biotransformation, excretion

No studies on the metabolism of 1,1,1-trifluoroethane were found.

The metabolism of 1,1,1,2,2-pentafluoroethane has been studied in experiments with rats. In one study, rats were exposed to up to 50,000 ppm pentafluoroethane 6 hours/day, 5 days/week for 4 or 13 weeks, and it is reported that no increase of fluoride concentration was found in either plasma or urine (6). In another study (5) rats were exposed to 9700 ppm pentafluoroethane for 6 hours, and it was found that the concentration of trifluoroacetic acid in urine was extremely low during the 12 hours immediately following the exposure (40 times lower than after exposure to 11,000 ppm halothane). Slight trifluoracetylation of protein was demonstrated in liver homogenates from the exposed animals (using immunochemical methods). These data suggest that metabolism of 1,1,1,2,2-pentafluoroethane by rats is extremely limited. To the extent that metabolism does occur, it is effected by the P450 system: the pentafluoroethane is probably transformed via a pentafluoroethyl radical to pentafluoroethanol, which may subsequently eliminate hydrogen fluoride and form trifluoroacetyl fluoride. The trifluoroacetyl fluoride may be hydrolyzed, creating trifluoroacetic acid and more hydrogen fluoride. Trifluoroacetyl fluoride can also eliminate fluoride and bind to protein (1, 3, 5).

The reported oil/gas distribution coefficient in vitro for 1,1,1,2,2-pentafluoroethane is 7.3 (9). The reported octanol/water distribution coefficient for 1,1,1,2,2-pentafluoroethane is 1.48 (4).

Toxicity

Human data

There is no information regarding effects of human health attributed to exposure to either 1,1,1-trifluoroethane or 1,1,1,2,2-pentafluoroethane.

Animal data

Rats exposed for 4 hours to 540,000 or 97,000 ppm 1,1,1-trifluoroethane had temporary, dose-dependent weight loss, but there were no deaths. Oxygen concentration in the exposures was about 20% (2). Dogs were given intravenous injections of adrenaline and exposed to 1,1,1-trifluoroethane for 10 minutes: a tendency to cardiac arrhythmia was noted at an air concentration of 300,000 ppm but not at 250,000 ppm or lower (2).

Rats exposed for 4 hours to 800,000 ppm 1,1,1,2,2-pentafluoroethane (20% oxygen by volume) developed ataxia and labored breathing, and their reaction to noise was impaired, but there were no deaths (6). When mice were exposed for six hours to 600,000 ppm (same oxygen concentration), observed effects included low activity, tremor and weight loss (6). When dogs were given intravenous injections of adrenaline and exposed to 1,1,1,2,2-pentafluoroethane for 10 minutes, they showed a tendency to cardiac arrhythmia at air concentrations of 100,000 ppm or higher, but none of the dogs showed a positive response to an exposure of 75,000 ppm under the same conditions (6). Rats exposed to 50,000 ppm

1,1,1,2,2-pentafluoroethane 6 hours/day during gestation had an unsteady gait during the exposures. Rabbits exposed to 50,000 ppm 1,1,1,2,2-pentafluoroethane 6 hours/day during gestation had somewhat slower weight gain and lower feed intake during the first few days of the exposure (6).

Rats were exposed to 1,1,1-trifluoroethane concentrations of 40,000, 10,000 or 2000 ppm 6 hours/day, 5 days/week for 4 weeks or 90 days. Exposures were either "nose only" (4 weeks) or whole-body (4 weeks or 90 days). Clinical signs, body weights, organ weights, hematology, biochemical blood analyses, urine analyses, tissue morphology, β-oxidation activity in the liver and effects on the eyes were studied. No treatment-related effects were seen in the animals after whole-body exposure. Males that had been exposed "nose only" for 4 weeks, however, had recurring weight loss (no change in food intake) and degenerative changes in testes at all dose levels. The authors regarded this as a result of stress caused mostly by extreme temperature conditions. No significant changes were noted in female rats after "nose only" exposures (2).

In another study (6), rats were given whole-body exposures to 50,000, 15,000 or 5000 ppm 1,1,1,2,2- pentafluoroethane 6 hours/day, 5 days/week for 4 or 13 weeks, and sacrificed at termination of exposure or at intervals for up to 4 weeks afterward. Clinical signs, body weights, organ weights, hematology, biochemical blood analyses, urine analyses, tissue morphology, β-oxidation activity in the liver and effects on eyes were studied. No treatment-related effects are reported.

Mutagenicity, carcinogenicity, effects on reproduction

One study (8) reports that 1,1,1-trifluoroethane had mutagenic effects on two of four strains of Salmonella typhimurium when tested in vitro, both with and without metabolic activation. Another study (2) reports that mutagenic effects could not be observed when the substance was tested, either with or without addition of metabolizing systems, on a total of six strains of Salmonella typhimurium and two strains of E coli. In cell transformation tests on mammalian cells in vitro, 1,1,1-trifluoroethane had no observed mutagenic effect (8). There was no significant increase in chromosome aberrations when human lymphocytes were exposed to the substance in vitro (2). Further, there was no significant increase of micronuclei in bone marrow cells of mice that had been exposed to 40,000, 10,000 or 2000 ppm 1,1,1-trifluoroethane 6 hours/day for two days (2).

No mutagenic effect was observed when 1,1,1,2,2-pentafluoroethane was tested in vitro, either with or without metabolic activation, on various strains of Salmonella typhimurium and one strain of E coli (6, 8). In tests on mammalian cells in vitro there was a statistically significant ($p < 0.01$) increase of cells with chromosome aberrations at cytotoxic doses and prolonged exposure times, but not at other doses/exposure times (6). The occurrence of chromosome aberrations did not increase when 1,1,1,2,2,-pentafluoroethane was tested on human lymphocytes in vitro (6). Nor was there a significantly higher frequency of micronuclei in the bone

marrow cells of mice exposed for 6 hours to 600,000, 120,000 or 24,000 ppm 1,1,1,2,2-pentafluoroethane (6).

In one study, a 3% solution of 1,1,1-trifluoroethane in corn oil was given to rats (36 of each sex) by gavage in doses of 300 mg/kg body weight, 5 days/week for 52 weeks. The animals were killed on week 125, and routine histopathological examinations of lungs, liver, spleen, kidneys and brain were made, and other organs were also examined if they looked abnormal. Males had significantly lower body weights from week 28 to week 88. No other exposure-related effects were observed: there was no significant increase of cancer incidence in any organ, for example (8). Shortcomings in the design of the experiment (limited histopathological examinations, brief exposure time, single dose level) make it difficult to draw any definite conclusions from this study.

In a reproduction study (2), rats and rabbits were exposed to 40,000, 10,000 or 2,000 ppm 1,1,1-trifluoroethane 6 hours/day on days 6 – 15 (rats) or 6 – 18 (rabbits) of gestation. A slight but significant increase of visceral and skeletal anomalies (due to retarded development) was observed in the rats, and a slight increase of skeletal aberrations in the rabbits, but, considering the unusually low incidence in the control groups and the lack of a clear dose-response relationship, the effects were judged to be unrelated to the exposures. No significant differences were reported for other reproduction parameters (pre- and post-implantation losses, embryo/fetus deaths, fetus weights), and it was concluded that 1,1,1-trifluoroethane was neither teratogenic nor fetotoxic. No effects on the mothers were observed.

In another reproduction study (6), rats and rabbits were exposed to 50,000, 15,000 or 5000 ppm 1,1,1,2,2-pentafluoroethane for 6 hours/day on days 6 – 15 (rats) or 6 – 18 (rabbits) of gestation. Pre- and post-implantation losses, embryo/fetus death, deformities/anomalies and fetal weights were registered. Transient effects on the mothers (rat: ataxia; rabbit; initial slow weight gain) were observed at 50,000 ppm, but no statistically significant difference in any of the studied reproduction parameters was found in any dose group.

Dose-effect/dose-response relationships

There are no data from which to derive a dose-effect or dose-response relationship for occupational exposure to either of these two substances. Effects on experimental animals are summarized in Tables 1 and 2.

Conclusions

There are no data on human exposures that would serve to define a critical effect for occupational exposure to either 1,1,1-trifluoroethane or 1,1,1,2,2-pentafluoroethane. In animal studies, no effects that can be ascribed with certainty to either substance have been reported at exposures to air concentrations below 50,000 ppm. Effects such as tendencies to irregular heartbeat have been reported at higher experimental exposures.

Table 1. Effects of inhalation exposure to 1,1,1-trifluoroethane observed in experimental animals (from Reference 2)

Exposure	Species	Effects
300,000 ppm, 10 minutes (+ adrenaline i.v.)	Dog	Cardiac arrhythmia
97,000 ppm, 4 hours	Rat	Temporary weight loss
40,000 ppm, 6 h/d 5 d/w, up to 90 days	Rat	No exposure-related effects
40,000 ppm, 6 h/d during gestation	Rat, rabbit	No exposure-related effects
40,000 ppm, 6 h/d, 2 days (micronucleus test)	Mouse	No exposure-related effects

Table 2. Effects of inhalation exposure to 1,1,1,2,2-pentafluoroethane observed in experimental animals (from Reference 6)

Exposure	Species	Effects
800,000 ppm, 4 hours	Rat	Mild ataxia, breathing difficulty, reduced reaction to noise
600,000 ppm, 6 hours (micronucleus test)	Mouse	Tremor, low activity, weight loss, no significant increase of micronuclei in bone marrow
100,000 ppm, 10 minutes (+ adrenaline i.v.)	Dog	Cardiac arrhythmia
50,000 ppm, 6 h/d, 5 d/w up to 13 weeks	Rat	No exposure-related effects.
50,000 ppm, 6 h/d during gestation	Rat, rabbit	Rat: Mild ataxia, no exposure-related effects on young. Rabbit: initial retardation in weight gain, no exposure-related effects on young.

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Consensus Report for Calcium Oxide and Calcium Hydroxide

February 24, 1999

Chemical and physical data. Occurrence

calcium oxide

CAS No.:	1305-78-8
Synonyms:	unslaked lime, caustic lime, quicklime, lime*
Formula:	CaO
Molecular weight:	56.08
Density:	3.25 – 3.38 g/cm^3
Boiling point:	2850 °C
Melting point:	2614 °C
Vapor pressure:	extremely low
Solubility:	disintegrates on contact with water

calcium hydroxide

CAS No.:	1305-62-0
Synonyms:	slaked lime, hydrated lime, lime*
Formula:	$Ca(OH)_2$
Molecular weight:	74.10
Density:	2.24 g/cm^3
Disintegration point:	disintegrates, releasing water, at 580 °C
Vapor pressure:	extremely low
Solubility:	1.7 g/liter water

*lime may refer to either calcium oxide or calcium hydroxide.

Calcium oxide is produced by heating limestone (calcium carbonate) to 950 – 1000 °C in a process called calcining:

$$CaCO_3 + \text{heat} \rightarrow CaO + CO_2$$

Calcium oxide is a white, amorphous substance that crystallizes with the same structure as sodium chloride. Most of it is used in chemical processing. Calcium oxide is used as a slag-forming substance and for production of sodium hydroxide, cement, glass, paper pulp, paper and sugar. Other areas of use are purification of

drinking water and sewage, ore enrichment and refining, and as an earth stabilizer in laying foundations for buildings. Commercial grade calcium oxide is usually about 90 to 95% calcium oxide. The usual impurities are water, calcium carbonate and iron.

Cement is produced by grinding limestone with clay or sand to a fine powder and heating the powder, which becomes a gravel-like material. During this process the calcium oxide reacts with the silicon oxide, forming calcium silicate, the most important component of Portland cement. The material is then ground to powder again, usually with a little gypsum. Addition of water forms calcium silicate hydrate and calcium hydroxide.

Calcium hydroxide is formed when calcium oxide and water are mixed:

$$CaO + H_2O \rightarrow Ca(OH)_2 + \text{heat}$$

This results in the creation of a dry, fine white powder. The process is called slaking. A surplus of water creates solutions of calcium hydroxide, called limewater. Calcium hydroxide is cheap, and is widely used to neutralize acid lakes and farmland. Sugar refining is another area of use.

Mortar is a plastic mixture of calcium hydroxide, sand and water. During the hardening process the calcium hydroxide reacts with the CO_2 in the air:

$$Ca(OH)_2 + CO_2 \rightarrow CaCO_2 + H_2O$$

The quartz in the sand also reacts with the calcium hydroxide, forming silicate, which gives the mortar additional strength.

Other areas of use are in lubricants and pesticides. Commercially available calcium hydroxide is usually 90 to 95% calcium hydroxide. The usual impurities are water, calcium carbonate and iron. Calcium hydroxide is widely used in dentistry for root canal treatments, since its high pH (around 12) neutralizes the acid environment in the tooth and thus has a bactericidal effect. It also has the ability to induce re-mineralization of demineralized dentin (5, 6). Calcium hydroxide is sometimes chewed in various mixtures that may contain tobacco, areca nut or other additives. These mixtures, called pan masala or betel quid, are quite popular in India and the Far East.

The concentration of dust (which consisted mostly of calcium oxide) was measured at a Swedish plant producing paper pulp. The geometric mean for total dust at the monitoring stations was 1.2 (0.1 – 7.7) mg/m^3 before the ventilation system was upgraded and 0.1 (0.1 – 0.2) mg/m^3 afterward. The two monitoring stations closest to the kiln showed the greatest decrease, and the personal monitors showed that the geometric mean for total dust concentration had dropped from 1.2 (0.4 - 5.8) mg/m^3 to 0.2 (0.1 - 0.6) mg/m^3 (25). Personal monitors were also used to measure the average exposure to calcium oxide at a American sugar refinery. Average exposures were 12.9 mg/m^3 around the lime kiln and 4.3 mg/m^3 during handling after the substance had been powdered (15). An unpublished NIOSH

report gives data from an American metal industry where calcium oxide was used as a lubricant in wire drawing: air concentrations ranged from 0.8 to 5.8 mg/m^3 for calcium oxide and from 0.4 to 2.4 mg/m^3 for the fraction in respirable form (9).

Uptake, biotransformation, excretion

Calcium oxide and calcium hydroxide react with skin and mucous membranes on contact by splitting fat and proteins. This facilitates continued penetration into the tissues by the remaining alkalis. Calcium oxide damages mucous membranes and damp skin by generating heat and dehydrating the tissues as the particles react with the moisture in the skin, and by the alkalinity of the calcium hydroxide that is formed in this process (1, 2).

Toxic effects

General effects
Effects on humans:
It is difficult to separate the effects of the two substances, since calcium oxide immediately becomes calcium hydroxide on contact with moisture. Calcium oxide, however, is considerably more irritating than calcium hydroxide. Symptoms involving the skin, eyes and respiratory passages are the predominant effects, and are described below. When the substances are ingested they cause irritation and smarting, and may also cause corrosive damage to the lining of the mouth, throat and esophagus. Stomach cramps, vomiting and in severe cases systemic effects can occur (11, 12). Chronic exposure to calcium hydroxide can result in inflammatory and ulcerous changes in the mouth as well as damage to the digestive tract (18).

Effects in experimental test systems:
The LD_{50} for calcium hydroxide in water given orally to rats is 7.3 (4.8 – 11.1) g/kg (23). In one study reviewed, male rats were given water containing 50 or 350 mg calcium hydroxide/liter. After two months the animals had become restless and aggressive, and their food intake was lower. After three months they had lost weight and had lower hemoglobin values and lower counts of red and white blood cells. Examination of the dead animals showed inflammation and damage in the stomach, small intestine, kidneys and liver (18). Two odontological root-treatment preparations containing up to 25% calcium hydroxide were applied to rat phrenic nerves in vitro. Nerve impulses were blocked within 30 –100 seconds. Exposures of 1.5 and 5 minutes yielded proportional degrees of reversibility, but 30 minutes of exposure resulted in blocking that did not abate during an observation period of a further 30 minutes (4). Another dental compound containing calcium hydroxide as the active component was tested on human cells in vitro: it caused degenerative changes in the cells and changes in various cell components within about a minute (20).

Eyes

Effects on humans:

Exposure to calcium oxide dust and to dust or solutions of calcium hydroxide have similar effects on humans. These strongly irritating and corrosive substances can cause severe damage, particularly to the cornea, resulting in permanent reduction of visual acuity. The severity of the effect is determined by the concentration of the substance, its pH and the exposure time. The hydroxyl ion is considered to be primarily responsible for the damage, which increases steeply between pH 11 and 12. Calcium hydroxide penetrates the epithelium of the cornea more slowly than other alkalis, which may explain why it causes less severe damage.

