

CLH report

Proposal for Harmonised Classification and Labelling

**Based on Regulation (EC) No 1272/2008 (CLP Regulation),
Annex VI, Part 2**

**Substance Name: 2,3-epoxypropyl methacrylate (Glycidyl
methacrylate, GMA)**

EC Number: 203-441-9

CAS Number: 106-91-2

Index Number: 607-123-00-4

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Part A.

1 PROPOSAL FOR HARMONISED CLASSIFICATION AND LABELLING

1.1 Substance

Table 1: Substance identity

Substance name:	<i>2,3-epoxypropyl methacrylate; glycidyl methacrylate</i>
EC number:	<i>203-441-9</i>
CAS number:	<i>106-91-2</i>
Annex VI Index number:	<i>607-123-00-4</i>
Degree of purity:	<i>>= 99.5 — <= 100.0 % (w/w)</i>
Impurities and additives:	<i>None relevant for classification</i>

1.2 Harmonised classification and labelling proposal

Table 2: The current Annex VI entry and the proposed harmonised classification

	CLP Regulation	Directive 67/548/EEC (Dangerous Substances Directive; DSD)
Current entry in Annex VI, CLP Regulation	Acute Tox. 4* H302 Acute Tox. 4* H312 Acute Tox. 4* H332 Eye Irrit. 2 H319 Skin Irrit. 2 H315 Skin Sens. 1 H317 Note D applies (Annex VI, Part 1, section 1.1.3.1) The * indicates a minimum classification (Annex VI, Part 1, section 1.2.1)	Xn; R20/21/22 Xi; R36/38 R43 Note D applies (Annex VI, Part 1, section 1.1.3.1) Specific concentration limits: Xi; R36/38: C ≥ 10 %
Current proposal for consideration by RAC	Acute Tox. 4 H302 Acute Tox. 3 H311 No classification for Acute Tox inhalation Eye Dam. 1 H318 Skin Corr. 1C H314 Skin Sens. 1 H317 Repr. 1B H360F Muta. 2 H341	Not relevant seen the repealing of this Directive

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

	<p>Carc. 1B H350 STOT SE 1 H370 (affected organs: respiratory tract; route of exposure: inhalation)</p>	
<p>Resulting harmonised classification (future entry in Annex VI, CLP Regulation)</p>	<p>Acute Tox. 4 H302 Acute Tox. 3 H311 Eye Dam. 1 H318 Skin Corr. 1C H314 Skin Sens. 1 H317 Repr. 1B H360F Muta. 2 H341 Carc. 1B H350 STOT SE 1 H370 (affected organs: respiratory tract; route of exposure: inhalation)</p> <p>Note D</p>	

1.3 Proposed harmonised classification and labelling based on CLP Regulation

Table 3a: Classification and labelling according to CLP / GHS for physicochemical properties

CLP Annex I reference and Hazard class	Proposed classification	Proposed SCLs and/or M-factors	Current classification	Reason for no classification
2.1.Explosives:			None	Not assessed
2.2 Flammable gases:			None	Not assessed
2.3 Flammable aerosols:			None	Not assessed
2.4 Oxidising gases:			None	Not assessed
2.5 Gases under pressure:			None	Not assessed
2.6 Flammable liquids:			None	Not assessed
2.7 Flammable solids:			None	Not assessed
2.8 Self-reactive substances and mixtures:			None	Not assessed
2.9 Pyrophoric liquids:			None	Not assessed
2.10 Pyrophoric solids:			None	Not assessed
2.11 Self-heating substances and mixtures:			None	Not assessed
2.12 Substances and mixtures which in contact with water emit flammable gases:			None	Not assessed
2.13 Oxidising liquids:			None	Not assessed

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2.14 Oxidising solids:			None	Not assessed
2.15 Organic peroxides:			None	Not assessed
2.16 Corrosive to metals:			None	Not assessed

Table 3b: Classification and labelling according to CLP / GHS for health hazards

CLP Annex I reference and Hazard class	Proposed classification	Proposed SCLs and/or M-factors	Current classification	Reason for no classification
3.1 Acute toxicity - oral:	Acute Tox. 4 (H302)	.	Acute Tox. 4* (H302)	
3.1 Acute toxicity - dermal:	Acute Tox. 3 (H311)		Acute Tox. 4* (H312)	
3.1 Acute toxicity - inhalation:			Acute Tox. 4* (H332)	conclusive but not sufficient for classification
3.2 Skin corrosion / irritation:	Skin Corr. 1C (H314)		Skin Irrit. 2 (H315)	
3.3 Serious eye damage / eye irritation:	Eye Dam. 1 (H318)		Eye Irrit. 2 (H319)	
3.4 Respiration sensitisation:			None	No data
3.4 Skin sensitisation:	Skin Sens. 1 (H317)	.	Skin Sens. 1 (H317)	
3.5 Germ cell mutagenicity:	Muta. 2 (H341)		None	
3.6 Carcinogenicity:	Carc. 1B (H350)		None	
3.7 Reproductive Toxicity:	Repr. 1B (H360F)		None	
3.7 Reproductive Toxicity: Effects on or	None		None	data lacking

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via lactation:				
3.8 Specific target organ toxicity - single:	STOT SE 1 (H370) Affected organs: respiratory tract Route of exposure: Inhalation		None	
3.9 Specific target organ toxicity - repeated:	None		None	conclusive but not sufficient for classification
3.10 Aspiration hazard:	None		None	conclusive but not sufficient for classification

The * indicates a minimum classification (Annex VI, Part 1, section 1.2.1)

Table 3c: Classification and labelling according to CLP / GHS for environmental hazards

CLP Annex I reference and Hazard class	Proposed classification	Proposed SCLs and/or M-factors	Current classification	Reason for no classification
4.1 Hazards to the aquatic environment (acute/short-term):			None	Not assessed
4.1 Hazards to the aquatic environment (long-term):			None	Not assessed
5.1 Hazardous to the ozone layer:			None	Not assessed

Labelling

Signal word: Danger

Hazard pictograms:



GHS05: corrosion



GHS06: skull and crossbones



GHS08: health hazard

Hazard statements:

- H302: Harmful if swallowed.
- H311: Toxic in contact with skin.
- H314: Causes severe skin burns and eye damage.
- H317: May cause an allergic skin reaction.
- H341: Suspected of causing genetic defects
- H350: May cause cancer
- H360F: May damage fertility
- H370: Causes damage to the respiratory tract by inhalation.

Precautionary statements:

(as applied by the registrants but not relevant for harmonisation)

- P201: Obtain special instructions before use.
- P202: Do not handle until all safety precautions have been read and understood.
- P260: Do not breathe dust/fume/gas/mist/vapours/spray.
- P264: Wash hands thoroughly after handling.
- P270: Do not eat, drink or smoke when using this product.
- P272: Contaminated work clothing should not be allowed out of the workplace.
- P363: Wash contaminated clothing before reuse.
- P280: Wear protective gloves/protective clothing/eye protection/face protection.
- P305+P351+P338: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
- P304+P340: IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing.
- P301+P312: IF SWALLOWED: Call a POISON CENTER or doctor/physician if you feel unwell.
- P301+P330+P331: IF SWALLOWED: rinse mouth. Do NOT induce vomiting.
- P303+P361+P353: IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower.
- P310: Immediately call a POISON CENTER or doctor/physician.
- P333+P313: If skin irritation or rash occurs: Get medical advice/attention.
- P321: Specific treatment (see on this label).
- P322: Specific measures (see on this label).
- P307+P311: IF exposed: Call a POISON CENTER or doctor/physician.
- P308+P313: IF exposed or concerned: Get medical advice/attention.
- P405: Store locked up.
- P501: Dispose of contents/container in accordance with local and national regulations

Proposed notes assigned to an entry: Note D

BACKGROUND TO THE CLH PROPOSAL

1.4 History of the previous classification and labelling

Much history is not available to the current submitters, but the substance has had the same harmonised classification for many years, initially since 1991 under the DSD Directive, now under the CLP Regulation, based on information such as for instance summarised in the OECD SIDS Initial Assessment Report of 1999.

However since then new studies have become available which required a revision within the framework of the substance evaluation as performed by its SIEF. This revised classification is now part of the most recent update (Quarter2 2013) of the REACH Registration dossier and consequently also the harmonised classification should be updated.

This CLH proposal was requested by the Lead Registrant for the substance (Stadex Nederland BV) who acts on behalf of the current 11 members of the SIEF. The draft of the proposed revision was performed by a sub-set of the SIEF and consisted of representatives of DOW, DuPont, Mitsubishi Rayon and Stadex. The proposed changes of the classification are agreed by the dossier submitter (The Netherlands). The proposal is submitted by the Netherlands as it contains changes of the harmonised classification (article 37.6).

1.5 Short summary of the scientific justification for the CLH proposal

Toxicokinetics

Results indicate that glycidyl methacrylate (GMA) is rapidly metabolized *in vivo* in rabbits as measured by disappearance of parent material (95% of intravenous administered dose of 200 mg/kg eliminated within 10 minutes). Maximum blood levels of GMA were increased by 10-fold in rabbits co-administered a carboxylesterase inhibitor indicating that GMA was likely metabolized by carboxylesterase to glycidol and methacrylic acid. Domoradzki (2004) observed similar findings in rat, rabbit and human liver homogenate, and rat and rabbit respiratory tract epithelial cell fractions *in vitro*. Glycidol was identified as the metabolite of GMA in this study.

Metabolism of GMA to glycidol has ramifications for hazard identification. Glycidol has a harmonised classification according to CLP (entry 603-063-00-8) as carcinogenic (category 1B), germ cell mutagenic (category 2) and toxic to reproduction (category 1B).

Acute toxicity

- Acute toxicity oral Category 4 (H302) (the LD50 values of all available studies are between 300 and 2000 mg/kg bw)
- Acute toxicity dermal Category 3 (H311) (the actual LD50 value of 480 mg/kg bw lies between 200 and 1000 mg/kg bw)
- No classification for Acute toxicity inhalation is required since the LC0 in rats exposed for 4 hours lies at 2394 mg/m³, the highest practically attainable vapour concentration and highest dose tested.

STOT SE

The findings in the acute toxicity inhalation studies do warrant a classification for Specific Target

Organ Toxicity, Single Exposure Category 1 (damage to the respiratory tract after inhalation) (H370). Local respiratory tract tissue injury is also expected given the corrosiveness seen on skin and eyes, while with repeated inhalation exposure degenerative effects occur at the portal of entry in both rats and rabbits at similar concentrations (around 10 to 60 mg/m³). These effects seem concentration related and are best addressed via a STOT SE Category 1.

Skin corrosion

In a DOT standard test (equivalent to OECD Test Guideline 404, key study), moderate necrosis occurred by 4 hours exposure in 2 out of 6 animals but not 1-hour exposure (only very slight or superficial).

GMA was corrosive to the skin of rabbits. The 0.1 ml applied area showed red, swelled and blistered after one or two days, subdermal bleeding and ulcers after three days, and hard, thicker, cracked, pigmentation after five days of exposure. The pathological changes were degeneration and necrosis of surface skin cells, disappearance of cellular boundaries, displaying pink staining material, bleeding in the corium cells and lymph cell infiltration with accompanying formation of abscesses.

A single covered topical application to the skin of albino rabbits for four hours induced moderate to severe skin irritation including necrosis with slight to moderate oedema. A 10% solution (aqueous) produced slight redness and oedema after 1 application (for 4 hours) and a moderate burn after 2 applications.

According to the available studies GMA must be classified according to CLP as: Skin corrosive Category 1C (H314)

Eye damage

GMA induced slight to moderate conjunctivitis in the unwashed eye where slight corneal injury cleared in one week in a study resembling the OECD eye irritation study but which was very limitedly reported.

In inhalation study using rats, eye irritation was also induced. Acute exposure for 4 hours induced eye irritation at 1563 mg/m³ and 2394 mg/m³. Corneal opacity was also observed slightly at 610 mg/m³, and moderately at 1563 mg/m³ and 2394 mg/m³. These changes did not heal within 14 days post-exposure. In sub-acute study, rats were exposed at 58.2, 223 and 931 mg/m³, 6 hours/day, 5 days/week for 2 weeks. As a result, eye irritation and corneal clouding were observed at 931 mg/m³.

As GMA is corrosive to the skin, GMA is also considered to induce serious eye damage (CLP chapter 3.3.2.3).

Skin sensitisation

The substance is a rapid and moderate sensitiser in guinea pigs and humans.

According to the available studies GMA must be classified according to CLP for Skin sensitisation Category 1 (H317) in view of the results found both in humans and animals.

Respiratory sensitisation

Respiratory sensitisation was not observed in any of the acute and sub-acute inhalation studies. However, these studies are not developed to determine the respiratory sensitising potential of substances.

Repeated dose toxicity

The major toxicity was tissue damages in the first exposure sites such as forestomach by oral administration and respiratory tract by inhalation, due to the corrosiveness of the substance and NOAELs were 30 mg/kg for rat oral (systemic effects), 10 mg/kg bw/day for rat oral (local effects), 12 mg/m³ for rat inhalation (local effects) and 2.91 mg/m³ for rabbit inhalation (local effects).

Based on the available studies and based on the other classifications already presented, no further classification of GMA for this endpoint is required under CLP.

Mutagenicity

Genotoxicity studies on GMA *in vitro* showed positive results. In micronucleus tests *in vivo*, oral administration of GMA increased the frequency of micronucleated polychromatic erythrocytes at the highest dose only, while mostly negative results were shown in other *in vivo* genotoxicity studies including micronucleus tests by intraperitoneal administration and including the gene mutation study using transgenic Big Blue Fischer 344 rats. However, *in vitro* and *in vivo* studies indicate carboxylesterase mediated hydrolysis of GMA to glycidol. Glycidol, a metabolite of GMA, is classified as a Category 2 germ cell mutagen under CLP. Based on the available studies for GMA itself and read across data for glycidol, GMA is considered to be a substance with mutagenic potential for somatic cells. There is only very limited information regarding germ cell mutagenicity in which only a slight and not dose-related increase in unscheduled DNA synthesis was observed in the germ cells. This is not sufficient for category 1B.

According to the available studies GMA must be classified according to CLP for Germ Cell Mutagenicity Category 2 (H341).

Carcinogenicity

There are reports on chronic exposure studies with GMA, but each one has significant methodological deficiencies such that the conclusion is that there are no acceptable chronic studies with GMA. In a very limited one-year study (Hadidian et al., 1968), rats (3 males and 3 females) were dosed 5 days/week by gavage at 0.1 mg/kg. Groups of 15 male and 15 female rats were also dosed at 0.3 mg/kg. The authors concluded that no tissue effects related to the treatment were found. However the doses applied are considered to be too low. There was also a 26-weeks inhalation toxicity study at concentrations of 15.3 and 206 mg/m³ in rats and rabbits (Ouyang Guoshun et al., 1990). A wide range of toxic effects were observed in both species at both concentrations. However, because of the low purity of the material used, the author suggested that the effects may have been caused by the impurities present. Therefore it is questionable if the systemic toxicity was caused by GMA itself. Consequently a Read Across for GMA was used. Rationale: although the kinetics of carboxylesterase-mediated hydrolysis of GMA appear to be species dependent, the primary metabolite of GMA found in humans, rats and rabbits is glycidol (see chapter 4.1). In terms of Annex XI of REACH, this means the similarity is based on the formation of a breakdown product (a metabolite) of GMA that is a known carcinogen (Cat 1B). Chronic bioassay data were located for glycidol in rats and mice and show carcinogenicity.