Slight erosion of the cornea produces a very superficial clouding of Bowman's membrane, which is just below the epithelium. This damage occurs immediately in humans and is particularly common when the epithelium has been damaged and the underlying tissue exposed. More prolonged or intense exposure results in more severe damage, with clouding and penetration even deeper into the supporting tissue (stroma) of the cornea. This occurs immediately at pH 12 or higher, and removes substance from the cornea. Penetration of this extent can also damage the deepest layer (endothelium). With severe corrosive damage the cornea becomes numb for several days. When the damage is less severe, the calcium ion plays a critical role in the clouding of the cornea: a thin layer of calcified material is formed in Bowman's membrane. With more severe damage there may be plaque formation. This type of clouding occurs sporadically in people. In the most severe type of damage, the stroma is completely and permanently eroded. Calcium hydroxide seldom penetrates into the eye far enough to damage the lens and iris, although glaucoma has been reported after severe damage. In a few of the cases it appeared after a few hours, and in others after six to twelve weeks. Simply rinsing the eye after exposure may not remove all the calcium hydroxide. Mechanical cleaning may also be necessary (7).

Effects in experimental test systems, effects on animals:

When the epithelium was removed from the corneas of rabbits, it was found that with up to 10 minutes of exposure and up to pH 11, damage to the stroma from alkalis was moderate and reversible. The degree of damage became much more severe when the pH was increased from 11 to 12, and at higher pH there is extensive breakdown of corneal tissue. In some species there is an immediate and severe clouding of the corneal stroma (7). After rabbits were exposed to a paste of calcium hydroxide for 1 minute, there was a gradual reduction of mucopoly-saccharides in the cornea, most pronounced after 24 hours and still not normalized after 3 months (18). In another study with rabbits, 0.01, 0.03 or 0.10 ml of a 100% calcium hydroxide solution was dropped into one eye and the animals were observed for three weeks. The exposures to the two higher doses had to be terminated after 7 and 14 days because of the severity of the irritation. Furthermore, it was apparent that the damage would be permanent. At the lowest concentration there was some irritation remaining after 3 weeks (8).

Skin

Effects on humans:

Calcium oxide is strongly irritating and corrosive to skin, and can cause open sores. Sores can also result from exposure to wet cement, which contains calcium hydroxide. Three factors contribute to the development of cement burns: alkalinity (a pH around 12), the mechanical abrasion of the particles in the cement, and the duration of contact with the skin. Two to three hours of exposure can cause severe skin damage. A burning sensation may arise, sometimes not until several hours after the contact, and the damage seems to continue even after the area is rinsed. This may be due to the difficulty of getting rid of all the alkali (17, 27). A cement burn often leaves a scar when it heals. In a survey at two cement factories in Australia, it was found that 5 of 117 employees had an occupation-related, non-allergic dermatitis (26).

Effects on animals:

No relevant data were found.

Respiratory passages

Effects on humans:

Calcium oxide is strongly irritating and corrosive to respiratory passages. Inflammation of respiratory passages and ulceration and perforation of the nasal septum, as well as pneumonia, have resulted from inhalation of calcium oxide dust. In an undated publication from the health department in Pennsylvania, it was stated that exposure caused severe nasal irritation at 25 mg/m^3 but not at 9 – 10 mg/m^3 (2). In the previously mentioned Swedish study, fifteen Swedish pulp mill workers exposed to calcium oxide were found to have lower nasal clearance than unexposed controls matched for age, sex and smoking habits (25). The exposed workers showed a significant improvement in nasal clearance after the total dust concentrations in the workplace had been reduced from 1.2 mg/m^3 to 0.1 mg/m^3, and the difference between exposed and unexposed workers disappeared. The improvement was regarded as due mostly to the reduction in dust exposure, although the possibility that it may have been at least partly due to the simultaneous reduction of temperature in the work environment (42 °C to 28 °C) could not be ruled out. There was no observed difference in lung function between the two groups. Air flow through the nose, measured as PEF (Peak Expiratory Flow), was somewhat lower among exposed workers both before and after the plant had been renovated. Nasal inflammation tended to be more common among the exposed workers before the dust concentration was reduced, but there was no statistically significant difference between the groups with regard to symptoms or inflammation markers in nasal lavage fluid. These results should be interpreted with caution, considering the size of the studied group.

An unpublished report from NIOSH states that irritation of nose and throat was a widespread complaint in an industry where measured concentrations of calcium oxide were 0.4 to 5.8 mg/m^3 (9). Studies of cement workers have revealed no

elevation in risk of lung diseases (13, 16). Atrophy of mucous membranes in nose and throat was a common observation among workers in a Polish cement plant (19).

Exposure to calcium hydroxide can trigger symptoms of acute irritation, coughing, pain and possibly chemical burns of mucous membranes. Massive exposure can result in pulmonary edema and shock (18). There are no data on the relationship between symptoms and exposure levels of calcium hydroxide.

Effects on animals
No relevant data were found.

Teratogenicity, mutagenicity, carcinogenicity

Effects on humans
A study of the most common form of cancer among the inhabitants of Papua New Guinea, squamous cell carcinoma in the mouth, showed that the placement of calcium hydroxide and the tumor location were in good agreement (the same side of the mouth in 106 of 162 cases) among betel nut chewers (24).

Studies of cement plant workers revealed elevated risk of cancer in the stomach (16) and caecum (13). Elevated incidences of cancers in lungs, bronchi, trachea, and bladder have been observed among masons (21). These findings might possibly be explained by exposure to quartz and/or chromium.

Effects in experimental test systems, effects on animals:
Calcium hydroxide at a concentration of 800 μg/ml showed no cytotoxic effects on human cells. At 50 - 800 μg/ml there were 20 to 40% more cells after five days of cultivation (compared with untreated cells), and at 100 μg/ml DNA synthesis was 23% higher. No genotoxicity, measured as DNA string breaks, was seen at 3 mg/ml, the highest tested concentration (14). Hamsters exposed to 4 mg calcium hydroxide 5 days/week for up to 14 months showed both hyperplasia and papillary formations in oral mucosa (3). Similar results are reported from other studies, e.g. atypical epithelium in the cheek pouches of hamsters after exposure to calcium hydroxide (10). When calcium hydroxide was applied to mucous membranes in the mouths of rats, it caused various degrees of hyperplasia in all of them and thickening of the mucous membrane (hyperkeratosis) in most of them. However, no malignancy was seen during the 12-month follow-up period (22).

Dose-response/dose-effect relationships

It is difficult to determine the smallest dose that has a definite effect on skin and eyes. Exposure of skin or mucous membranes often results in ulceration. The corrosive damage and penetration into the skin are considerably worse if the pH is above 11. Skin exposed to cement for two or three hours can develop severe chemical burns. Reduced nasal clearance was found in workers exposed to dust

consisting primarily of calcium oxide, where the geometric average for total dust was 1.2 mg/m^3 (25). The available literature supplies no further basis for determining a dose-response relationship for either calcium oxide or calcium hydroxide.

Conclusions

Direct contact with calcium oxide dust and dust or solutions of calcium hydroxide can cause severe chemical burns, especially of the eyes but also of skin and mucous membranes. The critical effects of exposure to calcium oxide and calcium hydroxide are reduced nasal clearance and irritation of the respiratory passages. Calcium oxide is the more irritating of the two substances. No conclusions can be drawn regarding the carcinogenic effects of either substance.

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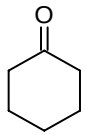
Consensus Report for Cyclohexanone

February 24, 1999

This report is based on two documents previously published here (4, 54) and on research published since 1987. It contains some supplementary information from older literature.

Chemical and physical data. Uses

CAS No.:	108-94-1
Synonyms:	cyclohexylketone, ketohexamethylene, pimelin ketone
Formula:	$C_6H_{10}O$
Structure:	



Molecular weight:	98.14
Density	
liquid:	0.95 (20 °C)
vapor:	3.4 (air = 1)
Boiling point:	155.6 °C
Melting point:	- 32.1 °C
Flash point	
open cup:	44 °C
closed cup:	54 °C
Vapor pressure:	0.69 kPa (25 °C)
Distribution coefficients	
olive oil/air:	3.83 (30 °C) (1)
olive oil/water:	0.79 – 1.23
Conversion factors:	1 ppm = 4 mg/m^3 (25 °C, 101.3 kPa)
	1 mg/m^3 = 0.249 ppm (25 °C, 101.3 kPa)
Odor threshold:	0.88 ppm (2)

Cyclohexanone is an alicyclic ketone which at room temperature is a colorless, oily liquid. It has an odor reminiscent of peppermint and acetone. Cyclohexanone is used in the production of polyamide (nylon) and as a solvent for paint, enamel,

printing ink, rubber, wax, various kinds of resin (vinyl, cellulose acetate, nitrocellulose), glue, insecticides and polyvinyl chloride (PVC). PVC is widely used in medical equipment and supplies, and there is a risk that residual amounts of cyclohexanone can escape from the polymer. Small amounts of cyclohexanone have been identified in intravenous solutions stored in bags made of PVC plastic (30, 57, 62), and leaching from dialysis tubes has also been reported (58). The use of cyclohexanone as a solvent in the production of audio and video tapes has increased in recent years. Cyclohexanone has been identified as an emission product in the thermal breakdown of PVC at 170 °C, and it is therefore possible that electromagnetic welding of PVC produces emissions of cyclohexanone (3). Cyclohexanone is also used as a monomer in the production of cyclohexanone resin.

Uptake, biotransformation, excretion

Uptake

It has been demonstrated in animal experiments that cyclohexanone is taken up by the lungs, digestive tract and skin. The blood/air distribution coefficient is 2150, implying high uptake via respiratory passages (26). This was confirmed in an inhalation study with human subjects, which reports a relative uptake during rest of 57 – 59% (40). Skin uptake of liquid cyclohexanone by persons (n = 3) who held one hand immersed up to the wrist in pure solvent for 30 minutes was estimated to be 1 – 2% of the dose absorbed by inhalation during 8 hours of exposure to a cyclohexanone concentration of 200 mg/m^3 (50 ppm) (40).

Biotransformation and excretion

In inhalation studies in which eight human subjects were exposed to 100 – 400 mg/m^3 cyclohexanone for 8 hours, 1,2- and 1,4-cyclohexanediol in urine accounted for 56 – 62% of the amount of uptake, and only 1% was excreted as cyclohexanol (40). Similar results were obtained when three volunteers were exposed to 415 mg/m^3 for 8 hours. Cyclohexanol and 1,2-cyclohexanediol were excreted primarily as glucuronides, whereas the 1,4-diol was excreted only in unconjugated form. The study also indicated that cyclohexanediol in urine occurs almost entirely as *trans* isomer (12). Repeated exposure to cyclohexanone vapor (200 mg/m^3, 8 hours/day, 5 days) resulted in a gradually increasing excretion rate for cyclohexanediol, at least for the first few days (40).

The urine of newborn babies that had been given intravenous nutrient solutions contaminated with cyclohexanone contained mostly unconjugated *trans*-1,2-cyclohexanediol, with small amounts of the 1,3- and 1,4-isomers. The *cis* form of 1,2-cyclohexanediol was also detected in a few samples (38).

In contrast to the studies described above, large amounts of glucuronidized cyclohexanol were seen in a case of acute poisoning. The half time for cyclohexanol in the patient's plasma was 4.75 hours (50). Cyclohexanol has also been identified in the urine of workers occupationally exposed to cyclohexanone (43, 44).

It has been proposed that the primary metabolic pathway in man is initial reduction to cyclohexanol by alcohol dehydrogenase, followed by cytochrome P-450 mediated hydroxylation to cyclohexanediol, which is excreted in urine (38). Available data do not indicate whether the glucuronide of the 1,2-diol is formed exclusively by glucuronidation of the diol or if oxidation of glucuronidized cyclohexanol occurs as well.

In laboratory animals, cyclohexanone is reduced to cyclohexanol, which conjugates with glucuronic acid and is excreted in urine. Rabbits excreted 66% of an oral dose as glucuronides, mostly of cyclohexanol (11). In beagles, the total excretion of cyclohexanol accounted for 74 – 100% of the given dose, and most of it (60% of the dose) was glucuronide. Less than 1% of the dose was excreted in urine as unconjugated cyclohexanone and cyclohexanol. Repeated exposure to cyclohexanone had no effect on the fraction of the dose that was transformed to cyclohexanol (34). When Wistar and Gunn rats were given an intravenous infusion of 50 mg/kg, conjugates of cyclohexanol accounted for 15 – 20% (Wistar) and 19 – 29% (Gunn) of the dose. After infusions of 100 mg/kg the conjugates accounted for 17 – 25% and 24 – 34% respectively (17). After oral administration of cyclohexanone to rabbits (1.9 mmol/kg) and rats (2.5 mmol/kg), trace amounts of *cis*-2-hydroxycyclohexyl mercapturic acid were found in their urine (28).

When beagles were given bolus doses of cyclohexanone by intravenous infusion (284 mg/kg/day, 18 days), cyclohexanone in plasma followed a two-compartment model with half times of 6.6 minutes and 81 minutes (31).

Since alcohol dehydrogenase catalyzes both the reduction of cyclohexanone to cyclohexanol and the oxidation of ethanol to acetaldehyde, interaction between ethanol and cyclohexanone can be expected. A man who drank 720 ml sake (10% ethanol w/v), mixed with about 40 g cyclohexanone and some acetone and methyl ethyl ketone, showed unusually rapid elimination of the ethanol and low levels of cyclohexanone in plasma (50). Rabbits that were given cyclohexanone and ethanol (4.8 mmol/kg) either separately or together showed accelerated metabolism of both substances when they were given simultaneously (51). This is probably because metabolism of one of the substances generates the right oxidation state of the cofactor NAD^+ / NADH for metabolism of the other substance.

Cyclohexanone induces the cytochrome P-450 system and has been shown to potentiate the hepatotoxicity of other chemicals (5).

Toxic effects

Human data

About ten people, both men and women, were exposed to cyclohexanone vapor for 3 to 5 minutes and asked to assess the degree of eye, nose and throat irritation and say whether they believed they could work in that atmosphere for 8 hours. The results were reported as the concentrations that a majority of the subjects considered irritating/acceptable. Irritation of eyes, nose and throat was reported after exposure to 75 ppm. Exposure to 50 ppm “was not tolerable,” mostly because of the

resulting throat irritation. The highest concentration judged acceptable for the proposed 8-hour exposure was 25 ppm (42).

Allergic contact eczema has been reported after exposure to paint containing cyclohexanone resin, but cyclohexanone alone showed no sensitizing ability when tested on guinea pigs (6). A later case report, however, describes allergic contact eczema caused by cyclohexanone (53).

Two cases of poisoning have been reported. The first was a man who drank 720 ml sake (10% alcohol) mixed with 100 ml of a liquid cement for PVC containing 39% cyclohexanone, 18% acetone and 28% methyl ethyl ketone. The man lost consciousness and was found to have metabolic acidosis and low blood oxygen. During the following days the man's blood-sugar level rose – probably caused by the acetone – and later he also had elevated serum levels of transaminases (liver enzymes). The kinetics of the three solvents and the time the man was unconscious indicate that cyclohexanol was the probable reason for the loss of consciousness. Whether the man recovered or not is not mentioned (50).

A 15-year-old boy who drank cyclohexanone became mentally affected and went into shock, with metabolic acidosis, chemical hepatitis, renal insufficiency and muscular degeneration (pain, elevated serum levels of creatine phosphokinase, myoglobulinuria) (63).

Workers (n = 75) in a furniture factory who coated wood with cyclohexanone reported symptoms such as mood swings, irritability, forgetfulness, insomnia and headaches more often than controls (n = 85). Irritation of eyes, upper respiratory passages and skin were also more common among the exposed workers than among controls, as were aching joints, muscles and bones. The statistical analysis of the symptom data (if one was made) is not given. Significant effects on both the central and peripheral nervous system were found. Reduced conductivity was observed in all three studied nerves (medianus, ulnaris and peroneus). Latency time and amplitude were also affected. As to CNS function, there were delayed reaction times to both visual and auditory stimuli. The average exposure time was 14 ± 3 (± SD) years (39). According to the authors, exposure measurements were made at the beginning, middle and end of twelve consecutive 8-hour shifts and in different places where cyclohexanone was used: the measured air concentrations ranged from 162 to 368 mg/m^3 (40 – 92 ppm). There may have been exposure to other substances. The control group was matched for socioeconomic factors, workload and shift (39).

Workers (n = 23) who had been exposed for 4 years to cyclohexanone (150 – 630 mg/m^3 or 37 – 158 ppm; unclear what the interval refers to), methyl ethyl ketone and minor amounts of toluene and acetone, showed declines in alertness and verbal memory on tests of cognitive performance (37).

A case report describes a 58-year-old man with lifelong exposure to solvents, mostly cyclohexanone, white spirit and isopropanol. For nearly 30 years the man had had repeated seizures resembling epilepsy, with reduced consciousness and occasionally a total loss of consciousness. There were never any convulsions. The man's exposure to solvents stopped, and the seizures stopped within a year. Four

years later the man was again exposed to high solvent levels (not further described) for three days and had a seizure resembling those he had had earlier (27).