According to the available studies on glycidol GMA must be classified according to CLP as Carcinogenic Category 1B (H350).

Reproductive toxicity

Oral toxicity study was performed on GMA in SD (Crj: CD) rats by an OECD combined repeated dose and reproductive/developmental toxicity screening test (OECD TG 422). Administration was conducted by gavage at doses of 10, 30 and 100 mg/kg/day from 14 days before mating to 14 days after mating in males and from 14 days before mating to day 3 of lactation in females. The fertility index (number of delivered animals/ number of mated animals) decreased significantly at 100 mg/kg.

Male mice injected i. p. with 5 consecutive daily doses of 0, 25, 50 or 100 mg/kg/day of GMA showed an increase in the percentage of abnormal sperm and decrease in the number of sperm. These results were confirmed in a subsequent study where mice were dosed i. p. with 0, 5, 25 or 100 mg/kg for five consecutive days. At 100 mg/kg, mice had decreased caudal epididymal weights, slightly lower testicular weights, decreased sperm counts and increased abnormal sperm. Mice given 25 mg/kg/day showed decreased sperm counts and increased abnormal sperm. These results might support the decreased fertility index of the rat study at 100 mg/kg/day.

Three reliable developmental studies by two different routes, oral and inhalation, indicated no teratogenicity even at the highest doses which showed maternal toxicity. The significant increase in fetal resorption was considered for classification for developmental toxicity. However, as this effect was not observed in the comparable OECD 422 study (same route and dose levels), only observed in the presence of maternal toxicity and the main metabolite glycidol has no classification for development, no classification is proposed.

However based on the available studies the substance must be classified under the CLP Regulation as Toxic to Reproduction Category 1B (H360F). In addition, glycidol, a metabolite of GMA, is classified as a Category 1B reproductive toxicant under CLP. Based on the available studies for GMA itself and read across data for glycidol, GMA is considered to be a reproductive toxicant. The relevant differentiation is effects on sexual function and fertility.

Other effects

There was no evidence of neurotoxic effects at any exposure level.

1.6 Current harmonised classification and labelling

1.6.1 Current classification and labelling in Annex VI, Table 3.1 in the CLP Regulation

- Acute Tox. 4* H302
- Acute Tox. 4* H312
- Acute Tox. 4* H332
- Eye Irrit. 2 H319
- Skin Irrit. 2 H315
- Skin Sens. 1

Note D applies (Annex VI, Part 1, section 1.1.3.1)

The * indicates a minimum classification (Annex VI, Part 1, section 1.2.1)

1.7 Current self-classification and labelling

1.7.1 Current self-classification and labelling based on the CLP Regulation criteria

Self-classification as applied by the registrants

- Acute Tox. 4 oral
- Acute Tox. 3 dermal
- No classification for Acute Tox inhalation but instead STOT SE 1
- STOT SE 1 (affected organs: respiratory tract; route of exposure: inhalation)
- Eye Dam. 1
- Skin Corr. 1C
- Skin Sens. 1
- Repr. 1B H360F
- Muta. 2
- Carc. 1B

Note D applies (Annex VI, Part 1, section 1.1.3.1)

Pictogram and signal word: GHS05, GHS06 and GHS08 and “Danger”

See table 3a/3b/3c for further details

2 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

The current harmonised classification is deemed by the registrants and the dossier submitter to be inadequate since:

- A change in an existing entry is considered justified due to new data becoming available after the current harmonised classification was agreed
- The classification as CMR substance is entirely missing
- The classifications for eye damage/irritation and for skin corrosion/irritation should become more strict (for eye from Cat. 2 to Cat. 1; for skin from Cat. 2 to Cat. 1C)
- The classification for acute dermal toxicity should become more strict (from Cat. 4 to Cat. 3)
- The classification for acute inhalation toxicity (Cat. 4) should be replaced by a STOT SE 1 (affected organs: respiratory tract; route of exposure: inhalation)

Part B.

SCIENTIFIC EVALUATION OF THE DATA

1 IDENTITY OF THE SUBSTANCE

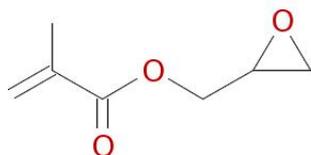
1.1 Name and other identifiers of the substance

The substance **2,3-epoxypropyl methacrylate** (GMA) is a mono constituent substance (origin: organic) having the following characteristics:

Table 4: Substance identity

EC number:	203-441-9
EC name:	2,3-epoxypropyl methacrylate
CAS number (EC inventory):	106-91-2
CAS number:	106-91-2
CAS name:	glycidyl methacrylate
IUPAC name:	oxiran-2-ylmethyl 2-methylprop-2-enoate
CLP Annex VI Index number:	607-123-00-4
Molecular formula:	C ₇ H ₁₀ O ₃
Molecular weight range:	142.1525

Structural formula:



1.2 Composition of the substance

Table 5: Constituents (non-confidential information)

Constituent	Typical concentration	Concentration range	Remarks
2,3-epoxypropyl methacrylate EC no.: 203-441-9	ca. 99.7 % (w/w)	≥ 99.5 — ≤ 100.0 % (w/w)	

Current Annex VI entry: see chapter 1.7.1

Table 6: Impurities (non-confidential information)

Impurity	Typical concentration	Concentration range	Remarks
Water	≤ 0.15 % (w/w)	≥ 0.0 — ≤ 0.15 % (w/w)	impurity from the synthesis
sodium chloride EC no.: 231-598-3	≤ 0.35 % (w/w)	≥ 0.0 — ≤ 0.35 % (w/w)	impurity from the synthesis

Current Annex VI entry: none

Table 7: Additives (non-confidential information)

Additive	Function	Typical concentration	Concentration range	Remarks
Mequinol EC no.: 205-769-8	stabiliser	ca. 0.01 % (w/w)	≥ 0.0050 — ≤ 0.015 % (w/w)	stabilizer to prevent hazardous polymerisation

Current Annex VI entry: 604-044-00-7:

Acute Tox. 4* H302

Skin Sens. 1 H317

Eye Irrit. 2 H319

1.2.1 Composition of test material

Information on the test material used in the different physic-chemical studies is given in each chapter when available while the information on the test material used in the toxicological studies is provided below for a global overview:

Table 8: Purity summary (blank if not available)

Study	Reference	Purity (%)
Toxicokinetic studies		
Intra-venous and subcutaneous (Rabbit)	Shi Tao et al. (1988)	
<i>In vitro</i> (rat, rabbit, human)	Domoradzki et al. (2004)	
Other	Bogdanffy (1987)	
	Dahl (1987)	
	Glatt (1984)	
	Mattes (1992)	
	Pacifici (1981)	
Acute toxicity studies		
Oral (rat, mouse, guinea pig)	Zdravko (1985)	
Oral (rat)	Olson (1960)	
Oral (rat, mouse)	Smyth (1969)	
Oral (rat)	EPA (1992)	97.8%
Inhalation (rat)	Nitschke (1990)	99.8%
Inhalation (rat)	Smyth (1969)	
Dermal (rabbit)		
Inhalation (rat, rabbit, guinea pig, dog)	Haag (1953)	
Intraperitoneal (rat, mouse)	Petrov (1973)	
Skin and/or eye irritation		
Skin (rabbit)	Ou-Yang (1988)	92%
Skin (rabbit)	Lockwood (1991)	
Skin and eye (rabbit)	Olson (1960)	
Skin (rabbit)	Smyth (1969)	

Table 8 cont: Purity summary (blank if not available)

Study	Reference	Purity (%)
Skin sensitisation studies		
Guinea pig	Dow (1990)	
Guinea pig	Ou-Yang (1988)	92%
Guinea pig	BIBRA (1988)	
Human	Dempsey (1982)	
Human	Matura (1995)	
Repeated dose toxicity		
Gavage (rat)	Ministry of Health and Welfare, Japan (1997)	99.93%
Oral (rat)	Hadidian (1968)	
Oral (rabbit)	Ou-Yang (1988)	92%
Inhalation (rat)	Landry (1996)	99.5%
Inhalation (rat)	Landry (1991)	99.61%
Inhalation (rat)	DuPont (1977)	
Inhalation (rat, rabbit)	Ouyang Guoshun (1990)	92%
Inhalation (rabbit)	Cieszlak (1996)	
<i>In vitro</i> mutagenicity studies		
	Ou-yang (1988)	92%
	Voogd (1981)	92%
	Linscombe (1995)	99.5%
	Ministry of Health and Welfare, Japan (1997)	99.93%
	Hude (1990)	97%
	Dorothy (1986)	98%
	Goodyear (1981)	
	Xie (1990a)	

Table 8: Purity summary (blank if not available)

Study	Reference	Purity (%)
	Hude (1991)	97%
	Xie (1990b)	
	Xie (1989)	
	Yang (1996)	
	Xie (1992)	
<i>In vivo</i> mutagenicity studies		
Intraperitoneal (mouse micronucleus assay)	Irwin (1990)	94%
Gavage (mouse micronucleus assay)	Ministry of Health and Welfare, Japan (1997)	99.93%
Inhalation (transgenic rodent mutation assay)	Gollapudi et al. (1999)	
Intraperitoneal (mouse micronucleus assay)	Ou-Yang (1988)	92%
Intraperitoneal (mouse micronucleus assay)	Lick (1995)	99.5%
Intraperitoneal (mouse micronucleus assay)	INBIFO (1979)	
(unscheduled DNA synthesis)	Xie (1990b)	
Carcinogenicity		
Gavage (rat)	Irwin et al. (1990)	94%
Reproductive toxicity studies		
Gavage (rat)	Ministry of Health and Welfare, Japan (1997)	99.93%
Intraperitoneal (mice)	Vedula (1994)	
Inhalation (rabbit)	Vedula (1995)	
Gavage (rat)	Ou-Yang (1988)	92%
Inhalation (rabbit)	Vedula (1996)	99.5%
Neurotoxicity studies		

Inhalation (rat)	Mattsson (1996)	
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The available information regarding the purity of the tested substance is limited with many studies for which the purity could not be retrieved. Also for several studies the applied test substance had a purity clearly below the purity as put on the market. Some test batches (example acute oral study EPA/OTS, 1992) contain epichlorohydrin and dichlorohydrin as impurities. Epichlorohydrin has a harmonised classification as Carc. 1B and may also show mutagenic properties seen the presence of an epoxy group. Also dichlorohydrin is classified as Carc. 1B. The presence of these impurities may affect the results of the tests especially for mutagenicity and carcinogenicity. As the impurities were not known in many studies this could potentially make the test results less reliable. However, as indicated in the relevant summaries, most key studies were performed with a test substance with a high purity considered relevant for the substance as put on the market. Therefore, the test results are considered relevant for the substance as put on the market.

1.3 Physico-chemical properties

Table 9: Summary of physico - chemical properties

Property	Value	Reference	Comment (e.g. measured or estimated)
State of the substance at 20°C and 101,3 kPa	Liquid	U.S. Coast Guard, Department of Transportation. CHRIS - Hazardous Chemical Data. Volume II. Washington, D.C.: U.S. Government Printing Office, 1984-5.	Measured
Melting/freezing point	-41.5 °C at 101.3 kPa	Lewis, R.J., Sr (Ed.). Hawley's Condensed Chemical Dictionary. 12th ed. New York, NY: Van Nostrand Rheinhold Co., 1993, p. 569	Measured
Boiling point	189 °C at 101.3 kPa	Dean, J.A. Handbook of Organic Chemistry. New York, NY: McGraw-Hill Book Co., 1987., p. 1-226	Measured
Relative density	1.07 at 20°C	Lide, D.R. (ed.). CRC Handbook of Chemistry and Physics. 76th ed. Boca Raton, FL: CRC Press Inc., 1995-1996., p. 3-291	Measured
Vapour pressure	420 Pa at 25 °C	OECD SIDS for Glycidyl methacrylate, 1999 (study by MITI, Japan)	Measured (98.2% purity)
Surface tension	25 mN/m at 20°C	U.S. Coast Guard, Department of Transportation. CHRIS - Hazardous Chemical Data. Volume II. Washington, D.C.: U.S. Government Printing Office, 1984-5.	Surface tension was measured for undiluted liquid
Water solubility	50 g/L at 25 °C	OECD SIDS for Glycidyl methacrylate, 1999 (study by MITI, Japan)	Measured

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Partition coefficient n-octanol/water	Log Kow (Pow): 0.96 at 25 °C	OECD SIDS for Glycidyl methacrylate, 1999 (study by MITI, Japan)	Measured (98.2% purity)
Flash point	76 °C at 1013 hPa	Kirk-Othmer Encyclopedia of Chemical Technology. 3rd ed., Volumes 1-26. New York, NY: John Wiley and Sons, 1978-1984., p. 15(81) 347	Closed cup (open cup was 84 degrees C)
Flammability	Flammable		Estimated. With a flashpoint of 76 (closed cup) – 84 (open cup) deg C (170 – 185 deg F) and a low volatility (vapour pressure of 4.2 x 100 Pa @ 25 deg C) a separate flammability test is not needed. GMA is a combustible liquid class IIIA and should be kept away from heat, sparks, flame and any source of ignition.
Explosive properties	Non explosive		There are no chemical groups associated with explosive properties in the molecule
Self-ignition temperature	389 °C at 1013 hPa	Seamanship Hazardous Chemical Database version 4.2.29	Measured
Oxidising properties	None		This substance is incapable of reacting exothermally with combustible materials on the basis of its chemical structure
Granulometry			The particle size distribution cannot be measured as the substance is a liquid.
Stability in organic solvents and identity of relevant degradation products			The stability of the substance in organic solvents is not considered to be critical.
Dissociation constant			It is scientifically not possible to perform the test (the substance is neither an acid nor a base).
Viscosity	Viscosity at 21°C: 5.481 mPa · s (dynamic)	U.S. Coast Guard, Department of Transportation. CHRIS - Hazardous Chemical Data. Volume II. Washington, D.C.: U.S. Government Printing Office, 1984-5.	Measured

2 MANUFACTURE AND USES

2.1 Manufacture

This substance is produced, handled and used in closed system under very well controlled conditions as a monomer unit to produce polymers or as an intermediate to produce further non-polymeric chemical products. The substance is not included in consumer products.

2.2 Identified uses

Only to be handled and used under very well controlled conditions in closed systems as industrial feedstock for further synthesis.

The substance is only approved and registered for a very limited number of industrial uses (PROC1/2/3/4/8b/9/15 and ERC 1/2/6a/6c/6d). Professional and consumer uses are advised against.