Anosmia (loss of the sense of smell), irritation of mucous membranes, loss of appetite, headaches, dizziness, concentration difficulty and alcohol intolerance were reported in a man who had been exposed for 20 years to dichloromethane, tetrahydrofuran, methyl ethyl ketone, cyclohexanone and small amounts of dimethylformamide. The inability to discern odors lingered for 15 months after exposure was terminated (41).

Animal data

The acute toxicity of cyclohexanone is low or moderate. Reported LD_{50} values are in the range 0.9 – 2.2 g/kg (20, 29, 61), depending on species and method of administration. When rats were exposed to 4000 ppm for 4 hours, all of them died (n = 6) (56). In a later study with rats, 4000 ppm was reported to be the level at which mortality was first observed ("approximate lethal concentration") (29). Simultaneous administration of oxime (not specified) increased the toxicity of cyclohexanone, measured as LD_{50}, for female rats (23). Oxime occurs together with cyclohexanone in the production of caprolactam, which is transformed to polyamide fiber by heating.

Female guinea pigs (n = 10) exposed to 4000 ppm for 6 hours showed effects on respiration and declining rectal temperature, and after about 1.5 hours of exposure declining pulse rate also (59).

When cyclohexanone was given to dogs in intravenous infusions (284 mg/kg/day, 18 – 21 days) it affected respiration, caused "vocalization," stimulated urine production, and caused salivation, watering of the eyes and ataxia (loss of muscular coordination). The effects intensified with increased concentration and infusion rate. Despite the indications of CNS effects, no damage to the brain could be detected histologically. The dogs given the dose as a 6% solution, 75 ml/minute, had to be killed after 18 days. Examination of these animals revealed metabolic acidosis, elevated absolute and relative liver weights with depletion of glycogen in the liver, plasma cell infiltrate around hepatic veins, hemosiderin deposits (accumulation of iron outside the blood) in the liver, spleen and lymph nodes, as well as mild extramedulary blood formation and bone marrow hyperplasia. An increase in the number of white blood cells, reduction in the number of erythrocytes and reduced hemoglobin contents were also observed, and in some cases elevated concentrations of hemoglobin in plasma. These findings, taken together with the enlarged and thickened epithelial cells and the protein droplets in the kidneys, indicate hemolysis. There were no pathological findings in the liver, kidneys or spleen in the groups of dogs given the same daily dose in a more diluted solution (0.75%) (31).

High doses of cyclohexanone (1000 – 1600 mg/kg) given orally to rabbits caused pronounced damage to lungs, heart muscle, liver, spleen and kidneys. Repeated skin application of high doses of cyclohexanone caused a pronounced drop in rectal temperature, convulsions and narcosis (61).

Slight narcosis, effects on respiration, ataxia and increased salivation were observed in rabbits exposed to 3000 ppm cyclohexanone vapor 6 hours/day, 5 days/week for 3 weeks. Eye irritation was observed at 300 ppm, but not at 190 ppm (10 weeks). After exposure to 190 ppm, low-grade degenerative changes were observed in liver and kidneys. There was no observed effect on hemoglobin levels or on number of red or white blood cells at any dose level (60).

Mice exposed to 19,000 mg/m^3 cyclohexanone vapor (4730 ppm) for up to 2 hours developed edema and local hemorrhages in the lungs, respiratory effects and CNS depression. Average survival time was about 100 minutes. The mice that survived the exposure were sacrificed 7 days later: histological examination revealed hyperplasia in the white pulp of the spleen. The same study reports a concentration-dependent weakening of contraction ability in isolated perfused rabbit heart after exposure to 1.93 – 19.3 mM (perfusate, 30 minutes). Cyclohexanone in oil instilled in one eye (5 – 40%) or applied to the skin under occlusion (12.4 – 99%) resulted in concentration-dependent irritation (rabbits) (20).

Groups of 10 newborn rats were given repeated intravenous injections of cyclohexanone (1, 10, or 25 mg/kg/day, 18 days): there were no observed histopathological changes in brain, heart, lungs, liver, spleen, kidneys, eyes, stomach, caecum or duodenum that could be related to the cyclohexanone. Nor were there any observed effects on clinical chemical or hematological parameters (16).

Male rats (Wistar and Gunn) were given cyclohexanone (0, 50 or 100 mg/kg/day) by repeated intravenous infusion for 28 days: no effects were noted on weight gain, hematological or clinical chemical parameters, other than significantly lower levels of serum calcium. No macroscopic (heart, lungs, liver, spleen and kidneys) or histopathological (spleen, skeletal muscles, kidneys, lungs, liver, stomach, duodenum, pancreas, bladder, brain and eyes) changes were reported. Nor were there any observed effects on the lens of the eye (17).

In another study, however, lens clouding was reported in 3 of 12 guinea pigs after repeated application of cyclohexanone directly to the skin. No such effects were noted in the controls (49). This result, however, could not be confirmed in a later study with rabbits and guinea pigs (18). The latter study reports changes in the lens after intravenous (0.5 or 5 mg/kg) and percutaneous (0.5 ml) administration (3 times a week for 3 weeks) of cyclohexanone in all guinea pig groups including the controls. There were no changes in exposed rabbits. The authors conclude that the changes observed in the guinea pigs are natural to the species and make guinea pigs unsuitable experimental animals in this context.

No damage to the peripheral nervous system could be demonstrated in experimental animals given cyclohexanone intraperitoneally in doses of 200 mg/kg, twice a day, 5 days/week for 13 weeks (46).

The duration of an electrically stimulated muscle contraction in male rats after 4 hours of exposure to cyclohexanone vapor was compared with an unexposed control group, and the air concentration that shortened the duration by 30% was determined to be 440 ppm. Female mice were exposed for 2 hours and the time to

maximum contraction after electrical stimulation was measured. Exposure to 490 ppm reduced the speed to contraction by 30%, i.e. latency time increased (13).

The mechanism behind cyclohexanone's neurotoxicity was investigated by studying its ability to induce or prevent convulsions. Intraperitoneal administration of cyclohexanone did not cause convulsions in female mice – it had an anti-convulsive action. Cyclohexanone impeded both chemically and electrically induced convulsions. The doses that had effect in 50 % of animals (ED_{50}). were 238 mg/kg (chemical induction) and 397 mg/kg (electrical induction). Cyclohexanone also acted as a competitive inhibitor for a ligand specific for picrotoxin receptors, which suggests that cyclohexanone has its effect via this mechanism (24).

Adult male rats exposed to 8 ppm cyclohexanone constantly for 10 weeks (45) and young rats exposed to 2 ppm constantly for 3 weeks (48) developed morphological changes in cells in their olfactory bulbs. Exposure to completely odorless air caused similar changes in adult animals, though not so pronounced. The result should be regarded as an adaptation to a restricted olfactory environment.

Cyclohexanone (0.01 M) reduced the uptake of ^{3}H-thymidine by human lymphocytes in vitro (47) and was cytotoxic to mouse fibroblasts in vitro (20). A concentration of 0.02 M in the culture medium reduced cell growth by 50%.

Mice exposed for 4 hours to cyclohexanone vapor (184, 255, 282, 334 or 577 ppm) showed changed behavior in swimming tests immediately following the exposures. The effect was dose-dependent and reversible (9). This performance measure does not differentiate between effects on the central nervous system and those on other organs.

Teratogenicity mutagenicity, carcinogenicity

Teratogenicity

Cyclohexanone given intraperitoneally to female mice (50 mg/kg/day, 28 days) had no observable effect on their fertility (21).

Sprague-Dawley rats were exposed to 100, 250 or 500 ppm cyclohexanone vapor 7 hours/day on days 5 to 20 of gestation: birth weight, sex distribution, resorptions and fetal deaths were not significantly different from controls. No significant increase in the frequency of aberrations was noted, although there was a weak increase in the proportion of rudimentary ribs per litter in the groups exposed to 250 and 500 ppm. In the absence of conventional indications of embryotoxicity, the small number of deformities noted in the cyclohexanone group was judged to be unrelated to the exposure (52).

Cyclohexanone given orally to pregnant mice in doses of 800 mg/kg/day on days 8 –12 of gestation had no effect on litter size or weight of pups on days 1 and 3 (7), nor on their performance in a maze on days 21, 58 and 200 (15). Cyclohexanone given orally to pregnant mice in doses of 2200 mg/kg/day on days 8 –12 of gestation caused significantly lower birth weights in a screening test with pregnant mice (n = 28). The treatment was lethal for 6 of the 28 mice (55).

Development of fertile chicken eggs exposed to an unspecified concentration of cyclohexanone vapor was retarded, and the neuromotor ability of the chicks was worse than that of controls (19).

When female mice were given 1% cyclohexanone in food during gestation and lactation, their pups were smaller and mortality was higher (14).

Mutagenicity, carcinogenicity

Cyclohexanone was not mutagenic in tests with Salmonella typhimurium (22) or in the L5178Y tk^+ / tk^- mouse lymphoma cell forward mutation assay, either with or without addition of metabolic systems (36). In another study, however, cyclohexanone was positive in Ames tests with Salmonella typhimurium. The number of forward mutations in Bacillus subtilis was also elevated (35). This study has been criticized on several points: the frequency of spontaneous mutations was elevated, the number of controls small and the procedures done only once (10).

Cyclohexanone (0.1 – 10 mM) has induced chromosome aberrations in human leukocytes in vitro (8, 32).

In a 2-year study with rats and mice, elevated incidence of some cancer forms was reported in exposed animals. Since the expected dose-response relationship did not appear, the evidence of carcinogenicity was judged to be minimal and the effect weak to nonexistent (33).

In 1989 the IARC stated in its assessment that cyclohexanone was “not classifiable as to its carcinogenicity to humans” (Group 3). Evidence of carcinogenicity to experimental animals was judged to be inadequate. There were no human data (25). No subsequent studies were found in the literature.

Dose-response/dose-effect relationships

Dose-response and dose-effect relationships observed in experimental animals exposed to cyclohexanone (inhalation and other means of administration) are summarized in Tables 1 and 2.

Occupationally exposed persons experience effects on the nervous system and eye, nose and throat irritation at air concentrations of 40 to 92 ppm. A majority of subjects exposed to cyclohexanone vapor for 3 to 5 minutes reported throat irritation at 50 ppm, and at 75 ppm irritation of eyes and nose as well. The highest concentration the majority of them judged tolerable for a hypothetical 8-hour working day was 25 ppm.

Rats showed no effects at 100 mg/kg. In a study of rabbits, marginal liver and kidney effects were observed at 190 ppm (760 mg/m^3) and eye irritation at 300 ppm (1200 mg/m^3).

Table 1. Dose-effect and dose-response relationships observed in experimental animals exposed to cyclohexanone by inhalation.

Species	Exposure (ppm)	Exposure time	Effects	Ref.
Mouse	4730	104 min.	2 of 5 animals died	20
Mouse	4730	78 min	CNS effects, irritation, respiratory effects, pulmonary edema	20
Guinea pig	4000	6 hours	Depressed respiration, pulse and rectal temperature; 3 of 10 died after 4 days.	59
Rat	4000	4 hours	All 6 animals died	56
Rat	2000	4 hours	1 of 6 died	56
Rabbit	3000	90 hours (6 h/d, 5 d/w, 3 w)	2 of 4 animals died; slight narcosis, depressed respiration, ataxia, salivation, irritation and blood vessel dilation in conjunctiva, weight loss	60
Rabbit	1400	300 hours (6 h/d, 5 d/w, 10 w)	Slight lethargy, conjunctivitis as above	60
Rabbit	760	300 hours (6 h/d, 5 d/w, 10 w)	Somewhat increased salivation, slight conjunctivitis	60
Monkey	600	300 hours (6 h/d, 5 d/w, 10 w)	Slight conjunctivitis	60
Rabbit	300	300 hours (6 h/d, 5 d/w, 10 w)	Very slight dilation of blood vessels in conjunctiva	60
Rabbit	190	300 hours (6 h/d, 5 d/w, 10 w)	Barely discernible degenerative changes in liver and kidneys	60
Mouse	184-575	4 hours	Effects on performance in swimming test	9

Table 2. Dose-effect and dose-response relationships observed in experimental animals exposed to cyclohexanone by means other than inhalation.

Species	Dose (mg/kg/d)	Administration method	Effects	Ref.
Rabbit	1600-1900	oral	Narcosis	61
Mouse	2200	oral	6/28 pregnant mice died. Pups had low birth weights	55
Mouse	800	oral	No effect on locomotor activity of pups	15
Mouse	800	oral, days 8-12 of gestation	No effect on litter size or weight of pups	7
Rat	200	i.p. 5 d/w, 13 weeks	No effect on neural conductivity in peripheral nerves	46
Dog	284	i.v. 18-21 days 6% solution 75 ml/minute	CNS effects that increased with concentration and rate of administration; respiratory effects, elevated liver and adrenal weight, metabolic acidosis	31
Dog	284	i.v. 18-21 days 0.75% solution 5 ml/minute	Lethargy, dilated pupils	31
Rat	0, 50, 100	i.v. 28 days	No histopathological changes in brain, no effects on clinical/chemical parameters	17
Rat	1, 10, 25	i.v., 18 days	No effects on hematology, chemistry, histopathology or organ weights	16
Rabbit	12.4-99%	epidermal	Concentration-dependent skin irritation, from mild to very severe	20
Rabbit	5-40%	instillation in eyes	Concentration-dependent irritation, mild to severe	20
Rabbit	2.5%	instillation in eyes	No eye irritation	20

Conclusions

The critical effect of occupational exposure to cyclohexanone is judged to be its effect on the nervous system. Occupationally exposed persons (40 – 92 ppm) report symptoms including CNS effects and irritation. Throat irritation was observed in volunteers exposed to 50 ppm for a few minutes.

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Consensus Report for Some Lactate Esters

June 2, 1999

This report is based primarily on a criteria document compiled jointly by the Nordic Expert Group and the Dutch Expert Committee (9). That document, in turn, is based on two recently published survey articles (5, 6). The Criteria Group published an earlier consensus report for lactate esters in 1995 (8). The present report takes up the following lactate esters: methyl lactate, ethyl lactate, isopropyl lactate, isobutyl lactate, n-butyl lactate, 2-ethylhexyl lactate, myristyl lactate and cetyl lactate. Some information on a few of the other lactate esters is given in the criteria document (9).

Lactates occur in two enantiomorphous (mirror image) forms, D (*dextro*) and L (*levo*). The forms often occur together: the DL (*dextrolevo*) form. 2-Ethylhexyl lactate also has diastereoisomeric forms.

Chemical and physical data. Uses

methyl lactate

CAS No.:	547-64-8 (DL form)
	27871-49-4 (L form)
Formula:	$C_4H_8O_3$
	$CH_3CH(OH)COOCH_3$
Molecular weight:	104.1
Melting point:	- 66 °C
Boiling point:	144 °C
Flash point:	57 °C
Density:	1.092 g/ml (20 °C)
Vapor pressure:	0.34 kPa (20 °C)
	23 kPa (100 °C)
Saturation concentration:	3302 ppm (20 °C)
Distribution coefficient:	log $P_{o/w}$ = - 0.53
Conversion factors:	1 ppm = 4.3 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.23 ppm (20 °C, 101.3 kPa)

Methyl lactate is a colorless, transparent liquid. It mixes with water at room temperature, and is also soluble in alcohol and ether. It is used as a solvent for cellulose acetate.

ethyl lactate

CAS No.:	97-64-3 (DL form)
	687-47-8 (L form)
Formula:	$C_5H_{10}O_3$
	$CH_3CH(OH)COOCH_2CH_3$
Molecular weight:	118.1
Melting point:	- 25 °C
Boiling point:	153 °C
Flash point:	61 °C
Density:	1.033 g/ml (20 °C)
Vapor pressure:	0.22 kPa (20 °C)
	17 kPa (100 °C)
Distribution coefficient:	log $P_{o/w}$ = 0.06
Conversion factors:	1 ppm = 4.9 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.20 ppm (20 °C, 101.3 kPa)

Ethyl lactate at room temperature is a colorless liquid with a mild, characteristic odor. The reported odor threshold is 0.89 mg/m^3, and the odor becomes irritating at 65 mg/m^3 (5). Ethyl lactate mixes with water, alcohols, ketones, esters, hydrocarbons and ethers. It occurs naturally, most notably in several different kinds of fruit. It is used as a solvent for nitrocellulose, cellulose acetate and many cellulose ethers. It is an ingredient in enamels, paints, polishes and various cosmetic products. It is used as a replacement for trichloroethylene in de-greasing and cleaning (9). An air concentration of 0.6 ppm was measured at a Swedish company that used ethyl lactate for de-greasing metal. There were peaks of 10 ppm around some operations, and an 8-hour average was calculated to be 4.2 ppm (4).

isopropyl lactate

CAS No.:	617-51-6 (DL form)
	63697-00-7 (L form)
Formula:	$C_6H_{12}O_3$
	$CH_3CH(OH)COOCH(CH_3)_2$
Molecular weight:	132.2
Boiling point:	157 °C
Flash point:	60 °C
Density:	0.991 g/ml (20 °C)
Vapor pressure:	0.17 kPa (20 °C)
	15 kPa (100 °C)
Distribution coefficient:	log $P_{o/w}$ = 0.39
Conversion factors:	1 ppm = 5.5 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.18 ppm (20 °C, 101.3 kPa)

Isopropyl lactate is soluble in water, alcohol, ether and benzene.

isobutyl lactate

CAS No.:	585-24-0 (DL form)
	702-84-0 (L form)
Formula:	$C_7H_{14}O_3$
	$CH_3CH(OH)COOCH_2CH(CH_3)_2$
Molecular weight:	146.2
Boiling point:	182 °C
Flash point:	76 °C
Density:	0.979 g/ml (20 °C)
Vapor pressure:	0.05 kPa (20 °C)
Distribution coefficient:	log $P_{o/w}$ = 1.10
Conversion factors:	1 ppm = 6.1 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.165 ppm (20 °C, 101.3 kPa)

Isobutyl lactate is soluble in water: 5.1 g/100 ml at 20 °C.

n-butyl lactate

CAS No.:	138-22-7 (DL form)
	34451-19-9 (L form)
Formula:	$C_7H_{14}O_3$
	$CH_3CH(OH)COO(CH_2)_3CH_3$
Molecular weight:	146.2
Melting point:	- 43 °C
Boiling point:	187 °C
Flash point:	79 °C
Density:	0.984 g/ml (20 °C)
Vapor pressure:	0.03 kPa (20 °C)
	4.7 kPa (100 °C)
Distribution coefficient:	log $P_{o/w}$ = 1.10
Conversion factors:	1 ppm = 6.1 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.165 ppm (20 °C, 101.3 kPa)

At room temperature, n-butyl lactate is a watery liquid with a mild odor. It mixes with water (4.5 g/100 ml), alcohol, ether and many solvents. In acids and alkalis it is hydrolyzed to lactic acid and butyl alcohol. The odor threshold is reported in one source (5) to be 0.095 mg/m^3, and in another (2) to be 7 ppm (42.6 mg/m^3). The reported irritation threshold for the odor is 9 mg/m^3. Butyl lactate has been used as a solvent for synthetic polymers, enamels, paints etc. It occurs in low concentrations (< 0.03%) in cosmetic products (9).

2-ethylhexyl lactate

CAS No.:	6283-86-9 (DL form)
	186817-80-1 (L form)
Formula:	$C_{11}H_{22}O_3$
	$CH_3CH(OH)COOCH_2CH(C_2H_5)(CH_2)_3CH_3$
Molecular weight:	202.3
Boiling point:	246 °C
Flash point:	113 °C
Density:	0.940 g/ml (20 °C)
Vapor pressure:	0.002 kPa (20 °C)
	0.6 kPa (100 °C)
Distribution coefficient:	log $P_{o/w}$ = 3.17
Conversion factors:	1 ppm = 8.4 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.12 ppm (20 °C, 101.3 kPa)

For 2-ethylhexyl lactate, the reported solubility in water is 30 mg/100 ml. The reported odor threshold is 0.45 mg/m^3 and the reported discomfort threshold is 40 mg/m^3 (5). Ethylhexyl lactate has been used as a degreaser.

myristyl lactate

CAS No.:	1323-03-1 (DL form)
Formula:	$C_{17}H_{34}O_3$
	$CH_3CH(OH)COO(CH_2)_{13}CH_3$
Molecular weight:	286.5
Density:	0.892 – 0.904 (25 °C)
Conversion factors:	1 ppm = 11.9 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.08 ppm (20 °C, 101.3 kPa)

Myristyl lactate occurs as a white to yellowish liquid or a soft solid. The substance dissolves in ethanol and propylene glycol but is insoluble in water and glycerin. Myristyl lactate is used as a softener in several cosmetic products, usually in concentrations of 5 to 10% (6).

cetyl lactate

CAS No.:	35274-05-6 (DL form)
Formula:	$C_{19}H_{38}O_3$
	$CH_3CH(OH)COO(CH_2)_{15}CH_3$
Molecular weight:	314.4
Melting point:	23 – 41 °C
Boiling point:	170 °C (at 2 x 10^{-3} kPa)
Density:	0.893 – 0.905 (25 °C)
Conversion factors:	1 ppm = 13.05 mg/m^3 (20 °C, 101.3 kPa)
	1 mg/m^3 = 0.077 ppm (20 °C, 101.3 kPa)

Cetyl lactate is a white to yellow, soft, waxy substance with a faint, pleasant and easily recognizable aroma. It is soluble in ethanol and propylene glycol. Cetyl lactate is used as a non-ionic softener in pharmaceuticals and cosmetics, usually in concentrations of 1 to 5% (9).

Uptake, biotransformation, excretion

There are no quantitative data on uptake of lactate esters. Twenty-four hours after ^{14}C-labeled ethyl lactate was applied to the skin of rats, the radioactivity was seen in sebaceous glands, hair follicles, epidermis and dermis (11).

Hydrolysis of lactate esters to lactic acid and alcohol has been reported to occur after both skin application and oral administration (3). Lactic acid is a naturally occurring metabolite, and its toxicity is mostly a result of its acidity. Concentrated lactic acid is irritating to skin and eyes. In in vitro studies, 80% of ethyl lactate in rat plasma was hydrolyzed within 60 minutes. Similar results have been demonstrated in homogenates of nasal mucosa, liver and skin from rats (5).

There are no quantitative data on excretion of lactate esters. Since hydrolysis of lactate esters is fairly rapid, elimination pathways are probably the same as those for lactic acid and alcohol.

Toxic effects

Human data

One case of allergic contact dermatitis has been reported. An acne medication (gel) containing 10% ethyl lactate caused acute erythema (skin reddening). Tests six weeks later yielded a positive response to the gel and to 1% ethyl lactate in petroleum jelly (10).

Unpublished reports on n-butyl lactate, cited in the ACGIH document, state that occupational exposure to 43 mg/m^3 (about 7 ppm) with brief peaks of about 67 mg/m^3 (11 ppm) caused headache, coughing and irritation of mucous membranes. Some symptoms also occurred with exposure to 24 mg/m^3 (4 ppm), whereas no symptoms resulted from exposures below 8 mg/m^3 (1.4 ppm). A later report, also unpublished, states that 7 ppm had a definitely unpleasant odor but caused no harm and was judged acceptable (1).

Animal data

Lactate esters dropped into the eyes of rabbits caused eye irritation. Tested esters that yielded a positive response were ethyl, n-propyl, n-butyl, lauryl, and myristyl lactate. Methyl lactate, however, was classified as non-irritating (5, 6, 7, 12).

A 50% solution of ethyl lactate caused no irritation when applied to the skin of rabbits, but undiluted butyl lactate caused mild to moderate reddening. Mild skin irritation can be caused by cosmetics containing up to 12% lauryl lactate or myristyl lactate (5).

The calculated RD_{50} for mice is 750 – 800 mg/m^3 for ethyl lactate and butyl lactate (5). (RD_{50} is the dose that causes a 50% reduction in respiratory rate.)

Cosmetics containing small amounts of lauryl lactate or cetyl lactate were tested on the skin of guinea pigs (the Magnusson-Kligman test). Both substances were judged to be non-sensitizing (6).

In inhalation studies, groups of rats (both sexes) were exposed 6 hours/day, 5 days/week for 28 days. Exposure levels for ethyl lactate were 0, 25, 75, 150, 200, 600 or 2500 mg/m^3. In the two highest dose groups there were degenerative changes in olfactory epithelia with hyperplasia in goblet cells. Similar effects were observed at high doses when the animals were exposed to isobutyl lactate or n-butyl lactate. The NOAEL for these three lactates is 200 mg/m^3. Further information is presented in Table 1 (9).

At the same laboratory, rats exposed by inhalation to 2-ethylhexyl lactate showed the same effects even at 75 mg/m^3, the lowest tested dose. The effects were the same whether the ester was in gas or aerosol phase. See also Table 1 (9).

Groups of rats (both sexes) were given myristyl lactate in oral doses of 0, 0.5, 2.5 or 5.0 mg/kg body weight, 5 days/week for 13 weeks. Elevated relative liver weights and enlargement and thickening of the walls of the stomach and duodenum were seen in rats in the two higher dose groups. Histological examination revealed diffuse hyperplasia in the mucous membranes of the duodenum. These changes were not observed at the lowest dose (6).

Mutagenicity, carcinogenicity, teratogenicity

Ethyl lactate has been tested on several different strains of Salmonella typhimurium, both with and without metabolizing systems. No mutagenic activity was observed. No mutagenic activity was observed when 2-ethylhexyl lactate was tested on Salmonella and E. coli (5).

No carcinogenicity studies were found on any of these lactate esters.

Ethyl lactate (doses of 0, 517, 1551 or 3619 mg/kg body weight/day) was applied to the skin of rats on days 6 – 15 of gestation. There was slight reddening at the site of application, but no clinical effects were seen in the mothers and there were no observed effects on the young (5).

Pregnant rats were exposed to an aerosol of 2-ethylhexyl lactate (0, 200, or 600 mg/m^3) 6 hours/day on days 6 to 15 of gestation. Animals in the high-dose group had lower food intake, but showed no toxic effects. Retarded ossification was noted in pups in both dose groups, but was attributed to stress rather than the toxicity of the exposure (5).

Dose-response/dose effect relationships

For most of the lactate esters, there are no data from which to derive a dose-effect or dose-response relationship. Data on human exposures are particularly sparse.

Data from inhalation exposure studies with rats are presented in Table 1.

Myristyl lactate given orally to rats increased liver weights at a daily dose of 2.5 mg/kg body weight or higher. The NOAEL in this study was 0.5 mg/kg body weight (6).

Table 1. Effects of some lactate esters on rats exposed 6 hours/day, 5 days/week for 28 days (from References 5 and 9).

Lactate	Exposure mg/m^3	Exposure ppm	Effect
ethyl (gas)	2500	500	Reduced growth, lower absolute liver weights, reduced food intake, elevated blood glucose, degenerative changes in olfactory epithelium, hyperplasia in goblet cells.
	600	120	Degenerative changes in olfactory epithelium, hyperplasia in goblet cells.
	200	40	NOAEL
n-butyl (gas)	600	99	Slight focal hyperplasia in nasal epithelium.
	200	33	NOAEL
isobutyl (gas)	800	132	"Disarrangement" of olfactory epithelium, hyperplasia in respiratory epithelium in nose.
	400	66	Hyperplasia in respiratory epithelium in nose.
	200	33	NOAEL
2-ethylhexyl (aerosol)	1800	216	Histopathological changes in nose, larynx, trachea and lungs; peroxisome proliferation.
	600	72	Histopathological changes in respiratory passages.
	200	24	Histopathological changes in respiratory passages.
	75	9	Histopathological changes in nasal cavity; LOAEL.
(gas)	75	9	Focal hyperplasia in respiratory epithelium in nose; LOAEL

Conclusions

The critical effect of occupational exposure to lactate esters is judged to be irritation of mucous membranes. The similarities in responses to the different esters implies that lactic acid is probably the underlying reason for the effects. For rats, the NOAEL for several tested lactate esters is 200 mg/m^3. For 2-ethylhexyl lactate, effects can be observed at an exposure as low as 75 mg/m^3. According to unpublished data on human subjects exposed to n-butyl lactate, effects appear at exposure to 7 ppm (43 mg/m^3) or higher.

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Consensus Report for Ethylene Glycol Methylether and Ethylene Glycol Methylether Acetate

June 2, 1999

This report is based on a criteria document published in Arbete och Hälsa (22).

Chemical and physical characteristics. Uses

Ethylene glycol methylether (EGME)

CAS No.:	109-86-4
Synonyms:	glycol methylether, 2-methoxyethanol, methyl glycol
Formula:	$CH_3–O–CH_2–CH_2–OH$
Molecular weight:	76.09
Density:	0.96 (20 °C)
Boiling point:	124 °C
Melting point:	- 85.1 °C
Vapor pressure:	1.3 kPa (9.7 mm Hg) (20 °C)
Evaporation rate:	0.5 (butyl acetate = 1)
Saturation concentration:	12,800 ppm (25 °C)
Relative density:	2.6 (air = 1)
Conversion factors:	1 ppm = 3.11 mg/m^3 (20 °C) 1 mg/m^3 = 0.322 ppm (20 °C)

Ethylene glycol methylether acetate (EGMEA)

CAS No.:	110-49-6
Synonyms:	ethylene glycol monomethylether acetate 2-methoxyethyl acetate, methyl glycol acetate
Formula:	$CH_3–O–CH_2–CH_2–O–CO–CH_3$
Molecular weight:	118.13
Density:	1.005 (20 °C)
Boiling point:	145 °C
Melting point:	- 65 °C
Flash point:	55.6 °C (open cup)
Vapor pressure:	0.27 – 0.50 kPa (2.0 – 3.7 mm Hg) (20 °C)
Evaporation rate:	0.3 (butyl acetate = 1)
Saturation concentration:	3100 – 6000 ppm (25 °C)
Relative density:	4.07 (air = 1)
Conversion factors:	1 ppm = 4.90 mg/m^3 (20 °C) 1 mg/m^3 = 0.200 ppm (20 °C)

EGME and EGMEA at room temperature are flammable, volatile, clear liquids with a weak, sweetish odor and bitter taste. Both substances dissolve readily and completely in water as well as polar and non-polar solvents.

EGME is produced by a reaction between methanol and ethylene oxide. EGMEA is produced from EGME by conventional estrification. Known impurities in EGME are reported to be < 0.1% methanol, < 0.1% diethylene glycol methylether and < 0.02% ethylene glycol (22).

EGME and EGMEA do not occur in nature. World-wide reported uses for the two glycol ethers are in paints and enamels; printer's ink; plastic packaging for foodstuffs; pigments for silk-screen printing; photographic and photolithographic processes (including the production of offset plates); CDs, circuit boards and integrated circuits; cleaners for household and industrial use; and antifreeze in hydraulic fluids and airplane fuel. In 1993 there were 23 Swedish chemical products containing EGME, and total annual use was about 260 tons of the pure substance. EGMEA was not listed in the product register. EGME was used primarily as a solvent, but it was also an ingredient in paints and enamels. In 1997 there were 27 Swedish products containing EGME, and annual use was 19 tons, most of which was used as photoresist in the telecommunications industry. EGMEA was a listed ingredient in 3 products, with an annual use of less than 0.1 ton. In 1994 EGME and EGMEA were classified by the EU as toxic to reproduction and their use in consumer products was prohibited (22).

Reported average exposure levels are in the range <0.1 to 23 mg/m^3 for EGME, and from <0.1 to 143 mg/m^3 for EGMEA. Exposure has been reported from semi-conductor and circuit board manufacture, printing, painting (especially automobile and ship painting), furniture finishing, paint production and automobile repair (22). No data were found on exposures in Swedish workplaces.

Uptake, biotransformation, excretion

As suggested by the chemical structure and the ready solubility of EGME and EGMEA, both substances are efficiently absorbed via all paths of uptake and rapidly distributed throughout the body. Uptake via respiratory passages has been measured at 76% of the amount inhaled (22).

Uptake of EGME by frozen and thawed human epidermis in vitro was 2.8 mg/cm^2/hour (12). An average absorption rate of 2.9 mg/cm^2/hour, with large inter-individual variations, was measured for liquid EGME in an in vivo study with volunteers. Exposure of hands and lower arms to EGME in liquid form was calculated to yield an absorption rate 100 times that of exposure to 5 ppm in the air. The authors also calculated that, with whole-body exposure to EGME vapor, 55% of total uptake occurs via the skin (24).

EGME is distributed fairly evenly between blood and other tissues, with the exception of low solubility in adipose tissue. Methoxyacetic acid (MAA) also has relatively even distribution in body tissues (22).

EGMEA is efficiently hydrolyzed to EGME by the carboxylesterases in nasal mucosa, liver, kidneys, lungs and blood. The most important metabolic pathway for EGME is oxidation via methoxyacetaldehyde (MALD) to MAA. This metabolism can be inhibited by ethanol, and the importance of alcohol dehydrogenase is illustrated by the fact that metabolism of EGME is almost completely suppressed in rats that have been pre-treated with pyrazol. When men were exposed to 5 ppm EGME for 4 hours (resting), an estimated 86% of the inhaled amount of EGME was excreted in urine as MAA. The reported half time for MAA in human urine is 77 hours. The half time for MAA in serum and plasma has been reported to be about 6 hours for mice and 20 hours for monkeys (22).