<i>PROC1</i>	<i>Use in closed process, no likelihood of exposure</i>
<i>PROC2</i>	<i>Use in closed, continuous process with occasional controlled exposure</i>
<i>PROC3</i>	<i>Use in closed batch process (synthesis or formulation)</i>
<i>PROC4</i>	<i>Use in batch and other process (synthesis) where opportunity for exposure arises</i>
<i>PROC8b</i>	<i>Transfer of chemicals from/to vessels/large containers at dedicated facilities</i>
<i>PROC9</i>	<i>Transfer of chemicals into small containers (dedicated filling line)</i>
<i>PROC15</i>	<i>Use as laboratory reagent</i>
<i>ERC1</i>	<i>manufacture</i>
<i>ERC2</i>	<i>formulation</i>
<i>ERC6a</i>	<i>Industrial use as intermediate for further processing (not under strictly controlled conditions)</i>
<i>ERC6c</i>	<i>Industrial use of monomers for manufacture of thermoplastics</i>
<i>ERC6d</i>	<i>Industrial use of process regulators for polymerisation processes in production of resins, rubbers, polymers</i>

Note that all PROCs can be combined with all ERCs

3 CLASSIFICATION FOR PHYSICO-CHEMICAL PROPERTIES

Table 10: Summary table for relevant physico-chemical studies

Method	Results	Remarks	Reference
Not relevant	Not relevant	Not relevant	Not relevant

3.1 Hazard classes for physico-chemical properties

Not relevant for the substance

3.1.1 Summary and discussion of classification for physico-chemical properties

Not relevant for the substance

3.1.2 Comparison with criteria

Not relevant for the substance

3.1.3 Conclusions on classification and labelling

Note D is warranted (see Table 8) because GMA contains a stabiliser (polymerisation inhibitor) to prevent spontaneous polymerisation.

4 HUMAN HEALTH HAZARD ASSESSMENT

4.1 Toxicokinetics (absorption, metabolism, distribution and elimination)

4.1.1 Non-human information

The results of studies on absorption, metabolism, distribution and elimination are summarised in the following table:

Table 11a: Summary of toxicokinetic studies

Method	Results	Remarks	Reference
Rabbit intra-venous and subcutaneous Doses/conc.: intravenous: 200 mg/kg subcutaneous: 800 mg/kg		2 (reliable with restrictions) key study experimental result Test material: GMA	Shi Tao et al. (1988) OECD (1999)
<i>in vitro</i> study	Main ADME results: metabolism: Under all circumstances	2 (reliable with restrictions)	Domoradzki et al. (2004)

Method	Results	Remarks	Reference
rat, rabbit and human direct addition to liver homogenate and nasal epithelial tissue Exposure regime: 6 hours Doses/conc.: 2 mM and 40 mM no data	within 30-120 minutes only one metabolite appeared, tentatively identified as glycidol Metabolites identified: yes Details on metabolites: Only glycidol (EINECS 209-128-3) At an initial starting concentration of GMA at 2 mM, half-lives of GMA hydrolysis were faster in incubations with rat and rabbit (30 min) tissue in comparison to humans (2 hrs). Carboxylesterase activity was lower in humans than in rats and rabbits.	key study experimental result Test material: GMA Form: liquid	
<i>in vitro</i> study	General information on the distribution of carboxylesterase and epoxy hydratase in different tissues and species.	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Bogdanffy (1987) Dahl (1987) Glatt (1984) Mattes (1992) Pacifici (1981) OECD (1999)

Key study summaries

Shi Tao et al. (1988)

Toxicokinetics of GMA was investigated in rabbits. After an intravenous injection at 200 mg/kg, the concentration-time curve of this substance could exactly fit the two-compartment open model, and over 95 % of the parent compound had disappeared from the blood within 10 minutes. Following a subcutaneous injection at 800 mg/kg, the toxicokinetics appeared to fit a first-order absorption one-compartment open model. This substance was metabolized by a first-order process in incubation with whole blood, plasma, erythrocyte suspension, and homogenates of brain, heart, liver, lung, spleen, kidney, small intestine, and muscle. The highest rate of elimination had been found in blood and liver homogenate. The subcutaneous co-administration of tri-o-cresyl-phosphate (a carboxylesterase inhibitor) with this substance resulted in about a tenfold increase in the maximum blood concentrations of this substance, compared to those of animals dosed with this substance alone. *In vitro*, metabolism rate could be also decreased by tri-o-cresyl phosphate.

Domoradzki et al. (2004)

The purpose of this investigation was to study the metabolism of GMA *in vitro* using tissues from humans, rats, and rabbits. Differences in carboxylesterase and epoxide hydrolase activities in tissues from these species may result in differences in formation of glycidol, methacrylic acid, GMA-diol and glycerol. Results from this study may provide a basis to judge the relative sensitivity of humans to rabbits and rats for the generation of toxic effects with GMA.

In vitro incubations of GMA with nasal tissue preparations and liver homogenate from human, rat and rabbit resulted in the formation of only one metabolite. This metabolite was tentatively identified as glycidol based on retention time match with ^{14}C glycidol. No other metabolites were found. At an initial starting concentration of GMA at 2 mM, half-lives of GMA hydrolysis were faster in incubations with rat and rabbit tissue than with human tissue (completed within 30 minutes for rat and rabbit versus 2 hours for humans). At a concentration of 2 mM, GMA metabolism in liver from the fastest to the slowest was rabbit >rat>>human (Table 11b). Even though cellular fractions were not tested for epoxide hydrolase activity, carboxylesterase activities using 4-nitrophenyl acetate as a substrate were measured in human, rat and rabbit nasal tissue homogenates and human nasal tissues used in this study were found to be lower in this activity than in the other species (Table 11c). Based on *in vitro* results obtained in this study, it appears that GMA is principally metabolized to one metabolite (glycidol) and that glycerol and GMA-diol were not formed. Since glycerol or GMA-diol were not observed as metabolites, it appears that epoxide hydrolysis was not a major *in vitro* route of metabolism for GMA using rat, rabbit and human tissue homogenate preparations in the incubations, although epoxide hydrolase activity in these preparations was not determined.

Figure 1: Carboxylesterase activity in nasal tissues (Domoradzki et al. 2004)

Carboxylesterase Activity in Nasal (Respiratory and Olfactory) Tissue of Human, Rabbit and Rat Using the Substrate, 4-Nitrophenyl Acetate.

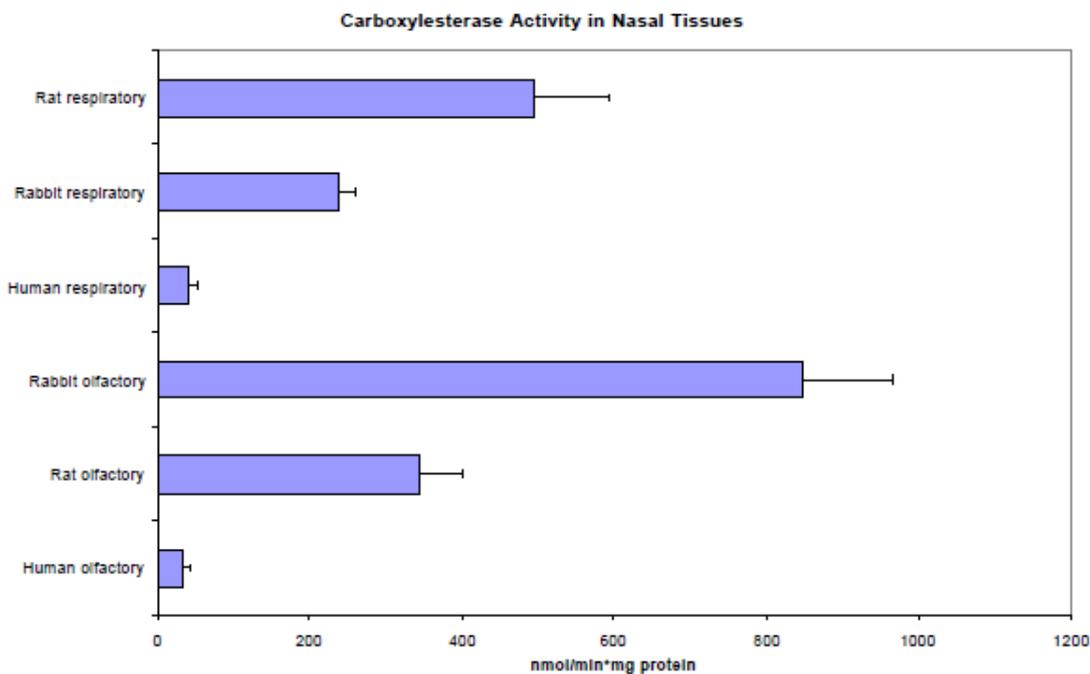


Table 11b: Metabolism of GMA in nasal epithelium and liver (Domoradzki et al. 2004)

METABOLISM OF GLYCIDYL METHACRYLATE IN NASAL EPITHELIUM AND LIVER OF FISCHER 344 RATS, NEW ZEALAND RABBITS, AND HUMANS

Table 1. Glycidyl Methacrylate and a Metabolite Present in *In Vitro* Preparations of Human, Rabbit and Rat Liver Incubated for Varying Times Following the Addition of ¹⁴C-Glycidyl Methacrylate at a 2 mM Concentration.

Sample Name	Incubation Time (minutes)	Metabolite ^a	GMA ^a
Rat Liver Control (Denatured) ^c	60	0	100
Rat Liver	1	4	96
	2	10	90
	5	26	74
	10	43	57
	30	89	11
	45	97	3
	60	99	1
Rabbit Liver Control (Thermal) ^b	60	0	100
Rabbit Liver Control (Denatured) ^f	60	0	100
Rabbit Liver	1	10	90
	2	22	78
	5	55	45
	10	78	22
	30	98	2
	45	100	0
	60	100	0
Human Liver Control (Thermal) ^b	360	0	100
Human Liver Control (Denatured) ^c	360	NA ^e	NA ^e
Human Liver 221981	1	2	98
	2	2	98
	5	5	95
	10	9	91
	30	21	79
	45	25	75
	60	39	61
	120	79	21
	240	87	13
	360	85	15
Human Liver Control (Denatured) ^c	360	0	100
Human Liver 624971	1	3	97
	2	4	96
	5	8	92
	10	19	81
	30	36	64
	45	51	49
	60	68	32
	120	96	4
	240	99	1
	360	99	1

^a Data are peak area percent response of ¹⁴C-RAM^b ¹⁴C-GMA in buffer at 37 degrees centigrade for 60 minutes^c denatured by heating at 100°C for 10 minutes and then incubated at 37°C for 60 minutes^d sample lost^e not applicable

Table 11c: Carboxylesterase activity measure (Domoradzki et al. 2004)

METABOLISM OF GLYCIDYL METHACRYLATE IN NASAL EPITHELIUM AND LIVER OF FISCHER 344 RATS, NEW ZEALAND RABBITS AND HUMANS

Table 11. Carboxylesterase Activity Measured with the Substrate, 4-Nitrophenyl Acetate, in Rat, Rabbit, and Human Nasal Tissues

Tissue Homogenate	mg protein	mOD/min	OD/min	nmoles/min	nmol/min*mg protein	Mean	SD
Rat respiratory	0.001178	30.2	0.0302	0.46037	390.803	493.602	98.08
		49.43	0.04943	0.75351	639.649		
	0.003534	136.12	0.13612	2.07500	587.153		
		110.42	0.11042	1.68323	476.296		
	0.00589	168.48	0.16848	2.56829	436.043		
		166.79	0.16679	2.54253	431.669		
Rat olfactory	0.003095	55.69	0.05569	0.84893	274.292	343.899	56.28
		89.81	0.08981	1.36905	442.344		
	0.009285	205.31	0.20531	3.12973	337.073		
		215.49	0.21549	3.28491	353.787		
	0.015475	314.69	0.31469	4.79710	309.991		
		351.15	0.35115	5.35290	345.906		
Rabbit respiratory	0.00393	67.51	0.06751	1.02912	261.862	237.146	23.70
		68.71	0.06871	1.04741	266.516		
	0.01179	171.46	0.17146	2.61372	221.690		
		188.68	0.18868	2.87622	243.954		
	0.01965	272.61	0.27261	4.15564	211.483		
		280.2	0.2802	4.27134	217.371		
Rabbit olfactory	0.003946	265.67	0.26567	4.04985	1026.317	846.955	120.13
		216.41	0.21641	3.29893	836.019		
	0.011838	682.14	0.68214	10.39848	878.398		
		702.48	0.70248	10.70854	904.590		
	0.01973	884.71	0.88471	13.48643	683.550		
		974.41	0.97441	14.85381	752.854		
Human respiratory	0.03069	90.666	0.090666	1.38210	45.034	41.727	13.00
		128.198	0.128198	1.95424	63.677		
	0.06138	115.981	0.115981	1.76800	28.804		
		180.364	0.180364	2.74945	44.794		
	0.09207	237.876	0.237876	3.62616	39.385		
		173.134	0.173134	2.63924	28.666		
Human olfactory	0.00298	8.629	0.008629	0.13154	44.141	32.802	11.00
		9.714	0.009714	0.14808	49.691		
	0.00596	7.014	0.007014	0.10692	17.940		
		8.094	0.008094	0.12338	20.702		
	0.00894	21.511	0.021511	0.32791	36.679		
		21.355	0.021355	0.32553	36.413		
	0.0149	27.751	0.027751	0.42303	28.392		
		27.819	0.027819	0.42407	28.461		

Supporting studies

Bogdanffy, 1987

Inhalation exposure of rats and mice to glycol ether acetates and acrylate esters causes degeneration of the olfactory epithelium but not of the respiratory epithelium. Since these compounds are metabolized via carboxylesterase to acids that are toxic to the olfactory epithelium, the activity and cellular distribution of carboxylesterase in the nasal passages of rats and mice were studied.

Olfactory mucosal carboxylesterase in both rats and mice was found to have a Vmax value for the hydrolysis of p-nitrophenyl butyrate approximately 3 to 6 times larger than that for respiratory mucosa. Similarly, the second-order rate constant for binding and catalysis, was approximately four

times greater in olfactory mucosa than in respiratory mucosa of both rats and mice. These data demonstrate that the olfactory mucosa of rats and mice hydrolyze carboxylesters more efficiently than the respiratory mucosae. Enzyme histochemistry was employed to identify the individual cells within the respiratory and olfactory mucosae which contain carboxylesterase activity. All cell types of the respiratory epithelium had some carboxylesterase activity, with varying intensities between individual cell populations. Ciliated and cuboidal epithelial cells were most active in this region. In the olfactory mucosa, however, Bowman's glands stained most intensely, sustentacular cells demonstrated moderate activity, and no activity was detectable in olfactory sensory cells. Together, these data quantitate carboxylesterase activity in nasal mucosal homogenates and localize the enzyme in individual cell types. The data suggest that olfactory mucosa may metabolize carboxylesters to acids more readily than respiratory mucosa. However, such metabolism does not occur in the target cell population, the olfactory sensory neurons, raising the possibility of intercellular migration of toxic acid metabolites.

Dahl, 1986

Esters are a widespread class of organic compounds found both in industry and the environment. Because esters are often volatile and, therefore, readily inhaled, the capacity of respiratory tract tissues as well as liver S-9 homogenates from rats, rabbits, and Syrian hamsters to hydrolyze a variety of esters was investigated. A new technique to determine hydrolysis rates by measuring carboxylic acid residues using ion chromatography was proven effective. The results indicated that esters, including potentially carcinogenic β -lactones, are readily hydrolyzed by respiratory tract enzymes. Species and tissue differences were apparent. The nasal ethmoturbinates had especially high levels of esterase activity with tissue weight-normalized activities from rabbits and hamsters for most substrates exceeding all other tissues tested, including liver. Phenyl acetate was the most rapidly hydrolyzed by ethmoturbinatate tissue of the esters tested. Among straight chain aliphatic alcohol acetates, hydrolysis rates increased with carbon number up to pentyl alcohol and then decreased. Branched 4-carbon alcohol acetates were less rapidly hydrolyzed than n-butyl acetate. Correlation of hydrophobicity constants with hydrolysis rates indicated that, for the straight chain aliphatic acetates, a bilinear model best fit the data.