In addition to MAA, methoxyethyl glucuronide, methoxyethyl sulfate, ethylene glycol, glycolic acid, glycine, methoxyacetyl glucuronide, methoxyacetyl glycine, methoxycitrate and methoxybutenic acid were identified in nucleomagnetic resonance spectrometry (NMR) analysis of urine samples from mice and rats given doses of ^{13}C -labeled EGME. Simultaneous administration of acetate, an endogenously formed substance and a precursor in the Krebs cycle, increased the proportion of EGME-related metabolites and reduced the proportion of MAA-related metabolites. These results show that ether cleavage can occur, and also that EGME after oxidation can form methoxyacetyl-coenzyme A. It has been suggested that this "false substrate" in the Krebs cycle may be related EGME's toxic effects on reproduction (22).

Mechanism studies

EGME had no effect when incubated with human erythrocytes, whereas 0.5 mM of MAA increased their osmotic fragility. When human erythrocyte membranes (ghosts) were incubated with MAA or EGME, membrane-bound acetylcholinesterase (IC_{50} = 5.5 mM) and ATPase (IC_{50} = 1.4 mM) were inhibited by MAA but not by EGME (26).

Simultaneous administration of a number of other substances (formate, acetate, glycine, glucose, serine, sarcosine) involved in the formation of pyridine and purine – which in turn are needed for synthesis of DNA and RNA – reduces or completely eliminates the malformed sperm and disruption of spermatogenesis caused by EGME in experimental animals (22).

Addition of 10 μM MAA, but not 1 μM, reduced the proliferative capacity of fetal mouse liver cells in vitro, observed as reduced incorporation of tritium-labeled thymidine. No effect on survival of the cells was observed, however (20).

Toxic effects

Animal data

EGME and EGMEA have moderate acute toxicity. The reported LD_{50} values for EGME range from 0.9 to 3.4 g/kg body weight, depending on species and method of administration. The reported LC_{50} for inhalation is 4600 mg/m^3 (1480 ppm).

Four hours of exposure to 1000 ppm resulted in atrophied sperm in male rats, and 625 ppm produced damaged spermatids within 24 hours. The reported LD_{50} values for EGMEA range from 1.3 to 5.6 g/kg. The reported LD_{50} for MAA with oral administration (in water) is 1 to 1.5 g/kg (22).

Short-term exposures via gavage, skin application, feed and inhalation have similar effects in several species, including reduced thymus, spleen and testes weights, lower counts of white and red blood cells and platelets, lower hematocrit, hemoglobin levels and bone marrow cellularity, higher numbers of immature granulocytes and disturbance of spermatogenesis. Spermatogenesis is disrupted at a particular phase, the late pachytene, and the effect shows up later in lower sperm counts or aspermia. Toxicity is about the same regardless of the method of exposure – gavage, drinking water, skin application or inhalation (22).

After tests with rabbits, EGME and EGMEA were classed according to EEC criteria as non-irritating to skin, and EGME as non-irritating to eyes (22).

Human data

Older studies report that repeated occupational exposure to products containing EGME can cause headaches, weakness, dizziness, ataxia, toxic encephalopathy and dampened reflexes (22). Further case reports are summarized in Table 1.

In a cross-sectional study of 65 workers who produced and packaged EGME, measured concentrations in workplace air were 4 to 20 ppm and personal monitors indicated 5.4 to 8.5 ppm (time-weighted averages). Trends (not significant) to lower leukocyte counts and lower Hb were seen in the 40 exposed workers when they were compared with the 25 unexposed workers. Closer study of a sub-group of 6 exposed and 9 unexposed workers showed tendencies to reduced leukocyte counts, lower hemoglobin, reduced testicle size, lower sperm counts, elevated levels of luteinizing hormone (LH) and lower levels of testosterone and follicle-stimulating hormone (FSH) in serum, none of which was statistically significant (7).

Of 73 painters at a shipyard, 10% had anemia and 5% had granulocytopenia, compared with 0% in an unexposed control group. No other hematologic differences between the groups were observed. Measured exposure levels were 0 – 5.6 (mean 0.8, median 0.4) ppm for EGME and 0 – 21.5 (mean 2.6) ppm for ethylene glycol ethylether (EGEE). A review of patient journals revealed that the conditions had arisen during employment as painters. The authors listed about 60 substances that painters at a shipyard might be exposed to, and of these lead, benzene and glycol ethers were identified as potentially harmful to blood-forming organs. All blood-lead levels were below 40 µg/dl and most of them were below 20 µg/dl. Air monitoring and product reviews indicated negligible exposure to benzene, and the authors concluded that the observed hematologic effects could not be explained by exposure to lead or benzene (33).

No reports on skin irritation, eye irritation or sensitization were found in the literature.

Mutagenicity

With the exception of the studies reviewed below, EGME and its metabolite MAA have been negative in all genotoxicity studies, including Ames tests, in all tested Salmonella strains, both with and without addition of metabolizing systems (for a survey see Reference 25).

EGME caused mutations in the gpt gene in a cell line from Chinese hamsters, but no mutations in the hprt gene in another cell line. The metabolite MALD was weakly mutagenic in Salmonella (TA97a) and gave rise to an increased number of mutations, sister chromatid exchanges and chromosome aberrations in Chinese hamster cells in vitro, in the concentration interval 5 – 40 mM. Chromosome damage from MALD was also seen in human lymphocytes after 1 hour at 40 mM and after 24 hours at 2.5 mM. No chromosome damage was observed in mice that had been given up to 1000 mg/kg MALD or up to 2500 mg/kg EGME by gavage (22).

EGME and its metabolites were tested for genotoxicity and epigenetic effects in different test systems. Increased numbers of micronuclei and mitotic irregularities were seen in vitro at 65 mM EGME, 0.12 mM MALD and 3.2 mM MAA. With MALD, an elevated frequency of mutations was seen at 1 – 10 mM, of sister chromatid exchanges and chromosome aberrations at 0.1 – 1 mM, and of morphological transformations at 0.1 – 0.3 mM. The authors regard the results as weakly positive for EGME and MAA and clearly positive for MALD (13).

EGMEA was tested with a number of Salmonella strains, both with and without metabolic activation, in two different laboratories. One judged it to be weakly mutagenic, and the other judged it to be possibly mutagenic (36).

Carcinogenicity

There are no reports of animal studies on the carcinogenicity of EGME or EGMEA. In McGregor's assessment, aside from a few positive results in Ames tests (see Mutagenicity), there is no experimental evidence that either of the substances is carcinogenic (25).

A review of 198 cases of acute myelotic leukemia (AML) was made in a French case-control study. Blind estimates of exposure to different types of glycol ethers and potential exposure levels were made by an expert panel. No relationship between AML and glycol ethers was seen (21).

Reproduction toxicity

Animal data

A large number of animal studies have provided a clear and unambiguous picture of the toxic effects on reproduction in both sexes. Males given low doses have reduced testes weights, histological changes in testes and low sperm counts; higher doses cause testicular atrophy and aspermia. The effects are temporary. Females have lower fertility and higher numbers of dead and resorbed fetuses. Young have lower post-natal survival rates and higher frequencies of skeletal anomalies, malformed

extremities and severe deformities. Effects on fetuses appear at doses too low to have visible effects on the mothers. At higher doses there is 100% fetal mortality. The degree of fetal damage is extremely sensitive to the time of exposure. These effects have been demonstrated for all administration methods and in several different species (22). A few of the more recent studies are summarized below.

Rabbits were given EGME in drinking water, 12.5 – 50 mg/kg/day, 5 days/week for 12 weeks: there were dose-dependent declines in several parameters of sperm quality. The effects were significant at 37.5 and 50 mg/kg, and the most marked effect was reduced number of sperm per ejaculate. Histologically, spermatogenesis (number of round spermatids per Sertoli's cell) was somewhat reduced at 25 and severely disrupted at 37.5 mg/kg. At 50 mg/kg spermatogenesis ceased almost completely in 5 of 7 rabbits. No effects were seen on the libido or fertility of the rabbits that still had functional sperm production, and no other pathological or histopathological effects were observed. The authors concluded that spermatogenesis in rabbits is about 10 times more sensitive to EGME than that in rats or mice (4, 16).

EGME given to female rats in doses of 300 mg/kg/day completely eliminated the estrus cycle: inhibition of ovulation, luteal body hypertrophy, permanently elevated progesterone levels and permanently low levels of estradiol, follicle-stimulating hormone (FSH), luteinizing hormone (LH) and prolactin. Addition of MAA to luteal cells in vitro resulted in elevated progesterone levels in the cultivation medium at all levels; 1 mM was the lowest tested concentration (9). MAA was also tested in an in vitro system with luteinized granulosa cells from humans. Incubation with 0 – 5 mM of MAA for 6 to 48 hours yielded a duration- and concentration-dependent increase of progesterone. The effect was significant at 1 mM, but the tendency was also apparent at 0.1 and 0.5 mM. The authors associate these observations with the effects on menstrual cycle and ovarian function in humans (1).

Pregnant monkeys (*Macaca fascicularis*) were given EGME by gavage in doses of 12, 24, or 36 mg/kg/day on days 20 to 45 of gestation (during organogenesis). Effects on the mothers (moderate to extreme loss of appetite, weight loss) were observed at all dose levels. Most of the animals in the two higher dose groups were therefore given nourishment and/or electrolytes by gavage. Caesarean sections were performed after 100 days: all 8 fetuses in the high-dose group were dead or resorbed, as were 3 of 11 in the middle group and 4 of 14 in the low-dose group. This can be compared with 0 of 6 in the untreated control group and 0 of 3 in a control group treated with ethanol (0.47 mmol/kg/day). The authors noted that the dead fetuses looked different from those seen in spontaneous, drug-induced or ethanol-induced fetal deaths, and drew the conclusion that the effect was not secondary to the maternal toxicity but a direct effect of the EGME. One of the dead fetuses in the highest dose group had no digits on the forelimbs. Deformities of this type had not previously been seen in monkeys, but had been observed earlier in mice and rabbits given EGME. Other deformities were also observed, but the

possibility that these were secondary to the death of the fetus could not be ruled out. No deformities were seen in the living fetuses (31).

Human data

In examination of sperm quality in painters at a shipyard, it was found that they had oligospermia (10/79 compared to 0/40) and azoospermia (4/79 compared to 0/40) more often than controls, as well as a tendency to a lower number of sperm per ejaculate. Average exposures of the painters were 0.8 ppm for EGME and 2.6 ppm for EGEE. Urine analyses for ethoxyacetic acid indicated considerable skin exposure (34).

A women whose job during two pregnancies was to rinse laboratory glassware in EGMEA had in both cases boys with genital defects (hypospadia, micropenis, bifid type of scrotum). The authors could find no other factors in the work environment, the home environment or heredity that could explain them (5).

There is a report on 44 patients in Matamoros, Mexico, who had a syndrome with characteristic facial distortions and mental retardation. All of them were born in 1971 – 1977 and were children of mothers who during their pregnancies had worked at a factory making condensers. There are no quantitative data on exposure, but during work the women had dipped their hands into a solution consisting mainly of EGME and ethylene glycol. There was no ventilation, and workers used no protective gloves or face masks. Indications of acute poisoning, with fatigue, dizziness, nausea and vomiting, had occurred during work. A closer examination of 28 of the cases revealed that all of them also had musculoskeletal defects and that about half of them had eye and ear defects as well. There was no familial relationship between the cases and birth defects of this nature had not occurred previously in any of the affected families (30).

An elevated frequency of spontaneous abortions (compared with unexposed controls) was noted in women in the semiconductor industry, and was associated particularly with diffusion/dipping and photolithography. (For a brief description of the production of integrated circuits, see e.g. Britannica Online (6)). Exposure to glycol ethers, xylene, toluene and hexamethyl disilazane was reported to occur during photolithographic work, and to arsine, phosphine and diborane during diffusion work. No exposure measurements were made (27). This report served to initiate several further epidemiological studies in the semiconductor industry.

In a cohort study of 6088 women employed in 14 semiconductor factories, 904 pregnancies and 113 miscarriages were examined. After control for age, smoking habits, ethnic background, education, income, date of pregnancy and stress levels, there was a tendency for women in production work to have a higher proportion of miscarriages than other employees. A significantly higher frequency was seen among women who worked with masking. In this group, the highest risk of miscarriage was associated with etching (3). In a sub-study, the outcomes of 891 pregnancies were sorted according to exposure during the first trimester. Women working with photolithography, who were exposed to ethylene glycol ethers

(EGME, EGEE and their acetates), fluorides and other substances, had a significantly higher risk of miscarriage (32).

In a prospective study made at the same companies, 403 women were followed for 6 months by analysis of chorionic gonadotropin in urine. After control for possibility of conceiving, use of contraceptives and age, there was significantly lower fertility in the women working in dipping and the same tendency in those exposed to glycol ethers (14). In addition, female production workers had a significantly higher risk of spontaneous abortions than those who held other types of jobs. All three pregnancies among the women who were exposed to ethylene glycol ethers terminated in miscarriages (14). The same group of women also kept diaries on their menstruations. Prolonged menstrual cycles were seen in women who worked with dipping, and shortened cycles and a greater number of irregular menstruations were seen in the photolithography group (17).

In exposure assessments made in the workplaces at the same time, it is stated that 15 – 20% of the factories used photochemicals (negative photoresist), usually containing 3% EGME. All personal monitors registered EGME levels below 10 ppb, average exposure to ethylene glycol ethylether acetate (EGEEA) was 22 ppb, and exposure to 1-methoxypropyl acetate was 8 ppb (18). Exposures to glycol ethers were strongly correlated to exposures to xylene and n-butyl acetate (19).

In a study of 454 pregnancies among 1368 women employed in the semiconductor industry, risk of spontaneous abortion tended to be higher for those who worked in chip production, and those with chemical exposure outside of chip production, than for unexposed subjects. The proportion of stillbirths also tended to be higher in the two exposed groups. The authors report that chip production involves exposure to glycol ethers and a number of other solvents, which they listed, but exposures were not measured (29).

Another study in the semiconductor industry covered both female employees (561 pregnancies) and the wives of male employees (589 pregnancies). For the female employees, those with the highest likelihood of exposure to ethylene glycol ethers had significantly reduced fertility and elevated risk of spontaneous abortion. No increased risk of miscarriage was found among the wives of employed men, but there was a tendency to lower fertility. No personal monitoring measurements were taken, and only general information on exposures is given. A few measurements yielded glycol ether levels below 0.2 ppm in the highest exposure group. The glycol ethers named in the study are diethylene glycol dimethylether (DEGDME) and ethylene glycol ethylether acetate (EGEEA); EGME was not mentioned. Simultaneous exposure to glycol ethers and hexamethyl disilazane occurred. No increase in frequency of the studied effects was noted with exposure to n-butyl acetate, N-methyl-2-pyrrolidone or xylene, unless there was simultaneous exposure to glycol ether (8).

None of the epidemiological studies made in the semiconductor industry contains detailed information on exposure levels. About 400 air samples were analyzed in a separate study, and average levels of 0.1 ppm EGME and 0.01 ppm EGMEA were

found (28). The studies are also alike in that the authors claim there was exposure to no factors, other than glycol ethers, known to have toxic effects on reproduction.

Immunotoxicity

Animal data

All clinical, morphological and histological indications of leukemia disappeared in male rats that had been given subcutaneous injections of human leukemia cells when they were given drinking water containing 2.5 mg/ml EGME. Addition of 0.25 mg/ml, equivalent to a daily dose of 15 mg/kg, halved the leukemia response. EGEE also retarded the leukemia response, but was ten times less potent than EGME. Seven other tested glycols and glycol ethers had no effect. In vitro tests with the same cell line showed a concentration-dependent reduction in number of cells in the dose interval 1 – 100 μM EGME. The metabolite MAA was about half as effective, which the authors regard as an indication that the mitosis-inhibiting effect of EGME is not due to a cytotoxic mechanism alone (11).

Mice given EGME by gavage in doses of 500 or 100 mg/kg/day for 5 to 10 days developed atrophy and decline in mature thymocytes in the thymal cortex, but the medulla was unaffected (23).

Female rats were exposed to EGME in drinking water, 2000 or 6000 mg/liter (equivalent to 161 or 486 mg/kg/day), for 21 days: the treatment resulted in a dose-dependent reduction of thymus weight, increased activity of killer cells, reduced antibody production and a lower number of cells in the spleen. At 6000 mg/l there was also reduced production of gamma interferon. Male rats exposed to 1600 or 4800 ppm in drinking water (200 or 531 mg/kg/day) showed all these effects as well as reduced testes weights at both dose levels. Thymus atrophy and reduced interleukin-2 production were also seen at the higher dose (15).

Single oral doses of 125 or 500 mg/kg caused a 3 or 8 times higher apoptosis index (programmed cell death) in the thymus, compared with unexposed rats. There was a parallel increase in the liver's capacity to metabolize MALD to MAA. Pre-treatment with phenobarbital suppressed this effect almost completely (2).