Mattes, 1992

Carboxylesterase activity in the rat nasal mucosa plays an important role in the response of this tissue to certain toxic inhaled esters. We have examined this activity in extracts of both Fischer-344 rat and human nasal tissue using the substrate α -naphthyl butyrate, the same substrate as that used for histochemical analysis of this activity. We find substantially higher activity in rat nasal extracts than in human nasal extracts. The Michaelis constant (K_m), however, is approximately the same for both activities and is significantly less than that reported for rat nasal carboxylesterase activity using dibasic esters as a substrate. p-Nitrophenyl butyrate is a competitive inhibitor of the rat α -naphthyl butyrate esterase, but surprisingly has no effect on the human activity. The assay reported here should prove a powerful tool in the development of a valid in vitro nasal toxicity assay system that uses cultured rat or human cells expressing carboxylesterase activity.

Pacifici, 1981

1. Glutathione transferase and epoxide hydratase activities were determined in liver, lung and kidney of nine species including man with [7-³H]styrene oxide as substrate.
2. Activity was detectable in all tissues. The activities of both enzymes were higher in the liver of all species than in either kidney or lung.

3. The baboon had the highest hepatic epoxide hydratase activity, 31 μ m 2nmol/mg per min, while the mouse had the lowest hepatic activity, 1.9 μ m 0.1 nmol/mg per min.
4. Rodent species had higher glutathione transferase activity than non-rodent species, mouse liver having the highest activity, 143 μ m 13nmol conjugate/mg per min.
5. Taking all the data into account, it is concluded that no single species of those studied is a suitable model for the disposition of epoxides in man.

4.1.2 Human information

See above mentioned Domoradzki et al. (2004) study.

4.1.3 Summary and discussion on toxicokinetics

Limited toxicokinetic and metabolism data are available for GMA. The metabolism of GMA in mammals is hypothesized to proceed by at least two different and competing enzyme systems, epoxide hydrolase and non-specific carboxylesterases. The two metabolic routes are shown in the Figure 2. Metabolism of GMA by carboxylesterase would result in formation of glycidol and methacrylic acid as metabolites, while initial metabolism by epoxide hydrolase would result in the formation of glycerol methacrylate. The relative speed at which the two competing metabolic reactions occur in different tissues and species is likely to be important for understanding the toxicity of GMA.

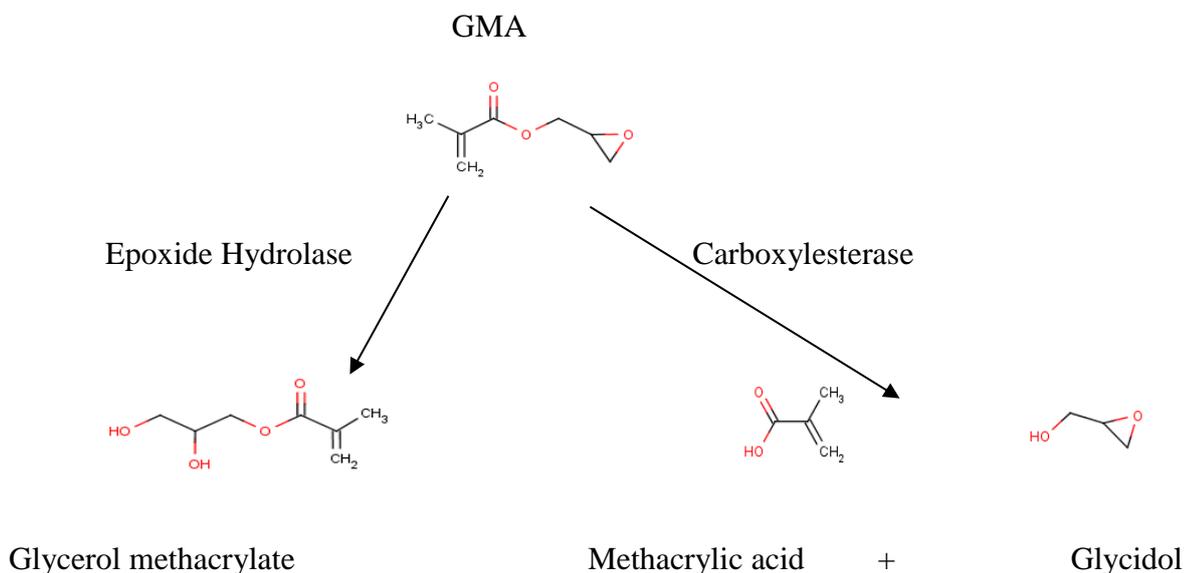


Figure 2: Hypothesized metabolism of GMA in mammals

Species differences in the activity of these enzymes suggest that the carboxylesterase route of metabolism may predominate in the nasal tissue of rabbits (yielding glycidol and methacrylic acid) while the epoxide hydrolase route was hypothesized to predominate in rats and humans (producing

glycerol methacrylate, then glycerol and methacrylic acid by carboxylesterase) (Bogdanffy et al.: 1987, Dahl et al.: 1987, Glatt et al.: 1984, Mattes and Mattes: 1992, Pacifici et al.: 1981).

Toxicokinetics of GMA were investigated in rabbits. After an intravenous injection at 200 mg/kg, over 95 % of the parent compound disappeared from the blood within 10 minutes according to a two-compartment open model. Following a subcutaneous injection at 800 mg/kg, the toxicokinetics appeared to fit a first-order absorption one-compartment open model. This substance was metabolized by incubation with whole blood, plasma, erythrocyte suspension, and homogenates of various tissues. The subcutaneous co-administration of tri-*o*-cresyl-phosphate (a carboxylesterase inhibitor) with this substance resulted in about a ten-fold increase in the maximum blood concentrations, compared to those of animals dosed with this substance alone (Shi Tao et al. 1988). This finding suggests a key role of carboxylesterase in GMA metabolism and although metabolites were not specifically measured, could implicate glycidol as that metabolite.

More definitive work on the metabolism of GMA was studied by Domoradzki et al. (2004) *in vitro* using liver homogenate and nasal epithelial tissues from humans, rats and rabbits. Radiolabeled GMA [¹⁴C 1,3- glycidyl] was used in this study and was 92% radio chemically pure. *In vitro* incubations of ¹⁴ C GMA with tissue preparations from human, rat and rabbit resulted in the formation of only one metabolite. This metabolite was tentatively identified as glycidol based on retention time match with ¹⁴C-glycidol. At an initial starting concentration of GMA at 2 mM, half-lives of GMA hydrolysis were faster in incubations with rat and rabbit tissue. Although the biotransformation was faster in rats and rabbits as compared to humans (completed within 30 minutes versus 2 hours), under all circumstances only one metabolite appeared which was tentatively identified as glycidol (EINECS 209 -128 -3).

Overall, the available studies show that GMA is metabolised into glycidol. Metabolism of GMA to glycidol has ramifications for hazard identification. Glycidol has a harmonised classification according to CLP as carcinogenic (category 1B), germ cell mutagenic (category 2) and toxic to reproduction (category 1B).

4.2 Acute toxicity

4.2.1 Non human information

4.2.1.1 Acute toxicity: oral

Table 12a: Summary table of relevant acute toxicity studies (oral)

Method	Results	Remarks	Reference
Rat oral: unspecified exposure regimen Dose: no data	LD50: 597 mg/kg bw	4 (not assignable) supporting study experimental result Test material: GMA	Zdravko (1985) OECD (1999)
Rat (n=2 per dose) oral: 10% in corn oil Dose: 126, 252, 500 and 1000 mg/kg bw	LD50: ca. 700 mg/kg bw	3 (not reliable) supporting study experimental result Test material : GMA	Olson (1960) Smyth (1969) OECD (1999)
Rat (albino) oral: unspecified dose	LD50: 451 mg/kg bw	4 (not assignable) supporting study experimental result Test material: GMA (purity 97.8%) Mixture: 0.3% epichlorohydrin + 0.6% dichlorohydrin	EPA (1992) OECD (1999)
Mouse oral: unspecified dose	LD50: 390 mg/kg bw	4 (not assignable) supporting study experimental result Test material: GMA	Zdravko (1985) OECD (1999)
Rat oral: unspecified dose	LD50: 1050 mg/kg bw	4 (not assignable) supporting study experimental result Test material: GMA	Smyth (1969) OECD (1999)
Guinea pig oral: unspecified dose	LD50: 697 mg/kg bw	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Zdravko (1985) OECD (1999)

All available acute oral studies are old and have limitations either in reporting (score 4) or in the conduct of the study (score 3). The OECD has adopted an oral LD50 value for GMA of 597 mg/kg bw for rat (Zdravko et al.: 1985). All other acute oral studies resulted in LD50 values in the same range.

4.2.1.2 Acute toxicity: inhalation

Table 12b: Summary table of relevant acute toxicity studies (inhalation)

Method	Results	Remarks	Reference
Rat (Fischer 344)	LC0 (4 h): > 2394 mg/m ³	2 (reliable with	Nitschke (1990)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
5 male+ 5 female/group inhalation: vapour (whole body), single 4-hour period, 14-day post- exposure observation period Analytical concentration: 105, 269, 412 ppm (310, 1563, 2394 mg/m ³) OECD Guideline 403 (Acute Inhalation Toxicity)	(412 ppm) (male/female): no mortality	restrictions) key study experimental result Test material: GMA purity 99.8%	OECD (1999) Nitschke (1999)
Rat 6 per group inhalation: vapour Concentration: saturated	Concentrated vapour inhalation by rats maximum for no deaths = 2 hours There was no recorded inhalation metered vapour concentration mortality in rats.	4 (not assignable) supporting study experimental result Test material: GMA	Smyth (1969) OECD (1999)
Rat inhalation: vapour Concentration: 1400 mg/m ³ air	LCL ₀ (6 h): 1400 mg/m ³ air Changes in lungs, thorax, respiration, etc.	3 (not reliable) supporting study experimental result Test material: GMA	Haag (1953) OECD (1999)
Rabbit inhalation: vapour Concentration: 1400 mg/m ³ air	LCL ₀ (6 h): 1400 mg/m ³ air Changes in lungs, thorax, respiration, etc.	3 (not reliable) supporting study experimental result Test material: GMA	Haag (1953) OECD (1999)
Guinea pig inhalation: vapour Concentration: 1400 mg/m ³ air	LCL ₀ (6 h): 1400 mg/m ³ air Changes in lungs, thorax, respiration, etc.	3 (not reliable) supporting study experimental result Test material: GMA	Haag (1953) OECD (1999)
Dog inhalation: vapour Concentration: 1400 mg/m ³ air	LCL ₀ (6 h): 1400 mg/m ³ air Changes in lungs, thorax, respiration, etc.	3 (not reliable) supporting study experimental result Test material: GMA	Haag (1953) OECD (1999)

LCL₀= (lethal concentration low): The lowest concentration of a substance, in air, that causes death to mammals in acute (<24h) or sub-acute or chronic (>24 h) exposure.

Nitschke (1990)

The purpose of this study was to determine the acute toxicity of inhaled glycidyl methacrylate vapors. Groups of 5 male and 5 female rats were exposed to nominal concentrations of 105, 255 or 412 ppm (0.61, 1.48 or 2.30 mg/l, respectively) of the test material for a single 4-hour period. The highest concentration was the maximum practically attainable vapor concentration.

All animals survived the exposure and 14-day post-exposure observation period. At 412 ppm, labored breathing was noted clinically at the end of the exposure period and eye irritation was noted in a few animals for several days following exposure. Body weights of animals exposed to 412 ppm were decreased as much as 15% from pre-exposure values but thereafter the animals achieved expected body weight gains. Similar but less severe effects were noted in animals exposed to 269 ppm. At the lowest concentration, 105 ppm, a very slight transitory body weight loss of 3% was noted only on the day following exposure to the test material, but thereafter the animals achieved expected body weight gains. Corneal opacities were noted in all but one rat exposed to the various concentrations of glycidyl methacrylate. While these lesions were consistent with corneal dystrophy, a spontaneous lesion in Fischer 344 rats, the high frequency of the corneal opacity is considered to be exposure-related.

In another inhalation toxicity study, acute exposure to rats with saturated vapour of this substance for 2 hours resulted in no deaths (Smyth et al.: 1969). It was reported that saturated vapour of GMA at 20 °C was 474 ppm (2754 mg/m³) (No reference in Draft Workplace Environmental Exposure Level Guide: 1999). Higher concentrations including the testing of aerosols were not performed. No other additional information or further details were provided.

4.2.1.3 Acute toxicity: dermal

Table 12c: Summary table of relevant acute toxicity studies (dermal)

Method	Results	Remarks	Reference
Rabbit	LD50: 480 mg/kg bw	4 (not assignable)	Smyth (1969)
Dermal, no data		key study	OECD (1999)
Dose: no data		experimental result	
		Test material: GMA	

The available information on the only available study is very limited and would normally not be usable for classification. However, there is already a harmonised minimum classification of GMA for acute dermal toxicity. It is likely that this study was the basis for the current harmonised classification. Therefore, this study can be used to adapt the current minimum classification.

4.2.1.4 Acute toxicity: other routes

Table 12d: Summary table of relevant acute toxicity studies (other routes)

Method	Results	Remarks	Reference
Rat Intraperitoneal Dose:	LD50: 290 mg/kg bw	4 (not assignable) supporting study experimental result Test material: GMA	Petrov (1973) OECD (1999)
Mouse Intraperitoneal Dose:	LD50: 350 mg/kg bw	4 (not assignable) supporting study experimental result Test material: GMA	Petrov (1973) OECD (1999)

4.2.2 Human information

No relevant information available

4.2.3 Summary and discussion of acute toxicity

All available acute oral studies are old and have limitations either in reporting (score 4) or in the conduct of the study (score 3). A key study can therefore not be determined. The OECD has adopted an oral LD50 value for GMA of 597 mg/kg bw for rat (Zdravko et al.: 1985). Although the available studies are only limitedly reported, all studies provide the same range of LD50 values of 390 – 1050 mg/kg bw. Therefore, overall the level of evidence is considered sufficient to allow classification for acute oral toxicity.

In an inhalation toxicity study by OECD TG 403 (key study), there was no mortality observed in rats exposed for 4 hours at 2394 mg/m³, the highest practically attainable vapour concentration. Change of respiration (laboured breathing) and eyes (irritation and corneal opacity), and decrease in body weight were induced even at the lowest concentration of 1563 mg/m³ (Nitschke et al.: 1990).

In another inhalation toxicity study, acute exposure to rats with saturated vapour of this chemical substance for 2 hours resulted in no deaths (Smyth et al.: 1969). It was reported that saturated vapour of glycidyl methacrylate at 20 °C was 474 ppm (2754 mg/m³) (No reference in Workplace Environmental Exposure Level Guide: 1999). Overall, higher concentrations including the testing of aerosols were not performed. This study is considered as supportive only because only very limited information is available and especially information on the mortality at exposure duration longer than 2 hours is missing.

The available information on the only available acute dermal study is very limited and would normally not be usable for classification. However, there is already a harmonised minimum

classification of GMA for acute dermal toxicity. It is likely that this study was the basis for the current harmonised classification. Therefore, this study can be used to adapt the current minimum classification. The dermal LD50 for rabbits in this study was 480 mg/kg bw (Smyth et al.: 1969).

Table 13: Summary of LD50s

Routes	Strain	Type	Values	Reference
Oral	Rats	LD50	597 mg/kg	Zdravko, 1985
	Rats	LD50	about 700 mg/kg	Olson, 1960; Smyth, 1969
	Rats*	LD50	451 mg/kg	EPA/OTS, 1992
	Mice	LD50	390 mg/kg	Zdravko, 1985
	Mice	LD50	1050 mg/kg	Smyth, 1969
	Guinea pigs	LD50	697 mg/kg	Zdravko, 1985
Inhalation	Rats	LC0	2394 mg/m ³ /4hr	Nitschke, 1990
	Rats**	LCL ₀	1400 mg/m ³ /6hr	Haag, 1953
	Rabbits**	LCL ₀	1400 mg/m ³ /6hr	Haag, 1953
	Guinea pigs**	LCL ₀	1400 mg/m ³ /6hr	Haag, 1953
	Dogs**	LCL ₀	1400 mg/m ³ /6hr	Haag, 1953
	Rats	LCL ₀	saturated/2 hr	Smyth, 1969
	Dermal	Rabbits	LD50	480 mg/kg

LC0 = lethal concentration 0%, the calculated concentration at which none of the population is expected to die.

LCL₀ = (lethal concentration low): The lowest concentration of a substance, in air, that causes death to mammals in acute (<24h) or sub-acute or chronic (>24 h) exposure.

* as a mixture of GMA (97.8 %), epichlorohydrin (0.3 %) and dichlorohydrin (0.6 %).

** Changes in lungs, thorax, respiration, etc. were observed.

4.2.4 Comparison with criteria

The oral LD50 of all studies are all between the borders of Acute oral category 4 of 300 to 2000 mg/kg bw.

The dermal LD50 of the only study (Smith, 1969) of 480 mg/kg bw is between the borders of Acute dermal category 3 of 200 to 1000 mg/kg bw.

The available inhalation studies indicate no mortality up to the saturated vapour pressure. Aerosols were not tested.

4.2.5 Conclusions on classification and labelling

Based on the available data GMA must be classified according to CLP as

- Acute toxicity oral Category 4 (H302) (the LD50 values of all available studies are between the Category 4 limits of 300 and 2000 mg/kg bw)

- Acute toxicity dermal Category 3 (H311) (the actual LD50 value of 480 mg/kg bw lies between the Category 3 limits of 200 and 1000 mg/kg bw)
- No classification for Acute toxicity inhalation is required since the LC0 in rats exposed for 4 hours lies at 2394 mg/m³, the highest practically attainable vapour concentration. As higher concentrations including aerosols were not tested, it is concluded that no classification for acute inhalation toxicity is based on absence of data.

4.3 Specific target organ toxicity – single exposure (STOT SE)

4.3.1 Summary and discussion of Specific target organ toxicity – single exposure

This assessment focusses on the inhalation route as a classification for acute oral and dermal toxicity is already proposed based on mortality. An additional classification for STOT SE for these routes is considered a double classification.

Key Studies

Laboured breathing was induced in rats by acute inhalation exposure for 4 hours at 1563 mg/m³ and 2394 mg/m³ (Nitscke et al. 1990). In another acute inhalation study, changes in lungs, thorax, respiration, etc. were observed in rats, rabbits, guinea pigs and dogs (Haag, 1953). In this study, exposure was conducted at 1400 mg/m³ for 6 hours. No further details are available. These changes may have resulted from respiratory irritation of this substance. The criteria for STOT SE Category 1 is Inhalation (rat) vapour mg/L/4h Concentration $\leq 10 \text{ mg/l} = 10000 \text{ mg/m}^3$.

Supporting Studies

In inhalation repeated dose toxicity studies, there were many changes in the respiratory tract, such as noisy and difficult respiration (mouth breathing), and hyperplasia, necrosis and inflammation in nasal tissues. In one sub-acute toxicity study, rabbits were exposed at 2.9, 12, 29 or 60 mg/m³ (6 hours/day, 5 days/week) for 13 consecutive days. Treatment-related degeneration of the nasal olfactory epithelium was observed at 12 mg/m³. At 29 and 60 mg/m³, there were olfactory epithelial degeneration, and hyperplasia, erosion, ulcers and inflammation of the nasal epithelium. After a 4-week recovery period, there was reversibility of these changes except for olfactory epithelial degeneration observed at 29 and 60 mg/m³, which showed only partial reversibility. At 12 mg/m³, nasal tissue was indistinguishable from controls at one-month post-exposure (Cieszlak et al., 1996).

Rats exposed to GMA at concentrations of 58.2, 233 or 931 mg/m³ (7.09, 28.4 or 113 mg/kg/day, respectively) for 2 weeks (6 hours/day, 5 days/week) (Landry et al.: 1991) showed general debilitation with noisy and difficult respiration (mouth breathing) was observed at 931 mg/m³ in day 4. The animals at 931 mg/m³ were terminated early on day 4 because of the severity of the respiratory effects. Microscopically, there was severe multifocal necrosis and inflammation of the olfactory and respiratory epithelium in the nasal cavity and also effects on the lungs including congestions, inflammation and necrosis. At 233 mg/m³, there were slight to moderate multifocal necrosis, and inflammation of the respiratory and olfactory nasal epithelium. At 58.2 mg/m³, microscopically there was very slight multifocal necrosis of individual respiratory epithelial cells in 3 of 5 males and in 2 of 5 females. These changes in respiratory tract were considered due to irritation of GMA. There were no histopathological changes in any other tissues. Therefore, 58.2 mg/m³ (7.09 mg/kg/day) was considered to be LOAEL because of tissue damages in respiratory tract.

Rabbits exposed to 2.91, 11.6, 29.1, 58.2 mg/m³ (6 hours/day, 5 days/week) (Cieszlak et al., 1996) showed treatment-related degeneration of the nasal olfactory epithelium was at 11.6 mg/m³.

Olfactory epithelial degeneration, and hyperplasia, erosions, ulcers and inflammation of the nasal epithelium were observed at 29.1 and 58.2 mg/m³. After 4-week recovery period, there was complete reversibility of these changes except for olfactory epithelial degeneration observed at 29.1 and 58.2 mg/m³, which showed only partial reversibility. At 11.6 mg/m³, nasal tissue was indistinguishable from controls at one month post-exposure. 2.91 mg/m³ (0.26 mg/kg/day) was considered to be NOAEL. Unfortunately purity of chemical and GLP were not mentioned.

A sub-chronic inhalation toxicity study in rats at concentrations of 2.9, 12 or 87 mg/m³ (0.35, 1.46 or 10.6 mg/kg/day, respectively) for 13 weeks (6 hours/day, 5 days/week) (Landry et al.: 1996). There were no treatment related in-life observations, and no significant treatment-related effects on body weight, urinalysis, clinical chemistry or hematology parameters, as well as gross pathologic changes or organ weights at any exposure level. Treatment-related effects were limited to hyperplasia of respiratory epithelium of the nasal tissues in all animals at 87 mg/m³. In all affected animals, the hyperplastic respiratory epithelium was approximately two to three times as thick as in control animals, and was located in the anterior portions of the nasal passages, involving the tips of the turbinates and the lateral walls of the nasal passages. These changes were considered to result from respiratory irritation. Therefore, NOAEL was considered 12 mg/m³ (1.46 mg/kg/day) for both sexes.

4.3.2 Comparison with criteria

Specific target organ toxicity (single exposure) is defined in the CLP Regulation as specific, non-lethal target organ toxicity arising from a single exposure to a substance or mixture. All significant health effects that can impair function, both reversible and irreversible, immediate and/or delayed and not specifically addressed in other sections are to be covered.

Specific target organ toxicity following a repeated exposure is classified as described in Specific target organ toxicity — Repeated exposure and is therefore excluded here.

The adverse health effects produced by a single exposure include consistent and identifiable toxic effects in humans, or, in experimental animals, toxicologically significant changes which have affected the function or morphology of a tissue/organ, or have produced serious changes to the biochemistry or haematology of the organism, and these changes are relevant for human health.

STOT SE recognizes three categories of which the most severe category 1 is described in the CLP Regulation as:

“Substances that have produced significant toxicity in humans or that, on the basis of evidence from studies in experimental animals, can be presumed to have the potential to produce significant toxicity in humans following single exposure. Substances are classified in Category 1 for specific target organ toxicity (single exposure) on the basis of:
(a) reliable and good quality evidence from human cases or epidemiological studies; or
(b) observations from appropriate studies in experimental animals in which significant and/or severe toxic effects of relevance to human health were produced at generally low exposure concentrations. Guidance dose/concentration values are provided below (see 3.8.2.1.9) to be used as part of weight-of-evidence evaluation”.

The CLP Guidance concentration value for single dose exposures for placing a substance in Category 1 is $C \leq 10$ mg/l/4h for vapours (rat). The experimental value found for GMA in an acute inhalation toxicity study was at 1.563 mg/l/4h (laboured breathing was observed at this concentration of 269 ppm or 1.563 mg/l/4h by Nitschke et al. (1990) and thus this substance must be classified in STOT SE Category 1 (damage to the respiratory tract after inhalation). This

classification is also justified because of the severe multifocal necroses and inflammation of the lung after 4 days of exposure to 0.931 mg/L for 6 hours. Although the exposure was repeated for 4 days and the exposure period per day was somewhat longer, the exposure concentration was clearly below the guidance value of 10 mg/L for category 1. Therefore it is very likely that comparable respiratory tract irritation would have occurred after single exposure. These effects have probably occurred in the available acute inhalation study. However, they were probably not observed due to the limited necropsy and the long post-exposure observation period in an acute study. Local respiratory tract tissue injury is also expected given the corrosiveness seen on skin and eyes.

As STOT SE effects are only observed after inhalation exposure and are due to local irritation, it is not expected that acute oral and dermal exposure will result in comparable effects. Further, for the oral route, GMA is already classified as acute toxic Cat. 4. For the dermal route the substance is already classified as acute toxic cat. 3. An additional classification for these routes with STOT SE would be a double classification.

4.3.3 Conclusions on classification and labelling

Classification with STOT SE in category 1 is required with the respiratory tract as target organ and the inhalation route.

4.4 Irritation

4.4.1 Skin irritation

4.4.1.1 Non-human information

Table 14a: Summary table of relevant skin irritation studies

Method	Results	Remarks	Reference
Rabbit (New Zealand White) OECD Guideline 404 (Acute Dermal Irritation / Corrosion)	Overall irritation score: 5 of max. 6 Corrosiveness occurred in 2 out of 6 animals (Time point: 4 hours)	2 (reliable with restrictions) key study experimental result Test material: GMA	Lockwood (1991) OECD (1999)
Rabbit (albino) Undiluted GMA	Overall irritation score: 4 of max. 6 Single rabbit died after 1 st application (Time point: 4 hours)	4 (not assignable) supporting study experimental result Test material: GMA	Olson (1960) OECD (1999)
Rabbit 10% solution in propylene glycol	Overall irritation score: 2 of max. 6 Single rabbit showed hypermia and edema after 1 st application and moderate	4 (not assignable) supporting study	Olson (1960) OECD (1999)

Method	Results	Remarks	Reference
	burn after 2 nd application (Time point: 4 hours)	experimental result Test material: GMA	
Rabbit (domestic) 0.1 mL (100 mg) no data	Overall irritation score: 4 of max. 6 (Time point: 5 days)	3 (not reliable due to low amount of substance, absence of information on exposure duration and repeated exposure) experimental result Test material: GMA purity 92%	Ou-Yang (1988) OECD (1999)

4.4.1.2 Human information

No relevant information available

4.4.1.3 Summary and discussion of skin irritation

Key studies

Lockwood (1991)

A sample of GMA was tested for evaluation of corrosiveness to the skin by the DOT (Department of Transportation) test. A 4-hour exposure of the test material (0.5 ml on a one by one inch gauze pad covered with plastic) to the clipped backs of 6 New Zealand White rabbits resulted in moderate to severe erythema and slight to moderate edema at 4, 24 or 48 hours. Moderate necrosis was observed in 2 out of 6 rabbits. A 1-hour exposure resulted in slight erythema and edema, and superficial necrosis (2 rabbits) to the skin of 6 rabbits. The scoring system deviated from the Draize score with grades ranging from 1 (no effect) to 6. No information is available after 48 hours.

Table 14b: Summary of the Lockwood (1991) severity scores

GRADE	ERYTHEMA	EDEMA	NECROSIS
1	NONE	NONE	NONE
2	very slight	very slight	very slight
3	SLIGHT PINK	SLIGHT	SUPERFICIAL
4	MODERATE PINK	MODERATE	MODERATE
5	RED	MARKED	DEEP
6	VERY RED	SEVERE	VERY DEEP

Table 14c: Summary of the Lockwood (1991) study with respective severity scores after 1-hour exposure

		Rabbit: 1			2			3			4			5			6		
		14	24	48	14	24	48	14	24	48	14	24	48	14	24	48	14	24	48
ON BACK INTACT	OBSERVATION TIME (HOURS)																		
	ERYTHEMA	1	2	1	1	1	1	2	3	3	1	3	3	1	3	3	1	4	4
	EDEMA	1	1	1	1	1	1	1	3	2	1	2	2	1	1	1	1	1	1
	NECROSIS	1	1	1	1	1	1	1	1	3	1	1	2	1	1	1	1	1	1

Table 14d: Summary of the Lockwood (1991) study with respective severity scores after 4-hour exposure

		Rabbit: 1			2			3			4			5			6		
		4	24	48	4	24	48	4	24	48	4	24	48	4	24	48	4	24	48
ON BACK INTACT	OBSERVATION TIME (HOURS)																		
	ERYTHEMA	3	3	3	3	3	2	4	4	4	5	5	5	4	4	4	4	4	4
	EDEMA	3	1	1	3	3	2	4	3	3	4	4	2	4	3	2	3	3	2
	NECROSIS	1	1	1	1	1	1	1	3	4	1	3	4	1	1	1	1	3	2

Supporting studies

Olson (1960)

A single covered topical application to the skin of an albino rabbit for four hours induced moderate to severe skin irritation including necrosis with slight to moderate edema and mortality (Olson: 1960). A 10% solution produced slight redness and edema after 1 application (for 4 hours) and a moderate burn after 2 applications (Olson: 1960). No additional information was available with regards to this study.

Ou-Yang (1988)

One piece of undamaged skin two by two centimetres on either side of the spine was chosen for the testing, one side for testing and the other for control (an equal amount of tap water). 0.1 ml of original concentration of GMA was applied for five days. The localized skin reaction was observed daily. After the experiments, the skin was removed for microscopic examination. GMA induced high irritation to the skin of rabbits. The 0.1ml applied area showed red, swelled and blistered after one or two days, subdermal bleeding and ulcers after three days, and hard, thicker, cracked, pigmentation after five days. The pathological changes were degeneration and necrosis of surface skin cells, disappearance of cellular boundaries, displaying pink staining material, bleeding in the corium cells and lymph cell infiltration with accompanying formation of abscesses.