Immune response was studied in rats and mice that had been given EGME, EGMEA, MALD or MAA in oral doses of 50 to 400 mg/kg/day for 10 days. In the rats, the four substances yielded similar immunosuppression, expressed as reduced thymus and spleen weights and reduced antibody plaque-forming cell (PFC) response. The effects were significant at the lowest dose level, and equimolar doses of the four substances produced equivalent immunosuppression. Pre-treatment with 4-methylpyrazole caused these effects to disappear, which indicates that metabolic activation is necessary. This immunosuppression was observed in all of the rat strains but in none of the mouse strains. Nor did MAA in subcutaneous doses of up to 1920 mg/kg/day produce immunosuppression in the mice, which indicates that the difference between the species can not be explained by differences in bioavailability or metabolic rate (22).

Atrophy, dose-dependent reduction in cellularity, and changes in thymocyte patterns indicating disturbances in thymocyte maturation were observed in thymus glands from the young of mice given EGME in doses of 100 – 200 mg/kg/day on days 10 to 17 of gestation (20).

There are a number of other animal studies that support these findings on the immunotoxic effects of EGME (22).

Human data

Effects on several kinds of leukocytes were observed in 9 floorlayers when they were compared with an unexposed, matched control group. The changes comprised reduced numbers of eosinophils and segmented neutrophils and increased numbers of rod neutrophils and lymphocytes. Among the lymphocytes there were lower numbers of T cells and helper cells, but higher numbers of NK and B cells. According to the authors, this lymphocyte pattern resembles the one seen in immune-deficiency diseases. Tendencies to lower hemoglobin values and lower numbers of erythrocytes were also observed. The floorlayers were exposed to a number of solvents, including EGME (mean 6.1, maximum 150 mg/m^3), EGEE, EGBE, butanol, isobutanol, toluene, xylene, methyl ethyl ketone and methyl isobutyl ketone. Solvent levels in blood indicated that EGME was the predominant exposure (10).

Dose-effect/dose-response relationships

The addition of 10 μM MAA reduced mitosis in fetal liver cells in vitro (20). According to a toxicokinetic model, this level is equivalent to 8 hours of inhalation exposure to 1 ppm EGME (35).

Increased fetal death was observed in monkeys given EGME in oral doses of 12 mg/kg/day during gestation (31). At 25 mg/kg/day there were effects on testes and sperm in rabbits (4, 16), and in rats prolonged gestation, smaller litters and deformed pups. Doses of 50 mg/kg/day affect the thymus, suppress the immune response, are toxic and teratogenic to fetuses of rodents and completely eliminate spermatogenesis in rabbits. At doses around 100 mg/kg/day these effects are considerably stronger, and bone marrow depression, disturbances in hematopoiesis and reduced fertility are also seen. Inhalation exposure to 50 ppm has fetotoxic effects on rodents, producing skeletal aberrations and deformities. It is noteworthy that most of these effects are seen in all species and with all methods of administration, although the necessary exposure level varies. These variations can probably be partly explained by differences in study design. It is therefore difficult to give a single critical effect. Immunological effects of inhalation exposure have not been studied with modern methods.

Relationships between occupational exposure and effects are given in Table 1. Because the exposure situations are not fully known – particularly with regard to skin exposure – it is difficult to draw quantitative conclusions on a dose-effect or dose-response relationship from the existing data on human exposures.

Table 1. Effects on human health associated with occupational exposure to EGME.

Level ppm	Exposure situation	Number of persons	Observed effects	Ref.
mean 0.8 median 0.4	Shipyard painters; high skin exposure, including EGEE (mean 2.6 ppm)	73 men	10% had anemia and 5% had granulocytopenia (0% in controls). Low sperm counts.	33, 34
mean 2 peak 48	Floorlayers; also exposed to EGEE and other solvents	9 men	Higher numbers of rod neutrophils, lymphocytes, NK and B cells. Lower numbers of eosinophils, segmented neutrophils, T cells and helper cells. Tendencies to lower Hb values and erythrocyte counts.	10
5 – 9	Production and packaging of EGME	65 men	Tendencies to lower white blood cell counts, Hb values, testicle size, sperm counts and testosterone and FSH levels in serum. Tendency to elevated serum level of leuteinizing hormone (studied in different sub-groups).	7
about 8	Manual cleaning, skin exposure	2 men	Bone-marrow depression, pancyto-penia	22
18 – 58	Production and cleaning of microfilm	1 man	Apathy, fatigue, increased need for sleep, low counts of red and white blood cells and platelets, low Hb and hematocrit.	22
60 – 4000 (reconstruc-tion)	Printshop cleaning; large surfaces, skin exposure	6 men	Symptoms of poisoning, CNS effects. Hypocellular bone marrow (examined in only one of the men)	22

Effects on blood composition, immune system, testes and spermatogenesis have been seen with occupational exposure to EGME at air levels of 0.4 to 10 ppm, combined with unknown, but probably quite high, skin exposure. These observations are in good agreement with the results of animal experiments, and should be attributed to EGME even though other agents can not be unequivocally ruled out.

Two case reports (5, 30) contain descriptions of 44 and 2 children with birth defects born to mothers who were massively exposed to EGME during their pregnancies, and where there seems to be no other explanation for the deformities.

Effects on the menstrual cycle, reduced fertility and higher frequencies of spontaneous abortions have been seen among women production workers in the semiconductor industry. These observations are also in accordance with the effects of EGME observed in laboratory animals, and ethylene glycol ethers are presented by

the authors as the only plausible agents that could be identified. The importance of EGME relative to that of other glycol ethers is unclear. To the extent that exposure to EGME occurs, the air levels in this context should be below 1 ppm.

Conclusions

Judging from both animal data and experience of occupational exposures, the critical effects of ethylene glycol methylether (EGME) are its toxic effects on reproduction and blood formation.

EGME and its acetate ester EGMEA are efficiently absorbed via both inhalation and skin exposure. Skin penetration can account for a large portion of the total uptake if the skin is exposed to liquids or vapors containing EGME or EGMEA. EGMEA is rapidly transformed to EGME in the body, and in animal experiments the two substances are equally toxic. The health hazards of EGMEA should therefore be regarded as equivalent to those of EGME.

Effects on blood composition, testes and spermatogenesis have been observed in men occupationally exposed to EGME at air levels of 0.4 to 10 ppm, probably combined with considerable skin exposure. Severe birth defects and disturbances in hematopoiesis have been associated with occupational exposure to EGME and EGMEA at unknown, but probably high levels. Daily oral doses of 12 mg/kg body weight caused fetal death in monkeys, and 25 mg/kg/day reduced spermatogenesis in rabbits.

Several studies report elevated incidences of miscarriage, menstrual irregularities and low fertility among women in the semiconductor industry. The importance of EGME in relation to other agents is not clear.

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Consensus Report for Thiourea

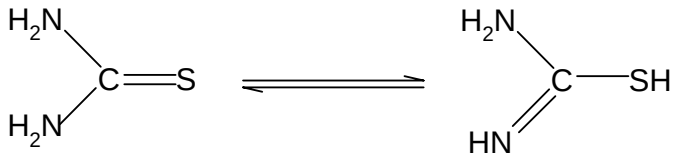
June 2, 1999

This report is an update of the Consensus Report published in 1988 (3).

Chemical and physical data

CAS No.:	62-56-6
Synonym:	thiocarbamide
Formula:	H_2N-CS-NH_2
Molecular weight:	76.12
Melting point:	176 – 178 °C
Boiling point:	disintegrates below boiling point
Density:	1.405 g/cm^3 (20 °C)
Solubility	
in water:	136 g/liter (20 °C)
in ethanol:	37 g/liter (20 °C)
Volatility:	does not evaporate from an aqueous solution (10)

Thiourea at room temperature is a white, crystalline powder. The compound occurs in two tautomeric forms, i.e. the molecule has three different reactive groups: amino, imino and sulfhydryl.



Occurrence and use

Thiourea is used as a catalyst in the production of fumaric acid, as an intermediate in the production of chemicals such as the dioxide of thiourea (formamidine sulfinic acid, FASA), thiouracil, thiobarbiturates and thiazol dyes, in metal refining, and as an antioxidant in photosensitive paper (diazo paper). Some metal polishes contain thiourea. Thiourea was once used medicinally as a treatment for hyperthyroidism (10).

Thiourea has been found to occur naturally in laburnum (14).

World production of thiourea in 1994 was about 10,000 tons, and Germany, Japan and China were the major producers (10).

Air monitoring during the years 1988-1991 at German workplaces where thiourea was used showed levels ranging from below the detection limit (not specified) to 0.32 mg/m^3, with an average of 0.085 mg/m^3 (10).

Uptake, distribution, excretion

Thiourea is taken up rapidly and completely from the digestive tracts of both animals and man (84). A peak concentration in blood was measured in human subjects 30 minutes after oral intake of 200 g thiourea, and at that point thiourea could also be found in urine.

Studies of skin uptake (rabbits) showed that 4% was absorbed if the thiourea (2.0 g/kg) was dissolved in water, whereas only 0.1% was absorbed if it was applied to the skin in solid form (10). Thiourea dissolved in acetone was applied to the lower arms of human subjects (4 µg/cm^2; total exposed surface was 13 cm^2) and they were instructed not to wash for 24 hours. Uptake was measured by determination of excretion in urine within the following 5 days: less than 1% was absorbed (20). In an in vivo study on hairless rats the flow of thiourea through the stratum corneum was determined to be 3.5 nmol/cm^2/hour (66). A Russian study with rats reports systemic effects on the thyroid after single exposures to 500 mg thiourea/kg body weight, applied to the back in the form of a 3% aqueous solution. The amount of uptake was not determined (45).

Thiourea is rapidly spread throughout the body. A whole-body autoradiography study with mice (^{14}C-thiourea, i.v.) showed that radioactivity began to accumulate in the thyroid after only 5 minutes, and remained higher in this tissue than in any other organ during the entire 4-day observation period. Elevated concentrations were also seen in the walls of the larger blood vessels, the adrenal cortex and mammary tissue, as well as liver, lungs and kidneys. Thiourea passes the placental barrier and also accumulates in the thyroid of the fetus (74). Red blood cells (25) and lungs (33, 34) contain proteins with high affinity for thiourea. Thiourea also seems to accumulate in melanomas in mice during the process of melanin synthesis (52). In an older study with rats, about 2% of an injected (i.p.) dose of ^{35}S-labeled thiourea was deposited in the thyroid mostly in the form of sulfate (56%) and bound to proteins (13%) (36).

In an experiment in which rats were given thiourea (100 mg/kg) by intraperitoneal injection, the half time in plasma was calculated to be 3.3 hours (25).

Thiourea is excreted primarily via the kidneys: when rats were given ^{35}S-labeled thiourea (1 mg, i.p.), 98% of the radioactivity was excreted in urine within 48 hours. Most of it was unchanged, but 6% was in the form of inorganic sulfate and another 6% in the form of ether sulfate (68). When rats were injected with ^{14}C-labeled thiourea (0.6 mg/kg, i.p.), 80 to 90% of the radioactivity was recovered in urine within 24 hours (35). When 12 healthy volunteers were given 500 mg thiourea intravenously, about a third of the dose was recovered in urine within 24 hours. Only a few percent were excreted during the following 24-hour period, and none thereafter (84).

Thiourea has been found in the urine of workers exposed to carbon disulfide (58).

Biotransformation

As mentioned previously, most thiourea is excreted unchanged, but some metabolic transformation may occur (see Figure 1). The microsomal flavin-containing mono-oxygenase (FMO) catalyzes an NADPH and O_2-dependent S-oxidation of thiourea to formamidine sulfinic acid (FASA) with the corresponding sulfenic acid as an intermediate product (60). This means that a more toxic product is formed (86, 88). Thiourea can also undergo autooxidiation to FASA (88). The oxidation of thiourea can also increase the formation of oxidized glutathione (GSSG) in the cells, since reduced glutathione (GSH) can reduce the sulfenic acid back to thiourea (46). Cyanamide and urea are other possible metabolites of thiourea (86). FMO occurs as multiple iso–enzymes in mammals. Five different isoenzymes of FMO have been identified in humans: FMO3 and FMO5 are expressed in the liver, and FMO1 in kidneys. It is not clear which isoenzymes oxidize thiourea. FMO1 from rats (37) and mice (38) has been shown in vitro to use thiourea as substrate. Inhibition experiments have shown that thiourea is probably the substrate for FMO3 from both mice and humans (18).

Thiourea functions as an antioxidant and has the ability to capture hydroxyl and superoxide radicals as well as hydrogen peroxide (42) and peroxynitrite (83). Experiments with yeast suggest that thiourea can also generate oxygen radicals (9).

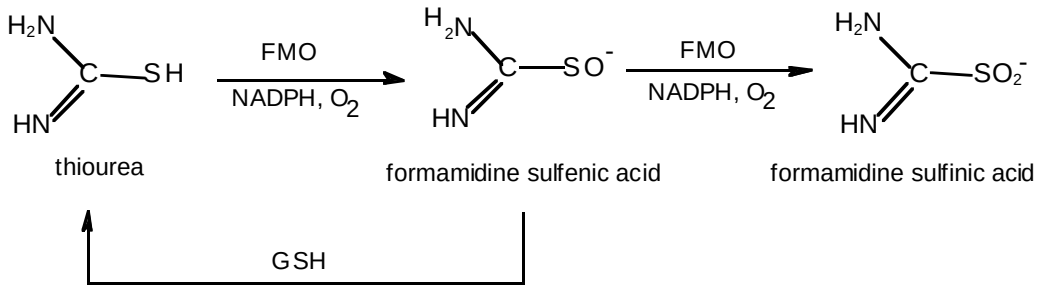


Figure 1. Metabolism of thiourea

Toxic effects

Human data

Of 525 patients given thiourea in initial doses of up to 2 – 3 g/day as a treatment for hyperthyroidism (antithyroid), 9% developed side effects in the form of fever, digestive upsets etc. (78). A maintenance dose in treatment of hyperthyroidism was 25 – 70 mg/day, whereas 10 – 15 mg/day had no effect (cited in Reference 15).

A pronounced drop in thrombocyte and granulocyte levels was seen in a woman who was treated with a total of 83 g thiourea over a period of 5 weeks. The toxic effect on bone marrow was reversible (54).

Indications of reduced thyroid function were observed in a Russian study of workers employed in thiourea manufacture. The study covered 45 exposed workers and 20 unexposed controls. Reported air concentrations of thiourea were in the range 0.6 to 12 mg/m^3. In the middle of the production hall the air concentration was 3.9 ± 1.0 mg/m^3, and concentrations around loading and cleaning were higher (9.0 ± 0.9 mg/m^3). The workers had been exposed for 9.5 ± 1.1 years, 73% had been exposed for at least 5 years, and 54.5% of them were over 40 years of age. The concentrations of thyroid hormones T_4 and T_3 were significantly lower in the exposed workers than in the controls (T_4: 78.0 ± 5.2 vs 109.4 ± 2.0 nmol /l, $p < 0.05$; T_3: 1.2 ± 0.1 vs 3.8 ± 0.1 nmol/l, $p < 0.001$). Thyroid hyperplasia was observed in 17 of the 45 exposed workers. Concentrations of T_4 and T_3 in this sub-group were 80.6 ± 1.8 and 0.9 ± 0.1 nmol/l respectively (76).

An elevated prevalence of hypothyroidism was observed, especially among men, in a study of workers in an English textile factory where thiourea (as well as resorcinol, which can also inhibit thyroid function) was used. The study was initiated because four cases of clinical hypothyroidism, three of which were men in their 40s, had been diagnosed among the factory workers within a period of 6 years. In a follow-up survey of 189 men and 48 women (44% of the employees: 115 production workers and 122 from offices, management and laboratories) 12 new cases of hypothyroidism of varying degrees of severity were discovered (classification according to Evered). Most of these cases were in the group considered to be unexposed. The air concentration of thiourea in the production hall was measured in the vicinity of the exhaust fans, and was reported to be about 5 μg/m^3, i.e. extremely low. The resorcinol concentration was also low: 20 μg/m^3. The authors suggest the possibility of exposure to chemicals outside the production facilities (63). The proportion of participants was low, the exposure situation was unclear, and no dose-response relationship was seen.

There are number of case reports of contact allergies from contact with photocopy paper containing thiourea (diazo paper), sometimes in combination with light (photoallergy) (16, 24, 41, 55). One case of contact and photoallergy after exposure to thiourea in silver polish has also been described (17). The number of reported cases of contact allergy to thiourea is low, considering how much of it is used (29, 39).

Exposure to thiourea in silver polish was recently suggested as a possible cause of a case of hepatitis (11).

Animal data

The LD_{50} for thiourea ranges from 125 to 10,000 mg/kg with oral exposure (mouse, rat, rabbit), and from 4 to 1340 mg/kg with intraperitoneal administration to various strains of rats (15). This indicates a wide variation in sensitivity between

different species and strains. The LC_{50} for rats (4 hours of inhalation) is above 170 mg/m^3 (10, 14).

Thiourea does not seem to be particularly irritating to eyes or skin (14).