4.4.1.4 Comparison with criteria

The CLP Regulation recognises for the skin corrosive category 1 three subcategories 1A, 1B and 1C. Category 1A shows corrosiveness within an exposure of 3 minutes or less (observation after max 1 hour), category 1B shows corrosiveness after 3 minutes but within 1 hour or less (observation after max 14 days), and category 1C shows corrosiveness after 1 hour but within 4 hours or less (observation after max 14 days).

Corrosive adverse effects for GMA were observed in 2 out of 6 rabbits after 4 hours exposure but not after 1 hour and thus this substance must be classified as 1C (Lockwood: 1991).

4.4.1.5 Conclusions on classification and labelling

According to the available studies GMA must be classified according to CLP as: Skin corrosive Category 1C (H314).

4.4.2 Eye irritation

4.4.2.1 Non-human information

Table 15a: Summary table of relevant eye irritation studies

Method	Results	Remarks	Reference
Rabbit (albino) Direct instillation Undiluted GMA directly instilled in eye; 10% GMA in propylene glycol	Overall irritation score: 4 of max. 6 (Time point: 7 days post-exposure) Undiluted/unwashed eye: slight to moderate conjunctivitis, slight corneal injury cleared in one week. Undiluted/washed eye: slight conjunctivitis, cleared in 48 hrs. 10% GMA in propylene glycol/unwashed eye: slight conjunctivitis, cleared in one hour. 10% GMA in propylene glycol/washed eye: slight conjunctivitis, cleared in 24 hrs.	4 (not assignable) supporting study experimental result Test material: GMA	Olson (1960) Smyth (1969) OECD (1999)
Rat Inhalation single 4-hour Concentration: 610, 1563 and 2394 mg/m ³	Overall irritation score: 4 of max. 6 (Time point: 4 hour treatment, 14-day post-exposure) Eye irritation, corneal opacity Changes did not heal within 14 days	2 (reliable with restrictions) supporting study experimental result Test material: GMA purity 99.8%	Nitschke (1990)
Rat Inhalation Concentration: 58.2, 223	Overall irritation score: 4 of max. 6 (Time point: 2-weeks treatment, no post-exposure time evaluation) Eye irritation, corneal clouding	4 (not assignable) Supporting study experimental result Test material:	Landry (1991)

Method	Results	Remarks	Reference
and 931 mg/m ³ 6 hours/day, 5 days/week for 2 weeks		GMA purity 99.61%	

Supporting studies

Olson (1960)

GMA was applied to both eyes of the rabbit and within 30 seconds, one eye was washed with tap water for two minutes. GMA was allowed to remain in the other eye. Results showed that with undiluted GMA, slight to moderate conjunctivitis was observed in the unwashed eye where slight corneal injury cleared in one week. In the washed eye, slight conjunctivitis was observed which cleared in one hour. In a solution of 10% GMA in propylene glycol, slight conjunctivitis was observed in the unwashed eye which cleared after 48 hours. In the washed eye, slight conjunctivitis was observed which cleared within 24 hours.

In the OECS SIDS (1999) and the registration file, it is reported that direct instillation of undiluted GMA to the eye of albino rabbits induced severe irritation and corneal damage. The corneal damage did not heal within 7-days post-dosing, which is contradictory to what study report findings (Table 15b). This ocular damage was prevented by washing with water within 30 seconds (Olson: 1960, Smyth: 1969).

Table 15b: Summary of Olson (1960) study

EYE CONTACT IRRITATION IN RABBITS

Material	Conc.	Eye Contact Irritation	
		Unwashed eye	Washed eye ②
Glycidyl methacrylate	Undiluted	Sl. to mod. conjunctivitis; sl. corneal injury cleared in one week	Sl. conjunctivitis cleaned in 48 hours.
Glycidyl methacrylate	10% in propylene glycol	Sl. conjunctivitis cleared in one hour	Sl. conjunctivitis cleared in 24 hours.
<u>Toxicity Test</u>		<u>Glycidyl Acrylate</u>	
Acute Oral		LD ₅₀ approximately 0.06 g/kg in rats.	LD ₅₀ approximately 0.7 g/kg in rats.
Eye Irritation		Undiluted and 10% in propylene glycol likely to produce permanent impairment of vision. Reacts rapidly with eye tissue. Chemical workers goggles recommended.	Undiluted: Moderately irritating. Side shield glasses recommended. 10% in propylene glycol: Slightly irritating, no corneal damage. Minimal eye protection satisfactory.

In inhalation study using rats, eye irritation was also induced. Acute exposure for 4 hours induced eye irritation at 1563 mg/m³ and 2394 mg/m³. Slight corneal opacity was also observed at 610 mg/m³, and moderately at 1563 mg/m³ and 2394 mg/m³. These changes did not heal within 14 days post-exposure (Nitschke et al.: 1990). In a sub-acute study, rats were exposed at 58.2, 223 and 931 mg/m³, 6 hours/day, 5 days/week for 2 weeks. As a result, eye irritation and corneal clouding were observed at 931 mg/m³ (Landry et al., 1991).

4.4.2.2 Human information

No relevant information available

4.4.2.3 Summary and discussion of eye irritation

The only eye irritation study has some resemblance to the OECD eye irritation study but is limitedly reported and there are differences in the described effects between the available report from Olson (1960) and the summary in the registration file and in the OECD summary. Therefore, there are no reliable results from relevant studies although the results indicate that GMA has potential to induce eye irritation. The supporting studies are performed using inhalation exposure and result in clear irritating effects on the eye. However, this is due to exposure of the vapour phase and not the liquid as required for classification for eye irritation.

4.4.2.4 Comparison with criteria

According to chapter 3.2.3 of CLP skin corrosive substances shall be considered as leading to serious eye damage. Seen the skin corrosivity of GMA as described in chapter 4.3, GMA must be considered as Eye damage category 1.

4.4.2.5 Conclusions on classification and labelling

According to the available studies GMA must be classified according to CLP as Eye damage Category 1 (H318).

4.4.3 Respiratory tract irritation

4.4.3.1 Non-human information

See section 4.4.3.3 below

4.4.3.2 Human information

No relevant information available

4.4.3.3 Summary and discussion of respiratory tract irritation

Not relevant for CLP as this effect is covered under STOT SE (chapter 4.3).

4.5 Corrosivity

Table 16: Summary table of relevant corrosivity studies

Method	Results	Remarks	Reference
Deliberately left empty			

4.5.1 Non-human information

In experimental data (see chapter 4.4.1, 4.4.2 and 4.4.3) the substance appeared corrosive to skin.

4.5.2 Human information

No relevant information available

4.5.3 Summary and discussion of corrosivity

The substance appears to be corrosive to skin.

4.5.4 Comparison with criteria

See section 4.4.1.

4.5.5 Conclusions on classification and labelling

See section 4.4.1.

4.6 Sensitisation

4.6.1 Skin sensitisation

4.6.1.1 Non-human information

Table 17a: Summary table of relevant skin sensitisation studies

Method	Results	Remarks	Reference
Guinea pig (Hartley) male Buehler test Delayed allergy reaction test Induction dose 25% reduced to 10%	No. with positive reactions: 7 out of 10 (test group); dose: 1% 1st reading: 8 out of 10 (positive control); dose: 5% Observations: slight erythema	2 (reliable with restrictions) key study experimental result Test material: GMA	Dow (1990) OECD (1999)
Guinea pig	No. with positive reactions: 1st reading: 6 out of 6 (test group);	4 (not assignable) supporting study	BIBRA (1988) OECD (1999)

Method	Results	Remarks	Reference
	dose: unknown Observations: skin sensitisation (eg contact dermatitis)	experimental result Test material: GMA	
Guinea pig delayed allergy reaction test delayed allergy reaction test	No. with positive reactions: 1st reading: 7 out of 10 (test group); induction dose: 0.1 ml of 1% GMA in acetone, skin smear application 1st reading: 6 out of 10 (test group); induction dose: 0.1 ml of 1% GMA in acetone, intradermal injection Observations: Hyperemia, edema, scleroma and necrosis were observed on the treated area and these changes reached a peak on the fourth day. But no obvious change was observed on the control area.	4 (not assignable) experimental result Test material: GMA purity 92%	Ou-Yang (1988) OECD (1999)
Guinea pig rapid allergic reactive test with active stimulation rapid allergic reaction test (active stimulation)	No. with positive reactions: 1st reading: 5 out of 5 (test group); dose: i.v. 0.5% GMA solution in homologous serum albumin 1st reading: 0 out of 3 (negative control); dose: zero Observations: Breathing difficulties, wheezing, increased mouth and nose secretions, spasms and death were observed in the test group, but no obvious changes in the control group.	4 (not assignable) experimental result Test material: GMA purity 92%	Ou-Yang (1988) OECD (1999)
Guinea pig rapid allergic reaction test with passive stimulation rapid allergic reaction test (passive stimulation) (injection of serum of allergic guinea pigs)	No. with positive reactions: 1st reading: 5 out of 5 (test group); dose: see details on study design Observations: Blue circles or spots were observed most markedly in the one to three areas, followed by the one to ten area and in the one to thirty areas, and there were a few scattered blue spots. This showed that the reaction is related to the dosage.	4 (not assignable) experimental result Test material: GMA purity 92%	Ou-Yang (1988) OECD (1999)

Guinea pigs received three topical applications with 0.4 ml of 10 % (third application) or 25 % (first and second application) GMA in dipropylene glycol monomethyl ether during the three-week induction phase. The single challenge application with 1% GMA induced slight erythema in these animals (7/10). (The Dow Chemical Company: 1990) This study deviates from the OECD TG 406 as no control animals were included which were only exposed to the solvent during induction and to the same concentration of the substance during the challenge.

Regarding the Bibra study (1988), no additional information is available than that it induced a positive reaction in first reading in 6 out of 6 guinea pigs.

Ou-Yang et al. (1988) reported on delayed and rapid allergy reaction tests in guinea pigs. In delayed allergy reaction test, induction with localized smear applications or intradermal injection with 0.1 ml of 1 % GMA in acetone for 10 days and challenge with an unknown concentration induced hyperaemia, oedema, scleroma and necrosis. Those changes belong to the strong allergenic category. As for rapid allergic reaction test, two tests by active and passive stimulation were conducted. In the active stimulation, 0.5 % GMA with homologous serum albumin was injected intradermally and the challenge was conducted intravenously. Breathing difficulties, wheezing, increased mouth and nose secretions, spasms and death were observed, belonging to the strong allergic category. In the passive stimulation, firstly, the diluted serum given from the sensitized guinea pig was injected subcutaneously to other animals and one hour later, 0.5 ml of 0.1 % GMA with homologous serum albumin was injected intravenously to the same animals. Blue circles or spots observed belonged to the strong allergic category. Both the delayed and rapid allergy test results showed that GMA was a moderate sensitiser. The author reported that this might be the reason that the epoxy radical of GMA easily combined with protein.

4.6.1.2 Human information

Table 17b: Summary of human patch tests

Method	Results	Remarks	Reference
Human Vehicle: petrolatum human patch test	No. with positive reactions: 1st reading: 3 out of 3 (test group); dose: 1% Observations: erythema, oedema and vesiculation	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Dempsey (1982) OECD (1999)
human female human patch test	No. with positive reactions: 1st reading: 1 out of 1 (test group); dose: 0.01% and 0.05% Observations: A patch test conducted on a 31-year-old non-atopic woman showed a reaction to GMA (0.01 and 0.05 % acetone)	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Matura (1995) OECD (1999)

There were two studies on human patch tests.

Three cases of allergic contact hypersensitivity to GMA used in adhesive sealant manufacturing were reported. Both closed and open patch testing with 1 % GMA solution in petrolatum was positive in all 3 cases. Symptoms included erythema, oedema, and vesiculation and

a strong 2⁺ reaction as scored according to the International Contact Dermatitis Research Group classification. (Dempsey: 1982).

Patch test was conducted for a 31-year-old non-atopic woman, who had worked as a chemist and mixed emulsions used to impregnate paper and textile materials to make them oil and water resistant. In this work, she had been in contact with acrylate derivatives (GMA, ethoxyethyl acrylate, etc.). In relation to this work, she had a history of recurrent acute vesiculopapular hand dermatitis with severe itching and burning mainly on the fingertips, palmar and dorsal aspects of the fingers, and both palms. As a result of patch test, she reacted only to nickel, GMA (0.01 and 0.05 % acet.) and ethoxyethyl acrylate among the European standard series and (meth) acrylate series. This reaction to nickel was relevant to her jewelry intolerance. (Matura et al.: 1995).

4.6.1.3 Summary and discussion of skin sensitisation

The key study (Dow, 1990) showed erythema in 7 out of 10 guinea pigs dermally induced with 25% GMA (reduced to 10% for the third induction) and dermally challenged with 1% GMA. The study resembles the Buehler study; however, a negative control group was missing. The induction dose of 10% induced some local effects. However, seen the strong reduction in concentration of the challenge dose it is expected that the observed effects are sensitisation and not irritation. The key study is supported by some other test with limited study information or using a different, non-standard, approach. Although the predictive value of these types of studies is not known, the results were considered positive. There were also a limited number of human cases.

4.6.1.4 Comparison with criteria

The CLP Regulation states that substances shall be classified as skin sensitisers (Category 1) where data are not sufficient for sub-categorisation in accordance with the following criteria:

- “(i) if there is evidence in humans that the substance can lead to sensitisation by skin contact in a substantial number of persons, or
- (ii) if there are positive results from an appropriate animal test”

For a non-adjuvant Guinea pig test, the criteria for a positive result is the induction of a response 15% of the animals or more. Although, the key study was not in line with the OECD requirements for a Buehler study, the positive response in 70% of the guinea pigs supported by positive results in other studies and the presence of a small number of human cases warrant classification for skin sensitisation. However, due to the changes during the study of the induction concentration in the key study from 25% to 10% it is not possible to conclude whether also the criteria for sub-category 1A for a Buehler are fulfilled because these require a response in more than 60% of the guinea pigs (fulfilled) after topical induction between 0.2 and 20% (unclear whether fulfilled). Therefore, the available data do not allow sub-categorisation and category 1 is proposed.

4.6.1.5 Conclusions on classification and labelling

According to the available studies GMA must be classified according to CLP for Skin sensitisation Category 1 (H317) in view of the results found both in humans and animals.

4.6.2 Respiratory sensitisation

Table 18: Summary table of relevant respiratory sensitisation studies

Method	Results	Remarks	Reference
Deliberately left empty			

4.6.2.1 Non-human information

In none of the acute and sub-acute inhalation studies respiratory sensitisation was observed.

4.6.2.2 Human information

No relevant information available. However, there are no reports found in public literature.

4.6.2.3 Summary and discussion of respiratory sensitisation

In none of the acute and sub-acute inhalation studies respiratory sensitisation was observed. However, these studies are not developed to determine the respiratory sensitising potential of substances.

4.6.2.4 Comparison with criteria

Not relevant.

4.6.2.5 Conclusions on classification and labelling

No classification for respiratory sensitisation is required based on absence of data.