Studies of lung damage have sometimes used chemicals including a thiourea group. Rats given 1.25 mg/kg thiourea by intraperitoneal injection (the LD_{50} for thiourea in this experiment was 3.55 mg/kg) developed pulmonary edema, probably because of effects on the endothelium that increased its permeability (26). Several studies of the mechanism behind this effect have shown that young rats are less sensitive, and that tolerance develops if the animals are first treated with a lower dose of thiourea (35). Toxicity to the lungs is correlated to the amount of thiourea that forms covalent bonds to proteins in the alveolar walls (35, 69) and to the histamine content of plasma (27, 28).

Repeated exposure to thiourea inhibits thyroid function in laboratory animals. Secondary effects are enlargement of the pituitary and atrophy of ovaries, uterus and prostate. Toxic effects on blood-forming organs have also been reported. Mice seem to be less sensitive than rats (15).

Young female rats (21 – 30 days old) were exposed for 10 days to various amounts of thiourea in drinking water (2 to 4 animals per dose). Inhibition of thyroid activity was registered in the form of induced compensatory thyroid hyperplasia. Animals exposed to 0.1% thiourea in drinking water (equivalent to 131 mg/kg body weight/day) developed greatly enlarged thyroids (hyperplasia). Exposure to 1% in drinking water (1.17 g/kg/day) had the same effect on the thyroid. A weak effect was observed at 21 mg/kg, and 12 mg/kg had no observed effect (5).

In a study with Sprague-Dawley rats, thiourea was given in drinking water (0.02 – 2.5 ppm) for 13 weeks. No clinical or histopathological effects were observed (reviewed in Reference 10).

Long-term exposure (3 to 63 weeks) of 11-month old female mice (0.25 – 0.375% thiourea in feed) caused changes in the adrenals, pituitary, ovaries, uterus and blood vessels, in addition to the effects on the thyroid (13).

Genotoxicity, mutagenicity

The results of a large number of experiments in which thiourea was tested for genotoxic/mutagenic activity have recently been summarized in a German report (4). The report concludes that thiourea is either not genotoxic or is only weakly so.

There are no data from bacterial test systems that clearly indicate genotoxicity (53, 85). Thiourea was not genotoxic in tests with Aspergillus nidulans (12). A weak mutagenic effect was observed in V79 cells (87) and in a host-mediated test system (72). Thiourea, without metabolic activation, was positive in a DNA repair test with E. coli (31) and in an in vitro micronucleus test (22). A cell transformation test with hamster embryo cells was considered negative, since a positive effect was seen only at the lowest dose (59). The results of various tests for DNA damage and repair in rat hepatocytes have been contradictory (1, 19, 47, 73, 87). Weak positive or

positive results (inter- and intrachromosomal recombinations), sometimes at toxic concentrations, have been observed in yeasts (23, 67). Formation of free radicals seems to be involved in the recombination effect in yeast (9). Some in vivo studies of genotoxicity with Drosophila have had positive results (8, 81) while others have been negative (6, 7, 64). Thiourea was not clastogenic in an in vivo micronucleus test with rats (study reviewed in Reference 4). Results of mouse lymphoma tests (in vivo) have been inconsistent (48, 51, 82).

It is not clear just how thiourea exercises its genotoxicity, but metabolism to a more reactive substance is probably necessary. Formamidine sulfinic acid, or FASA (see Biotransformation) is a conceivable candidate. FASA has been shown to be genotoxic in several in vitro tests (88). Chemical oxidation of ^{14}C-labeled thiourea with H_2O_2 in the presence of calf thymus DNA caused the formation of FASA, cyanamide and urea, as well as covalent bonding of the radioactivity to DNA (88).

Carcinogenicity

Thiourea has been tested for carcinogenic activity in several older studies. The results of these studies have been well summarized by the IARC (36) and the German working group for MAK values (15). The IARC has placed thiourea in Group 2B: "possibly carcinogenic to humans." The EU has made a similar assessment, and placed thiourea in Category 3 ("substances that may be carcinogenic to humans") in accordance with Directive 67/548/EEC. There are no reported studies in which thiourea was tested according to present praxis for cancer tests on animals (See Table 1).

Oral administration of thiourea to rats has caused tumors in the thyroid (61), liver (21), ear canal (Zymbal's gland), and eyelid (Meibom's gland) (65). These different tumor locations have usually been observed in different studies. In a 2-year study with rats in which thiourea was mixed in feed (80 ppm), no increase of tumor frequency was observed (62).

Thiourea's ability to cause tumors in the thyroid is attributed to hormonal disturbances. Rats are regarded as a sensitive species in this respect. Thiourea inhibits the enzyme thyroid peroxidase, causing a drop of thyroid hormones T_3 and T_4 in serum. This in turn stimulates the hypothalamus and pituitary to produce more thyroid-stimulating hormone (TSH). TSH stimulates thyroid growth, and chronically elevated levels of TSH in serum can result in thyroid hyperplasia, which may eventually develop into tumors (2, 30, 32).

A medium-term study, in which rats were exposed to 0.25% thiourea in drinking water (which would be about 200 mg/kg/day) for up to 26 weeks, showed that thiourea had a promotive effect on the formation of thyroid tumors in animals previously initiated with a nitrosamine. The hormonal changes were greater in the initiated animals than in those receiving thiourea alone (40). Elevated TSH levels seem to be most important during the earliest stages of tumor development (57, 71).

Table 1. Cancer studies with rats

Strain	Number	Exposure	Location	Response (animals with tumors)	Ref.
Local albino (R. norw.) males, females, Wistar males	3 x 10	0.25% in drinking water, 5 to 24 months. No controls	Thyroid	< 12 months: 0/5 > 12 months:22/25	61
Albino	8 x 18	up to 2 years: 0 0.01% in diet 0.025% 0.05 0.1 0.25 0.5 1%	Liver	Controls: 0/18 Surviving 2 years:14/29 (not dose-dependent) 3/5 4/8 2/8 5/8 ≤0.25%: all animals died within 17 months (1 liver tumor)	21, 36
Albino (Hebrew university) males	12 controls 16 (a) 19 (b)	(a) 4 ml 10% solution i.p. 3 times/week, 6 months; thereafter 0.2% in drinking water, up to 26 months (b) 0.2% in drinking water, up to 26 months	Nasal epithelium, eyelid (Meibom's gland), ear	Controls: 0/12 (a) 2-11 months: 0/4 died >12 months:10/12 (b) 18/19	65
Osborne-Mendel, males and females	4 x 30	24 months: 0 ppm 80 ppm in diet	Several different	Males:3 malignant/30 (1 lung, 2 subcutaneous) Females: 6 malignant (liver, lung, intestine, mammary, adrenal, lymph node metastasis) + 6 benign (mammary)/30 Males: 1 benign (testis + subcutaneous)/30 Females:1 malignant (lung + mammary) + 10 benign (9 mammary + 1 adrenal)/30	62

In a similar promotion study (initiation with a nitrosamine, followed by 0.2% thiourea in drinking water for 19 weeks) it was shown that simultaneous exposure to massive amounts of vitamin A enhanced the effect of the thiourea on the levels of T_3, T_4 and TSH in serum (49). A possible explanation for this may be induction of glucuronyltransferase, an enzyme that collaborates in the metabolism of thyroid hormone (32), which was observed in the livers of the animals treated with both

thiourea and vitamin A. Thiourea (0.2%) in drinking water, either with or without vitamin A, also increased the level of CYP2E1 in the liver (75).

Thiourea showed no initiating or promoting ability in a liver foci test with rats. Instead, treatment with 0.05 – 0.2% thiourea in drinking water for 51 to 70 days reduced both the number and the volume of foci (which lacked ATPase) in diethylnitrosamine-initiated male and female Sprague-Dawley rats (56). In another study with rats (F-344 males, initiation with another nitrosamine) thiourea (0.1% in drinking water for 19 weeks) increased the number of GSTP-positive foci. This treatment also resulted in nodules and neoplasias in thyroids. Thiourea and phenobarbital were observed to have a slight synergistic effect on liver foci (70).

Thyroid tumors have not been observed in mice after exposure to thiourea. In one study with mice in which thiourea was given orally (5 g/kg in diet), benign bone tumors were observed in the skull. According to the authors, the bone tumors were probably a side-effect of a direct toxic effect on the pituitary (50). In an older study with C3H mice it was found that high doses of thiourea inhibited the occurrence of spontaneous mammary tumors in surviving animals – probably because of thiourea's toxic effect on ovaries in this strain of mice and the consequent reduction in estrogen production (79).

No significant elevation in risk of bladder cancer was found in an epidemiological case-control study (relative risk 1.2; 95% confidence interval 0.4 – 3.3) in which exposure to thiourea was estimated from job or business sector (80).

Teratogenicity

High doses of thiourea given orally to pregnant rats (1 or 2 g/kg, day 12) and mice (1 g/kg, day 10) were fetotoxic, causing an increased number of resorbed embryos. No weight reductions and no deformities were observed in surviving fetuses (77).

When pregnant rats were exposed to thiourea (0.2% in drinking water) during the third or the second and third week of gestation, there were effects on the thyroids of the fetuses (day 20), the newborn pups and the mothers. The treatment had no effect on the body weights of the mothers. Exposure during the entire period of gestation was altogether too toxic for the fetuses. Increased thyroid weights and reduced iodine content were also observed in the young when the females were exposed to the same dose of thiourea during the nursing period. The effects on the thyroid were reversible (44). In another study by the same research group, female rats were given 0.2% thiourea in drinking water during the first 14 days of gestation. The exposure caused deformities in the bones and nervous systems of the young. General hemorrhaging was also observed (43).

Dose-effect/dose-response relationships

It was found in one study that occupational exposure to thiourea inhibited thyroid function, measured as lower levels of the thyroid hormones T_3 and T_4. Measured air concentrations in the factory were reported to be between 0.6 and 12 mg/m^3.

Thyroid hyperplasia was observed in 17 of 45 workers (76). If it is assumed that the workers weighed 70 kg and inhaled 1 m^3 per hour for 8 hours/day, and that uptake was complete, this air concentration is equivalent to a dose of 0.07 to 1.4 mg thiourea/kg body weight/day. Data from therapeutic use of thiourea indicate clearly that a daily oral dose of 10 to 15 mg/day (about 0.1 - 0.2 mg/kg/day) has no effect on the thyroid, and that doses of 25 to 70 mg/day (about 0.4 - 1.0 mg/kg/day) do have an effect (15).

The Dutch expert group has reported a NOEL of 4 mg/kg (80 ppm in diet, Osborne-Mendel rats) for the tumorigenic effect on rats with oral exposure (14, 62). An LD_{50} of about the same size, 3.55 mg/kg, has been reported for Sprague-Dawley rats after intraperitoneal injection of thiourea (26).

For rats, The NOEL for thiourea in drinking water (13-week study) was determined to be > 2.5 ppm (10), equivalent to a dose of about > 0.25 mg/kg/day.

Conclusions

The critical effect of thiourea is inhibition of thyroid function, and this has been reported with occupational exposure. Thiourea causes tumors in experimental animals. In addition to thyroid tumors, which are probably caused by hormonal disturbances to which the test animals are particularly sensitive, tumors in other locations – notably liver, ear canal and eyelid – have been observed. Documentation of this carcinogenic activity is weak, however. The results of tests for genotoxicity are inconsistent. Skin contact with thiourea can cause contact allergy and photoallergy. Thiourea can cross the placental barrier.

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Summary

Montelius J (ed). Scientific Basis for Swedish Occupational Standards. XX. Arbete och Hälsa 1999:26, pp 1-116.

Critical evaluation of those scientific data which are relevant as a background for discussion of Swedish occupational exposure limits. This volume consists of the consensus reports given by the Criteria Group at the Swedish National Institute for Working Life between July, 1998 and June, 1999.

Key Words: Calcium oxide, Calcium hydroxide, Cyanamide, Cyclohexanone, Dimethyl adipate, Dimethyl glutarate, Dimethyl succinate, Ethylene glycol monomethyl ether, Ethylene glycol monomethyl ether acetate, Glutaraldehyde, Lactate esters, Methyl tertiary-butyl ether, Occupational Exposure Limit (OEL), Pentafluoroethane, Phosphorus trichloride, Phosphorus pentachloride, Phosphoryl chloride, Scientific Basis, Thiourea, Trifluoroethane.

Sammanfattning

Montelius J (ed). Vetenskapligt underlag för hygieniska gränsvärden. XX. Arbete och Hälsa 1999:26, s 1-116.

Sammanställningar baserade på kritisk genomgång och värdering av de vetenskapliga fakta, vilka är relevanta som underlag för fastställande av hygieniskt gränsvärde. Volymen omfattar de underlag som avgivits från Kriteriegruppen för hygieniska gränsvärden under perioden juli 1998 - juni 1999.

Nyckelord: Cyanamid, Cyklohexanon, Dimetyladipat, Dimetylglutarat, Dimetylsuccinat, Etylenglykolmetyleter, Etylenglykolmetyleteracetat, Fosfortriklorider, Fosforpentaklorid, Fosforylklorid, Glutaraldehyd, Hygieniskt gränsvärde, Kalciumoxid, Kalciumhydroxid, Laktatestrar, Metyl-tert-butyleter, Pentafluoretan, Tiourinämne, Trifluoretan, Vetenskapligt underlag.

En svensk version av dessa vetenskapliga underlag finns publicerad i Arbete och Hälsa 1999:25.

APPENDIX

Consensus reports in this and previous volumes

Substance	Consensus date	Volume in Arbete och Hälsa	(No.)
Acetaldehyde	February 17, 1987	1987:39	(VIII)
Acetamide	December 11, 1991	1992:47	(XIII)
Acetic acid	June15, 1988	1988:32	(IX)
Acetone	October 20, 1987	1988:32	(IX)
Acetonitrile	September 12, 1989	1991:8	(XI)
Acrylamide	April 17, 1991	1992:6	(XII)
Acrylates	December 9, 1984	1985:32	(VI)
Acrylonitrile	April 28, 1987	1987:39	(VIII)
Aliphatic amines	August 25, 1982	1983:36	(IV)
Aliphatic hydrocarbons, C_{10}-C_{15}	June 1, 1983	1983:36	(IV)
Aliphatic monoketons	September 5, 1990	1992:6	(XII)
Allyl alcohol	September 9, 1986	1987:39	(VIII)
Allylamine	August 25, 1982	1983:36	(IV)
Allyl chloride	June 6, 1989	1989:32	(X)
Aluminum	April 21, 1982	1982:24	(III)
revised	September 14, 1994	1995:19	(XVI)
p-Aminoazobenzene	February 29, 1980	1981:21	(I)
Ammonia	April 28, 1987	1987:39	(VIII)
Amylacetate	March 23, 1983	1983:36	(IV)
Aniline	October 26, 1988	1989:32	(X)
Anthraquinone	November 26,1987	1988:32	(IX)
Arsenic, inorganic	December 9, 1980	1982:9	(II)
revised	February 15, 1984	1984:44	(V)
Arsine	October 20, 1987	1988:32	(IX)
Asbestos	October 21, 1981	1982:24	(III)
Barium	June 16, 1987	1987:39	(VIII)
revised	January 26, 1994	1994:30	(XV)
Benzene	March 4, 1981	1982:9	(II)
revised	February 24, 1988	1988:32	(IX)
Benzoyl peroxide	February 13, 1985	1985:32	(VI)
Beryllium	April 25, 1984	1984:44	(V)
Borax	October 6, 1982	1983:36	(IV)
Boric acid	October 6, 1982	1983:36	(IV)
Boron Nitride	January 27 1993	1993:37	(XIV)
Butadiene	October 23, 1985	1986:35	(VII)
1-Butanol	June 17, 1981	1982:24	(III)
Butanols	June 6, 1984	1984:44	(V)
Butyl acetate	June 6, 1984	1984:44	(V)
Butyl acetates	February 11, 1998	1998:25	(XIX)
Butylamine	August 25, 1982	1983:36	(IV)
Butyl glycol	October 6, 1982	1983:36	(IV)
Cadmium	January 18, 1980	1981:21	(I)
revised	February 15, 1984	1984:44	(V)
revised	May 13, 1992	1992:47	(XIII)
Calcium hydroxide	February 24, 1999	1999:26	(XX)
Calcium nitride	January 27, 1993	1993:37	(XIV)
Calcium oxide	February 24, 1999	1999:26	(XX)