4.7 Repeated dose toxicity

4.7.1 Non-human information

4.7.1.1 Repeated dose toxicity: oral

Table 19a: Summary table of relevant repeated dose toxicity studies (oral)

Method	Results	Remarks	Reference
Rat (Crj: CD(SD)) 12 male/12 female combined repeated dose and reproduction / developmental screening (oral: gavage) 10, 30, 100 mg/kg bw/day (nominal)	NOAEL: 10 mg/kg bw/day (nominal) (male) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach) NOAEL: 30 mg/kg bw/day (nominal) (female) based on: test mat. (local adverse effect: squamous hyperplasia in	1 (reliable without restriction) key study experimental result Test material: GMA purity 99.93%	Ministry of Health and Welfare, Japan (1997) OECD (1999)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
<p>Vehicle: corn oil</p> <p>Exposure:</p> <p>Duration of test: Males: 46 days</p> <p>Females: 40-47 days</p> <p>Post exposure observation period: 1 day</p> <p>Duration of exposure:</p> <p>Males: 45 days</p> <p>Females: from 14 days before mating to day 3 of lactation (daily)</p> <p>OECD Guideline 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)</p>	<p>forestomach)</p> <p>NOAEL: 30 mg/kg bw/day (nominal) (male/female) based on: test mat. (systemic adverse effect: increase in absolute and relative kidney and adrenal weights; increase in total protein and albumin)</p> <p>LOAEL: 30 mg/kg bw/day (nominal) (male) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach)</p> <p>LOAEL: 100 mg/kg bw/day (nominal) (female) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach)</p> <p>LOAEL: 100 mg/kg bw/day (nominal) (male/female) based on: test mat. (systemic adverse effect: increase in absolute and relative kidney and adrenal weights; increase in total protein and albumin)</p>		
<p>Rabbit</p> <p>Sub-acute (oral: unspecified)</p> <p>50 mg/kg/day</p> <p>Exposure: 15 days (daily)</p>	<p>no NOAEL identified: 50 mg/kg bw/day (nominal) (Fifteen days after treatment, slow reactions and some head shaking was observed in the treated group. In addition, 2 of 10 animals succumbed to prostration and died. There were decrease in body weight, increase in relative weights of heart, liver and kidneys, and some haematological changes (increase in white blood cells, platelets and lymph cells, decrease in leukoplasts and intermediate cells). In pathological examination, there was heart bleeding, deterioration of the heart muscle fibres, widespread fatty changes in the liver with focal necrosis. In kidneys, extravasated blood and haemorrhaging were observed, and the upper part of the renal tubules was swollen. There was also red stained protein in the tubular</p>	<p>3 not (reliable) supporting study experimental result</p> <p>Test material: GMA purity 92%</p>	<p>Ou-Yang (1988)</p> <p>OECD (1999)</p>

Method	Results	Remarks	Reference
	cavity, cranial haemorrhaging, small focal necrosis of the grey matter, and ulceration and necrosis of the mucous membrane of the stomach. However, incidence of these changes was not reported.		
Rat male/female chronic (oral: gavage) 0.1 and 0.3 mg/kg/day Exposure: 1 year (5 days/week)	NOAEL: > 0.3 mg/kg bw/day (nominal) (male/female) (No effects in all tissues were found, which could clearly be related to treatment. However, there was no more information provided)	4 (not assignable); experimental result Test material: GMA	Hadidian (1968) OECD (1999)

Key studies

Oral toxicity study of GMA was performed in SD (Crj: CD) rats (n=12) by an OECD combined repeated dose and reproductive/developmental toxicity screening test (OECD TG 422). Administration was conducted at doses of 10, 30 and 100 mg/kg/day by gavage for 45 days in males and from 14 days before mating to day 3 of lactation in females (Ministry of Health and Welfare, Japan: 1997).

Salivation was observed at 30 mg/kg (5/12) and 100 mg/kg (12/12) in males. In males, there was an increase in absolute and relative kidney and adrenal weights at 100 mg/kg. In blood chemistry of males, increase in total protein and albumin was observed. These changes were not considered as adverse effects. In histological examination, squamous hyperplasia in forestomach was observed at 30 and 100 mg/kg in males and cellular infiltration in forestomach at 100 mg/kg in females (see Table 19b). These histological changes were considered to be due to the irritation of GMA. NOAEL (including local effects) for oral repeat toxicity was considered to be 10 mg/kg/day for males and 30 mg/kg/day for females. The NOAEL for repeated dose toxicity (systemic effects) was considered to be 30 mg/kg/day (both sexes).

Table 19b: GMA-induced histological changes in forestomach in rats as reported by the Ministry of Health and Welfare, Japan (1997, pg. 46)

	Male				Female			
	0	10	30	100 mg/kg/day	0	10	30	100
DIGESTIVE SYSTEM								
forestomach	(12)	(11)	(12)	(11)	(12)	(12)	(12)	(12)
edema	4	7	12**	7	0	0	0	0
microgranuloma	0	0	1	0	0	0	0	0
cellular infiltration	6	2	8	9	0	2	3	4*
cellular infiltration, lymphocyte	0	0	0	1	0	0	0	0
hyperkeratosis	0	0	1	0	0	0	0	1
squamous hyperplasia	1	1	11**	11**	3	2	1	5

Two other orally repeated toxicity studies were reported.

One-year study is very limited (Hadidian et al., 1968). Rats (3 males and 3 female) were dosed 5 days/week by gavage at 0.1 mg/kg. Groups of 15 male and 15 female rats were also dosed at 0.3 mg/kg. The authors concluded that no tissue effects related to the treatment were found. These dosages are considered to be too low.

In another study, five male and female rabbits were given orally at 50 mg/kg daily for 15 days (Ou-Yang et al.: 1988). Some animals showed slow reactions, head shaking and prostration, and two animals died. There were several haematological and pathological changes including bleeding, necrosis and so on in heart, liver, kidneys and stomach.

These studies cannot be accepted for hazard assessment because of unreliability such as no Test Guideline, no GLP, unlikely severe systemic toxicity compared to the key study, and insufficient information on protocol and data analysis including purity of substance and pathological data.

4.7.1.2 Repeated dose toxicity: inhalation

Table 19c: Summary table of relevant repeated dose toxicity studies (inhalation)

Method	Results	Remarks	Reference
Rat (Fischer 344) male/female Sub-chronic (inhalation: vapour) 0.5, 2 and 15 ppm (2.9, 12 and 87 mg/m ³ , calculated daily dose 0.35, 1.46 and 10.59 mg/kg/day) (nominal conc.) Exposure: 13 weeks (6 hours per day, 5 days per week)	NOAEC: 12 mg/m ³ air (nominal) (male/female) (Hyperplasia of respiratory epithelium of the nasal tissues, graded as very slight, in all animals.) LOAEC: 87 mg/m ³ air (nominal) (male/female) (Hyperplasia of respiratory epithelium of the nasal tissues, graded as very slight, in all animals).	2 (reliable with restrictions) key study experimental result Test material: GMA purity 99.5%	Landry (1996) OECD (1999)
Rat (Fischer 344) male/female sub-acute (inhalation: vapour) 0, 10, 40 and 160 ppm (0, 58.2, 233, and 931 mg/m ³ , calculated daily dose 0, 7.09, 28.4 and 113 mg/kg/day) (nominal conc.) Exposure: 2 weeks (6 hours per day, 5 days per week)	LOAEC: 58.2 mg/m ³ air (nominal) (male/female) (Very slight effects in the nasal cavity in 5 of the 10 rats exposed).	2 (reliable with restrictions) key study experimental result Test material: GMA purity 99.61%	Landry (1991) OECD (1999)
Rat Sub-acute (inhalation: vapour) 35 ppm (204 mg/m ³ , calculated daily dose 24.9 mg/kg/day) (nominal conc.) Exposure: 2 weeks (6 hours/day, 5 days/week)	LOAEC: 204 mg/m ³ air (Decrease in body weight gain, respiratory symptoms, and higher red blood cell count than that of control were observed. There were no histopathologic effects. No remaining exposure-related effects were observed at two weeks after exposure.)	2 (reliable with restrictions) supporting study experimental result Test material: GMA	DuPont (1977) OECD (1999)
Rat chronic (inhalation: vapour) 15.3 and 206 mg/m ³ (calculated daily dose: 2.24 and 30.1 mg/kg/day)	LOAEL: 15.3 mg/m ³ air (nominal) (A wide range of chronic toxic effects, such as changes of liver and spleen weight, and enzyme (transaminase) levels in blood or tissue, and lesion in central	2 (reliable with restrictions) supporting study experimental result	Ouyang Guoshun (1990) OECD (1999)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
(nominal conc.) Exposure: 26 weeks (6 hours/day, 6 days/week)	nervous system, cardiovascular system, liver and kidney, were observed. At 206 mg/m ³ , all the changes were more pronounced and the pathological lesions only got worsened after the exposure was ceased. On the other hands, the changes at 15.3 mg/m ³ were sparse and slight, and almost all vanished one month after the exposure was ended. Because of the higher vapour pressure and lower purity, the author suggested that the test material used in this study contained components other than GMA, which may have contributed to the toxicity observed.)	Test material: GMA purity 92%	
Rabbit (New Zealand White) female Sub-acute (inhalation: vapour) 2, 5 and 10 ppm (11.6, 29.1, 58.2 mg/m ³ , calculated daily dose 1.04, 2.62, 5.24 mg/kg/day) (nominal conc.) Exposure: 13 days (6 hours/day, daily)	LOAEC: 11.6 mg/m ³ air (nominal) (Olfactory epithelial degeneration)	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Cieszlak (1996) OECD (1999)
Rabbit chronic (inhalation: vapour) 15.3 and 206 mg/m ³ (calculated daily dose 1.18 and 15.9 mg/kg/day) Exposure: 26 weeks (6 hours/day, 6 days/week)	LOAEC: 15.3 mg/m ³ air (nominal) (A wide range of chronic toxic effects, such as lesion in central nervous system, cardiovascular system, liver and kidney, and other degenerative changes in brain and coverings, were observed. Moreover, there were changes of cardiac EKG (not diagnostic of specified effects) and erythrocyte count. At 206 mg/m ³ , all the changes were more pronounced and the pathological lesions only got worsened after the exposure of GMA was ceased. On the other hands, the changes at 15.3 mg/m ³ were sparse and slight, and almost all vanished one month after the exposure was ended.	2 (reliable with restrictions) supporting study experimental result Test material: GMA purity 92%	Ouyang Guoshun (1990) OECD (1999)

Method	Results	Remarks	Reference
	Because of the higher vapour pressure and lower purity, the author suggested that the test material used in this study contained components other than GMA, which may have contributed to the toxicity observed.)		

Key study: Landry (1996)

Sub-chronic inhalation toxicity study was conducted in rats at concentrations of 2.9, 12 or 87 mg/m³ for 13 weeks (6 hours/day, 5 days/week) (Landry et al.: 1996). These three doses were calculated as approximately 0.35, 1.46 or 10.6 mg/kg/day, respectively. There were no treatment related in-life observations, and no significant treatment-related effects on body weight, urinalysis, clinical chemistry or haematology parameters, as well as gross pathologic changes or organ weights at any exposure level. Treatment-related effects were limited to hyperplasia of respiratory epithelium of the nasal tissues in all animals at 87 mg/m³. In all affected animals, the hyperplastic respiratory epithelium was approximately two to three times as thick as in control animals, and was located in the anterior portions of the nasal passages, involving the tips of the turbinates and the lateral walls of the nasal passages. These changes were considered to have resulted from respiratory irritation. Therefore, NOAEC was considered 12 mg/m³ (1.46 mg/kg/day) for both sexes.

Supportive studies

Sub-acute inhalation toxicity studies were performed in rats and rabbits. Rats were exposed to GMA at concentrations of 58.2, 233 or 931 mg/m³ for 2 weeks (6 hours/day, 5 days/week) (Landry et al.: 1991). These three concentrations were calculated as 7.09, 28.4 or 113 mg/kg/day. Decrease in body weight was observed at 233 and 931 mg/m³. At 931 mg/m³, general debilitation with noisy and difficult respiration (mouth breathing), eye irritation, corneal clouding and distended abdomen (day 4) were observed. The animals at 931 mg/m³ were terminated early on day 4 because of the severity of the respiratory and ocular effects. Microscopically, there was severe multifocal necrosis and inflammation of the olfactory epithelium in the nasal cavity. At 233 mg/m³, there were slight to moderate multifocal necrosis, and inflammation of the respiratory and olfactory nasal epithelium. At 58.2 mg/m³, microscopically there was very slight multifocal necrosis of individual respiratory epithelial cells in 3 of 5 males and in 2 of 5 females. These changes in respiratory tract were considered due to irritation of GMA. There were no histopathological changes in any other tissues. Therefore, 58.2 mg/m³ (7.09 mg/kg/day) was considered to be LOAEC because of tissue damages in respiratory tract.

Rabbits were exposed at 2.91, 11.6, 29.1, 58.2 mg/m³, 6 or 7 hours/day, daily for 13 consecutive days. (Cieszlak et al., 1996, Vedula et al., 1996. Two separate studies). Treatment-related degeneration of the nasal olfactory epithelium was observed at 11.6 mg/m³. At 29.1 and 58.2 mg/m³, there were olfactory epithelial degeneration, and the hyperplasia, erosions, ulcers and inflammation of the nasal epithelium. After 4-week recovery period, there was complete reversibility of these changes except for olfactory epithelial degeneration observed at 29.1 and 58.2 mg/m³, which showed only partial reversibility. At 11.6 mg/m³, nasal tissue was indistinguishable from controls at one month post-exposure. 2.91 mg/m³ (0.26 mg/kg/day) was considered to be NOAEC.

In a 26-week inhalation toxicity study at concentrations of 15.3 and 206 mg/m³ in rats and rabbits (Ouyang Guoshun et al.: 1990), a wide range of toxic effects, such as lesion in central nervous system, cardiovascular system, liver and kidney, were observed in both species at the low and high doses. However, because of the higher vapour pressure and lower purity, the author suggested that the test material used in this study contained components other than GMA, which may have contributed to the toxicity observed. Therefore these systemic toxicities observed in the studies are questionable.

4.7.1.3 Repeated dose toxicity: dermal

No relevant information available on GMA.

4.7.1.4 Repeated dose toxicity: other routes

No relevant information available.

4.7.1.5 Human information

No relevant information available

4.7.1.6 Other relevant information

Several oral and inhalation studies were available for glycidol. No dermal studies for glycidol were available.

Glycidol

Table 19d: Oral repeated dose toxicity studies with glycidol copied from the C&L proposal of glycidol (ECBI-92/95-add.3).

Species/strain	Dose (mg/kg bw)	Duration of treatment	Observations and remarks (specify group size, NOAEL, effects of major toxicological significance)
Rat (F344/N) 5 males and 5 females per dose group	0, 37.5, 75, 150, 300, 600	14 days	All rats receiving 600mg/kg died before end of study period. No other deaths occurred. Final mean body weights of males dosed at 150 or 300mg/kg were respectively 10% and 21% lower than that of vehicle controls. Final mean body weights of dosed and vehicle control female rats were similar. Oedema and degeneration of epididymal stroma was observed in 4/5 males given 300mg/kg. Atrophy of the testis and granulomatous inflammation of the epididymis were seen in the fifth animal (NTP, 1990).
Mouse (B6C3F ₁) 5 males and 5 females per dose group	0, 37.5, 75, 150, 300, 600	14 days	All mice receiving 600mg/kg died within 4 days. 3/5 males and 2/5 females given 300mg/kg died before end of study. Other deaths occurring in lower dose groups considered to be due to mis-dosing. Final mean body weights of dosed males were similar to the controls. Final female mean body weights in groups dosed at 150 or 300mg/kg were respectively, 7% and 8% lower than that of controls. Females receiving 600mg/kg and males and females receiving 150mg/kg had diarrhoea. Inactivity and ruffled hair coats were observed for 2/5 males and 2/5 females dosed at 600mg/kg and 3/5 males and 2/5 females dosed at 300mg/kg. Focal demyelination in the medulla and thalamus of the brain was present in all female mice given 300mg/kg (NTP, 1990).