Caprolactam	October 31, 1989	1991:8	(XI)
Carbon monoxide	December 9, 1981	1982:24	(III)
Cathecol	September 4, 1991	1992:47	(XIII)
Chlorine	December 9, 1980	1982:9	(II)
Chlorine dioxide	December 9, 1980	1982:9	(II)
o-Chlorobenzylidene malononitrile	June 1, 1994	1994:30	(XV)
Chlorocresol	December 12, 1990	1992:6	(XII)
Chlorodifluoromethane	June 2, 1982	1982: 24	(III)
Chlorophenols	September 4, 1985	1986:35	(VII)
Chloroprene	April 16, 1986	1986:35	(VII)
Chromium	December 14, 1979	1981:21	(I)
revised	May 26, 1993	1993:37	(XIV)
Coal dust	September 9, 1986	1987:39	(VIII)
Cobalt	October 27, 1982	1983:36	(IV)
Copper	October 21, 1981	1982:24	(III)
Cotton dust	February14, 1986	1986:35	(VII)
Creosote	October 26, 1988	1989:32	(X)
Cresols	February 11, 1998	1998:25	(XIX)
Cumene	June 2, 1982	1982:24	(III)
Cyanamid	September 30, 1998	1999:26	(XX)
Cyanoacrylates	March 5, 1997	1997:25	(XVIII)
Cycloalkanes, C_5-C_{15}	April 25, 1984	1984:44	(V)
Cyclohexanone	March 10, 1982	1982:24	(III)
revised	February 24 1999	1999:26	(XX)
Cyclohexanone peroxide	February 13, 1985	1985:32	(VI)
Cyclohexylamine	February 7, 1990	1991:8	(XI)
Desflurane	May 27, 1998	1998:25	(XIX)
Diacetone alcohol	December 14, 1988	1989:32	(X)
Dichlorobenzenes	February 11, 1998	1998:25	(XIX)
1,2-Dibromo-3-chloropropane	May 30, 1979	1981:21	(I)
Dichlorodifluoromethane	June 2, 1982	1982:24	(III)
1,2-Dichloroethane	February 29, 1980	1981:21	(I)
Dichloromethane	February 29, 1980	1981:21	(I)
Dicumyl peroxide	February 13, 1985	1985:32	(VI)
Dicyclopentadiene	March 23, 1994	1994:30	(XV)
Diethanolamine	September 4, 1991	1992:47	(XIII)
Diethylamine	August 25, 1982	1983:36	(IV)
2-Diethylaminoethanol	January 25, 1995	1995:19	(XVI)
Diethylene glycol	September 16, 1992	1993:37	(XIV)
Diethyleneglycol ethylether + acetate	December 11, 1996	1997:25	(XVIII)
Diethyleneglycol methylether + acetate	March 13, 1996	1996:25	(XVII)
Diethyleneglycol monobutylether	January 25, 1995	1995:19	(XVI)
Diethylenetriamine	August 25, 1982	1983:36	(IV)
revised	January 25, 1995	1995:19	(XVI)
Diisocyanates	April 8, 1981	1982:9	(II)
revised	April 27, 1988	1988:32	(IX)
Diisopropylamine	February 7, 1990	1991:8	(XI)
N,N-Dimethylacetamide	March 23, 1994	1994:30	(XV)
Dimethyl adipate	December 9, 1998	1999:26	(XX)
Dimethylamine	December 10, 1997	1998:25	(XIX)
N,N-Dimethylaniline	December 12, 1989	1991:8	(XI)
Dimethyldisulfide	September 9, 1986	1987:39	(VIII)
Dimethylether	September 14, 1994	1995:19	(XVI)
Dimethylethylamine	June 12, 1991	1992:6	(XII)
Dimethylformamide	March 23, 1983	1983:36	(IV)
Dimethyl glutarate	December 9, 1998	1999:26	(XX)
Dimethylhydrazine	January 27, 1993	1993:37	(XIV)

Dimethyl succinate	December 9, 1998	1999:26	(XX)
Dimethylsulfide	September 9, 1986	1987:39	(VIII)
Dimethylsulfoxide, DMSO	December 11, 1991	1992:47	(XIII)
Dioxane	August 25, 1982	1983:36	(IV)
revised	March 4, 1992	1992:47	(XIII)
Diphenylamine	January 25, 1995	1995:19	(XVI)
4,4'-Diphenylmethanediisocyanate	April 8, 1981	1982:9	(II)
Dipropylene glycol	May 26, 1993	1993:37	(XIV)
Dipropyleneglycol monomethylether	December 12, 1990	1992:6	(XII)
Disulfiram	October 31, 1989	1991:8	(XI)
Enzymes, industrial	June 5, 1996	1996:25	(XVII)
Ethanol	May 30, 1990	1991:8	(XI)
Ethanolamine	September 4, 1991	1992:47	(XIII)
Ethylacetate	March 28, 1990	1991:8	(XI)
Ethylamine	August 25, 1982	1983:36	(IV)
Ethylamylketone	September 5, 1990	1992:6	(XII)
Ethylbenzene	December 16, 1986	1987:39	(VIII)
Ethylchloride	December 11, 1991	1992:47	(XIII)
Ethylene	December 11, 1996	1997:25	(XVIII)
Ethylene chloride	February 29, 1980	1981:21	(I)
Ethylene diamine	August 25, 1982	1983:36	(IV)
Ethylene glycol	October 21, 1981	1982:24	(III)
Ethylene glycol methylether + acetate	June 2, 1999	1999:26	(XX)
Ethyleneglycol monoisopropylether	November 16, 1994	1995:19	(XVI)
Ethyleneglycol monopropylether + acetate	September 15, 1993	1994:30	(XV)
Ethylene oxide	December 9, 1981	1982:24	(III)
Ethylether	January 27, 1993	1993:37	(XIV)
Ethylglycol	October 6, 1982	1983:36	(IV)
Ferbam	September 12, 1989	1991:8	(XI)
Ferric dimethyldithiocarbamate	September 12, 1989	1991:8	(XI)
Flour dust	December 10, 1997	1998:25	(XIX)
Formaldehyde	June 30, 1979	1981:21	(I)
revised	August 25, 1982	1983:36	(IV)
Formamide	December 12, 1989	1991:8	(XI)
Formic acid	June 15, 1988	1988:32	(IX)
Furfural	April 25, 1984	1984:44	(V)
Furfuryl alcohol	February 13, 1985	1985:32	(VI)
Gallium + Gallium compounds	January 25, 1995	1995:19	(XVI)
Glutaraldehyde	September 30 1998	1999:26	(XX)
Glycol ethers	October 6, 1982	1983:36	(IV)
Glyoxal	September 13, 1996	1996:25	(XVII)
Grain dust	December 14, 1988	1989:32	(X)
Graphite	December 10, 1997	1998:25	(XIX)
Halothane	April 25, 1985	1985:32	(VI)
2-Heptanone	September 5, 1990	1992:6	(XII)
3-Heptanone	September 5, 1990	1992:6	(XII)
Hexachloroethane	September 15, 1993	1994:30	(XV)
Hexamethylenediisocyanate	April 8, 1981	1982:9	(II)
Hexamethylenetetramine	August 25, 1982	1983:36	(IV)
n-Hexane	January 27, 1982	1982:24	(III)
2-Hexanone	September 5, 1990	1992:6	(XII)
Hexyleneglycol	November 17, 1993	1994:30	(XV)
Hydrazine	May 13, 1992	1992:47	(XIII)
Hydrogen bromide	February 11, 1998	1998:25	(XIX)

Hydrogen fluoride	April 25,	1984	1984:44	(V)
Hydrogen peroxide	April 4,	1989	1989:32	(X)
Hydrogen sulfide	May 4,	1983	1983:36	(IV)
Hydroquinone	October 21,	1989	1991:8	(XI)
Indium	March 23,	1994	1994:30	(XV)
Industrial enzymes	June 5,	1996	1996:25	(XVII)
Isophorone	February 20,	1991	1992:6	(XII)
Isopropanol	December 9,	1981	1982:24	(III)
Isopropylamine	February 7,	1990	1991:8	(XI)
Isopropylbenzene	June 2,	1982	1982:24	(III)
Lactates	March 29,	1995	1995:19	(XVI)
Lactate esters	June 2,	1999	1999:26	(XX)
Lead, inorganic	February 29,	1980	1981:21	(I)
revised	September 5,	1990	1992:6	(XII)
Lithium boron nitride	January 27,	1993	1993:37	(XIV)
Lithium nitride	January 27,	1993	1993:37	(XIV)
Maleic anhydride	September 12,	1989	1991:8	(XI)
Manganese	February 15,	1983	1983:36	(IV)
revised	April 17,	1991	1992:6	(XII)
revised	June 4,	1997	1997:25	(XVIII)
Man made mineral fibers	March 4,	1981	1982:9	(II)
revised	December 1,	1987	1988:32	(IX)
Mercury, inorganic	April 25,	1984	1984:44	(V)
Mesityl oxide	May 4,	1983	1983:36	(IV)
Metal stearates, some	September 15,	1993	1994:30	(XV)
Methacrylates	September 12,	1984	1985:32	(VI)
Methanol	April 25,	1985	1985:32	(VI)
Methyl acetate	March 28	1990	1991:8	(XI)
Methylamine	August 25,	1982	1983:36	(IV)
Methylamyl alcohol	March 17,	1993	1993:37	(XIV)
Methyl bromide	April 27,	1988	1988:32	(IX)
Methyl chloride	March 4,	1992	1992:47	(XIII)
Methyl chloroform	March 4,	1981	1982:9	(II)
Methylene chloride	February 29,	1980	1981:21	(I)
4,4'-Methylene dianiline	June 16,	1987	1987:39	(VIII)
Methyl ethyl ketone	February 13,	1985	1985:32	(VI)
Methyl ethyl ketone peroxide	February 13,	1985	1985:32	(VI)
Methyl formare	December 12,	1989	1991:8	(XI)
Methyl glycol	October 6,	1982	1983:36	(IV)
Methyl iodide	June 30,	1979	1981:21	(I)
Methylisoamylamine	September 5,	1990	1992:6	(XII)
Methyl mercaptane	September 9,	1986	1987:39	(VIII)
Methyl methacrylate	March 17,	1993	1993:37	(XIV)
Methyl pyrrolidone	June 16,	1987	1987:39	(VIII)
Methyl-t-butyl ether	November 26,	1987	1988:32	(IX)
revised	September 30,	1998	1999:26	(XX)
Mixed solvents, neurotoxicity	April 25,	1985	1985:32	(VI)
Molybdenum	October 27,	1982	1983:36	(IV)
Monochloroacetic acid	February 20,	1991	1992:6	(XII)
Monochlorobenzene	September 16,	1993	1993:37	(XIV)
Monomethylhydrazine	March 4,	1992	1992:47	(XIII)
Mononitrotoluene	February 20,	1991	1992:6	(XII)
Monoterpenes	February 17,	1987	1987:39	(VIII)
Morpholine	December 8,	1982	1983:36	(IV)
revised	June 5,	1996	1996:25	(XVII)

Naphthalene	May 27, 1998	1998:25	(XIX)
Natural crystallinic fibers (except asbestos)	June 12, 1991	1992:6	(XII)
Nickel	April 21, 1982	1982:24	(III)
Nitroethane	April 4, 1989	1989:32	(X)
Nitrogen oxides	December 11, 1985	1986:35	(VII)
Nitroglycerin	February 13, 1985	1985:32	(VI)
Nitroglycol	February 13, 1985	1985:32	(VI)
Nitromethane	January 6, 1989	1989:32	(X)
Nitropropane	October 28, 1986	1987:39	(VIII)
2-Nitropropane	March 29, 1995	1995:19	(XVI)
Nitroso compounds	December 12, 1990	1992:6	(XII)
Nitrosomorpholine	December 8, 1982	1983:36	(IV)
Nitrotoluene	February 20, 1991	1992:6	(XII)
Nitrous oxide	December 9, 1981	1982:24	(III)
Oil mist	April 8, 1981	1982:9	(II)
Organic acid anhydrides, some	September 12, 1989	1991:8	(XI)
Oxalic acid	February 24, 1988	1988:32	(IX)
Ozone	April 28, 1987	1987:39	(VIII)
Paper dust	February 7, 1990	1991:8	(XI)
Pentaerythritol	November 16, 1994	1995:19	(XVI)
1,1,1,2,2-Pentafluoroethane	February 24, 1999	1999:26	(XX)
Peroxides, organic	February 13, 1985	1985:32	(VI)
Phenol	February 13, 1985	1985:32	(VI)
Phosphorous chlorides	September 30, 1998	1999:26	(XX)
Phosphorous oxides	February 11, 1998	1998:25	(XIX)
Phthalates	December 8, 1982	1983:36	(IV)
Phthalic anhydride	September 12, 1989	1991:8	(XI)
Piperazine	September 12, 1984	1985:32	(VI)
Plastic dusts	December 16, 1986	1987:39	(VIII)
Platinum	June 4, 1997	1997:25	(XVIII)
Polyaromatic hydrocarbons	February 15, 1984	1984:44	(V)
Polyisocyanates	April 27, 1988	1988:32	(IX)
Potassium aluminium fluoride	June 4, 1997	1997:25	(XVIII)
2-Propanol	December 9, 1981	1982:24	(III)
Propene	September 13, 1996	1996:25	(XVII)
Propionic acid	November 26, 1987	1988:32	(IX)
Propylacetate	September 14, 1994	1995:19	(XVI)
Propylene glycol	June 6, 1984	1984:44	(V)
Propylene glycol-1,2-dinitrate	May 4, 1983	1983:36	(IV)
Propylene glycol monomethylether	October 28, 1986	1987:39	(VIII)
Propylene oxide	June 11, 1986	1986:35	(VII)
Pyridine	May 13, 1992	1992:47	(XIII)
Quartz	March 13, 1996	1996:25	(XVII)
Resorcinol	September 4, 1991	1992:47	(XIII)
Selenium	December 11, 1985	1986:35	(VII)
revised	February 22, 1993	1993:37	(XIV)
Sevoflurane	May 27, 1998	1998:25	(XIX)
Silica	March 13, 1996	1996:25	(XVII)
Silver	October 28, 1986	1987:39	(VIII)
Stearates, metallic, some	September 15, 1993	1994:30	(XV)
Stearates, non-metallic, some	November 17, 1993	1994:30	(XV)
Strontium	January 26, 1994	1994:30	(XV)

Styrene	February 29,	1980	1981:21	(I)
revised	October 31,	1989	1991:8	(XI)
Sulfur dioxide	April 25,	1985	1985:32	(VI)
Sulfur fluorides	March 28,	1990	1991:8	(XI)
Synthetic inorganic fibers	March 4,	1981	1982:9	(II)
revised	December 1,	1987	1988:32	(IX)
Synthetic organic and inorganic fibers	May 30,	1990	1991:8	(XI)
Talc dust	June 12,	1991	1992:6	(XII)
Terpenes, mono-	February 17,	1987	1987:39	(VIII)
Tetrabromoethane	May 30,	1990	1991:8	(XI)
Tetrachloroethane	June 4,	1997	1997:25	(XVIII)
Tetrachloroethylene	February 29,	1980	1981:21	(I)
1,1,1,2-Tetrafluoroethane	March 29,	1995	1995:19	(XVI)
Tetrahydrofuran	October 31,	1989	1991:8	(XI)
Tetranitromethane	April 4,	1989	!989:32	(X)
Thioglycolic acid	June 1,	1994	1994:30	(XV)
Thiourea	December 1,	1987	1988:32	(IX)
revised	June 2,	1999	1999:26	(XX)
Thiram	October 31,	1989	1991:8	(XI)
Thiurams, some	October 31,	1989	1991:8	(XI)
Titanium dioxide	February 21,	1989	1989:32	(X)
Toluene	February 29,	1980	1981:21	(I)
Toluene-2,4-diisocyanate	April 8,	1981	1982:9	(II)
Toluene-2,6-diisocyanate	April 8,	1981	1982:9	(II)
1,1,1-Trifluoroethane	February 24,	1999	1999:26	(XX)
Trichlorobenzene	September 16,	1993	1993:37	(XIV)
1,1,1-Trichloroethane	March 4,	1981	1982:9	(II)
Trichloroethylene	December 14,	1979	1981:21	(I)
Trichlorofluoromethane	June 2,	1982	1982:24	(III)
1,1,2-Trichloro-1,2,2-trifluoroethane	June 2,	1982	1982:24	(III)
Triethanolamine	August 25,	1982	1983:36	(IV)
Triethylamine	December 5,	1984	1985:32	(VI)
Trimellitic anhydride	September 12,	1989	1991:8	(XI)
Trimethylolpropane	November 16,	1994	1995:19	(XVI)
Trinitrotoluene	April 17,	1991	1992:6	(XII)
Vanadium	March 15,	1983	1983:36	(IV)
Vinyl acetate	June 6,	1989	1989:32	(X)
Vinyl toluene	December 12,	1990	1992:6	(XII)
White spirit	December 16,	1986	1987:39	(VIII)
Wood dust	June 17,	1981	1982:9	(II)
Xylene	February 29,	1980	1981:21	(I)
Zinc	April 21,	1982	1982:24	(III)
Zinc dimethyl dithiocarbamate	September 12,	1989	1991:8	(XI)
Ziram	September 12,	1989	1991:8	(XI)

Sent for publication December 1999