Table 19d: Oral repeated dose toxicity studies with glycidol copied from the C&L proposal of glycidol (ECBI-92/95-add.3).

Species/ strain	Dose (mg/kg bw)	Duration of treatment	Observations and remarks (specify group size, NOAEL, effects of major toxicological significance)
Rat (F344/N) 10 males and 10 females per dose group	0, 25, 50, 100, 200, 400	5 days per week, for 13 weeks	All rats dosed at 400mg/kg died by week 2. 3/10 males and 1/10 females given 200mg/kg died before end of study. Final mean body weights of rats in 50, 100 or 200mg/kg groups were respectively, 9%, 4% and 15% lower than that of the vehicle controls for males, and 6%, 7% and 11% lower for females. The frequencies of non-neoplastic lesions in 0, 100, 200 and 400mg/kg groups were respectively as follows: Cerebellar necrosis (males: 0/10, 0/10, 2/10, 10/10; females: 0/10, 0/10, 4/10, 9/10), brain demyelination (males: 0/10, 0/10, 5/10, 6/10; females: 0/10, not reported, 0/10, 6/10), renal tubular cell degeneration/necrosis (males: 0/6, not reported, 0/8, 6/10; females: 0/6, not reported, 0/6, 10/10), thymic lymphoid necrosis (males: 0/6, not reported, 0/8, 2/9; females: 0/6, not reported, 1/7, 9/10), testicular atrophy (2/10, 3/9, 10/10, 9/10) (NTP, 1990).
Mouse (B6C3F ₁) 10 males and 10 females per dose group	0, 19, 38, 75, 150, 300	5 days per week, for 13 weeks	Due to an error, mice were given 125% of the nominal doses during week 2. All mice given 300mg/kg died by week 2. 4/10 males and 3/10 females given 150mg/kg died before end of study. Final mean body weights of treated animals were 6-10% lower than vehicle controls except for males dosed at 38mg/kg when the value was 5% greater than that for the control group. The frequencies of non-neoplastic lesions observed at 0, 75, 150 and 300mg/kg were respectively as follows: Brain demyelination (males: 0/10, 0/10, 5/10, 1/10; females: 0/10, 0/10, 3/10, 6/10), renal tubular cell degeneration (males: 0/10, not reported, 0/10, 4/10; females: 0/10, not reported, 0/10, 0/10), testicular atrophy/degeneration (2/10, 3/9, 5/10, 1/10) (NTP, 1990). Data do not support classification with R48 (see Annex 1)

Glycidol- Inhalation studies

Table 19e: Inhalation repeated dose toxicity studies with glycidol copied from the C&L proposal of glycidol (ECBI-92/95-add.3).

Species	conc. mg/l	Exposure time	Duration of treatment	Observations and remarks (specify group size, NOAEL, effects of major toxicological significance)
Rat (Long- Evans) (10 males)	0, 1.2	7 hours	Daily for 50 days	One rat died of bronchopneumonia between days 49 and 50, but there were no other deaths. Very slight irritation of the eyes, with slight lacrimation and encrustation of the eyelids and slight respiratory distress were observed following the first few exposures to glycidol. These signs of toxicity did not increase in severity with subsequent exposures. Necropsy at the end of dosing revealed no significant gross or microscopic lesions (Hine <i>et al</i> , 1956).

Table 19f: Comparison between GMA and glycidol repeated dose-toxicity studies

GMA	Glycidol
<p>Rat: Males: 46 days; Females: 40-47 days Ministry of Health and Welfare, Japan (1997)</p> <p>NOAEL: 10 mg/kg bw/day (nominal) (male) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach)</p> <p>NOAEL: 30 mg/kg bw/day (nominal) (female) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach)</p> <p>NOAEL: 30 mg/kg bw/day (nominal) (male/female) based on: test mat. (systemic adverse effect: increase in absolute and relative kidney and adrenal weights; increase in total protein and albumin)</p> <p>LOAEL: 30 mg/kg bw/day (nominal) (male) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach)</p> <p>LOAEL: 100 mg/kg bw/day (nominal) (female) based on: test mat. (local adverse effect: squamous hyperplasia in forestomach)</p> <p>LOAEL: 100 mg/kg bw/day (nominal) (male/female) based on: test mat. (systemic adverse effect: increase in absolute and relative kidney and adrenal weights; increase in total protein and albumin).</p> <p>The fertility index decreased significantly at 100 mg.kg group due to low sperm motility.</p>	<p>Rat oral 91-day study</p> <p>All rats that received 400 mg/kg died by week 2; three males and one female that received 200 mg/kg died during weeks 11-12.</p> <p>Sperm count and sperm motility were reduced in male rats that received 100 or 200 mg/kg. Necrosis of the cerebellum, demyelination in the medulla of the brain, tubular degeneration and/or necrosis of the kidney, lymphoid necrosis of the thymus, and testicular atrophy and/or degeneration occurred in rats that received 400 mg/kg.</p> <p>Mouse oral 91-day study</p> <p>All mice that received 300 mg/kg died by week 2; deaths of mice that received 150 mg/kg occurred during weeks 4-8 for males and weeks 1-5 for females.</p> <p>Sperm count and sperm motility were reduced in dosed male mice. Compound-related histopathologic lesions included demyelination of the brain in males and females that received 150 or 300 mg/kg, testicular atrophy in males at all doses, and renal tubular cell degeneration in male mice that received 300 mg/kg.</p> <p>Based on reduced survival, reduced weight gain, and histopathologic lesions in the brain and kidney in rats that received 200 or 400 mg/kg and on reduced survival and histopathologic lesions of the brain in mice that received 150 or 300 mg/kg, doses selected for the 2-year studies of glycidol were 37.5 and 75 mg/kg for rats and 25 and 50 mg/kg for mice.</p>
<p>Rat 90-day Inhalation</p> <p>Landry (1996)</p> <p>NOAEC: 12 mg/m³ air (nominal) (male/female) (Hyperplasia of respiratory epithelium of the nasal tissues, graded as very slight, in all animals.)</p> <p>LOAEC: 87 mg/m³ air (nominal) (male/female) (Hyperplasia of respiratory epithelium of the nasal tissues, graded as very slight, in all animals).</p> <p>Rat 14-day inhalation study</p> <p>DuPont (1977)</p> <p>LOAEC: 204 mg/m³ air (Decrease in body weight gain, respiratory symptoms, and higher red blood cell count than that of control were observed. There were no histopathologic effects. No remaining exposure-related effects were observed at two weeks after</p>	<p>Rat 50-day Inhalation study (1.2 mg/L; 1200 mg/m³)</p> <p>One rat died of bronchopneumonia between days 49 and 50, but there were no other deaths.</p> <p>Very slight irritation of the eyes, with slight lacrimation and encrustation of the eyelids and slight respiratory distress were observed following the first few exposures to glycidol. These signs of toxicity did not increase in severity with subsequent exposures.</p> <p>Necropsy at the end of dosing revealed no significant gross or microscopic lesions.</p>

<p>exposure).</p> <p>Rat 182-day inhalation study</p> <p>Ouyang Guoshun (1990)</p> <p>LOAEL: 15.3 mg/m³ air (nominal) (A wide range of chronic toxic effects, such as changes of liver and spleen weight, and enzyme (transaminase) levels in blood or tissue, and lesion in central nervous system, cardiovascular system, liver and kidney, were observed. At 206 mg/m³, all the changes were more pronounced and the pathological lesions only got worsened after the exposure was ceased. On the other hands, the changes at 15.3 mg/m³ were sparse and slight, and almost all vanished one month after the exposure was ended.</p> <p>Rabbit 13-day inhalation study</p> <p>Cieszlak (1996)</p> <p>LOAEC: 11.6 mg/m³ air (nominal) (Olfactory epithelial degeneration).</p> <p>Rabbit 182-day study</p> <p>Ouyang Guoshun (1990)</p> <p>LOAEC: 15.3 mg/m³ air (nominal) (A wide range of chronic toxic effects, such as lesion in central nervous system, cardiovascular system, liver and kidney, and other degenerative changes in brain and coverings, were observed. Moreover, there were changes of cardiac EKG (not diagnostic of specified effects) and erythrocyte count. At 206 mg/m³, all the changes were more pronounced and the pathological lesions only got worsened after the exposure of GMA was ceased. On the other hands, the changes at 15.3 mg/m³ were sparse and slight, and almost all vanished one month after the exposure was ended.</p>	
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4.7.1.7 Summary and discussion of repeated dose toxicity

See chapter 4.8.1.

4.8 Specific target organ toxicity (CLP Regulation) – repeated exposure (STOT RE)

4.8.1 Summary and discussion of repeated dose toxicity findings relevant for classification as STOT RE according to CLP Regulation

The observed effects in the available oral and inhalation repeated dose studies indicate mainly local effects at the port of entry. Systemic effects were absent or limited and are not severe enough for classification. The local effects are due to the irritating/corrosive properties of GMA and are expected to be much more acute than repeated dose effects. Partially different toxicity effects were observed with repeated dose toxicity studies with glycidol probably because GMA is corrosive whereas glycidol is irritant (Table 19f). GMA induced more severe local effects at lower

doses/concentrations (oral: squamous hyperplasia in forestomach; inhalation: hyperplasia of respiratory epithelium of the nasal tissues), in comparison to glycidol (oral: no reported local effects; inhalation: Very slight irritation of the eyes, with slight lacrimation and encrustation of the eyelids and slight respiratory distress were observed following the first few exposures to glycidol). In terms of systemic effects, both compounds induced kidney and male reproductive (GMA: reduction in sperm motility; glycidol: testicular atrophy) toxic effects at comparable doses. However, glycidol induced effects on the brains that were not observed with GMA at an external dose level that was (or could) not be tested with GMA.

4.8.2 Comparison with criteria of repeated dose toxicity findings relevant for classification as STOT RE

According to Annex I: 3.9.1.1. of the CLP, ‘*specific target organ toxicity (repeated exposure) means specific, target organ toxicity arising from a repeated exposure to a substance or mixture. All significant health effects that can impair function, both reversible and irreversible, immediate and/or delayed are included.*’ In addition, Annex I: 3.9.1.6. of the CLP states that ‘*Non-lethal toxic effects observed after a single-event exposure are classified as described in Specific target organ toxicity — Single exposure (section 3.8) and are therefore excluded from section 3.9.*’. The effects observed after repeated oral and dermal exposure were limited to local effects on the port of entry and were caused by the irritating/corrosive properties of GMA. It is considered likely that these effects are concentration dependent and not on dose per kg bw and occur already after single exposure at concentrations not so different from the dose levels at which these effects were observed in the repeated dose study. Such effects are considered more relevant for acute classification. Therefore, based on the available studies and based on the other classifications already presented (STOT-SE Category 1 for inhalation and Acute Tox. category 3 and 4 for dermal and oral exposure), no further classification of GMA for this endpoint is required under CLP (see also sections 4.4.3.3 and 4.4.3.5).

4.8.3 Conclusions on classification and labelling of repeated dose toxicity findings relevant for classification as STOT RE

Based on the available studies and based on the other classifications already presented, no further classification of GMA for this endpoint is required under CLP (see also sections 4.4.3.3 and 4.4.3.5).

4.9 Germ cell mutagenicity (Mutagenicity)

4.9.1 Non-human information

4.9.1.1 *In vitro* data

Table 20a: Summary table of relevant *in vitro* mutagenicity studies

Method	Results	Remarks	Reference
Bacterial gene mutation assay (gene mutation) Salmonella typhimurium TA95, TA100 (met. act.: with and	Test results: positive for gene mutations in Salmonella typhimurium TA100 but not in TA95 ; met. act.: with and without ;	2 (reliable with restrictions) supporting study	Ouyang (1988) OECD (1999)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
without) Test concentrations: 112, 224, 448, 896 µg/plate Positive control substance(s): MMS, 2-aminofluorene, 2-amine anthracene	cytotoxicity: unknown	experimental result Test material: GMA purity 92%	
Bacterial gene mutation assay (gene mutation) bacteria, other: Klebsiella pneumoniae (met. act.: without) Test concentrations: 0.05, 0.1, 0.2, 0.5, 1.0 mmol/L	Test results: positive for gene mutations in bacteria, other: Klebsiella pneumoniae ; met. act.: with and without ; cytotoxicity: unknown	2 (reliable with restrictions) supporting study experimental result Test material: GMA purity 92%	Voogd (1981) OECD (1999)
mammalian cell gene mutation assay (gene mutation) Chinese hamster Ovary (CHO) (met. act.: with and without) Test concentrations: With metabolic activation: 25-600 microgram/ml Without metabolic activation: 5-80 microgram/ml Positive control substance(s): With metabolic activation: 20-methylcholantrene. Without metabolic activation: ethyl methanesulfonate chinese hamster ovary cell/hypoxanthine-guanine-phosphoribosyl transferase (CHO/HGPRT) forward gene mutation assay	Test results: Positive for gene mutations (Positive with metabolic activation, negative without metabolic activation) for Chinese hamster Ovary (CHO) (all strains/cell types tested) ; met. act.: with and without ; cytotoxicity: yes (With metabolic activation: 500 microgram/ml and more. Without metabolic activation: 50 microgram/ml and more)	2 (reliable with restrictions) supporting study experimental result Test material: GMA purity 99.5%	Linscombe (1995) OECD (1999)
<i>in vitro</i> mammalian chromosome aberration test (chromosome aberration) CHL/IU cell (met. act.: with and without); plates/test: 3; replicates: 2 Test concentrations: -S9 (continuous treatment): 0.0031, 0.0063, 0.013, 0.025, 0.050 mg/ml -S9 (short-term treatment):	Test results: Positive for chromosomal aberrations (Clastogenicity positive with and without metabolic activation. Polyploidy positive without metabolic activation, ambiguous with metabolic activation) for CHL/IU cell (all strains/cell types tested) ; met. act.: with and without ; cytotoxicity: yes (only without metabolic activation)	1 (reliable without restriction) key study experimental result Test material: GMA purity 99.93%	Ministry of Health and Welfare, Japan (1997) OECD (1999)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
<p>0.0055, 0.011, 0.022, 0.044, 0.088 mg/ml</p> <p>+S9 (short-term treatment): 0.022, 0.044, 0.088, 0.18, 0.35 mg/ml</p> <p>Positive control substance(s): mitomycine C for continuous treatment, cyclophosphamide for short term treatment</p> <p>OECD Guideline 473 (<i>In vitro</i> Mammalian Chromosome Aberration Test)</p>			
<p>SOS/umu test (SOS-Chromotest)</p> <p>E. coli, other: PQ37 (met. act.: with and without)</p> <p>Test concentrations: 0.1, 0.3 and 1.0 mmol/L</p>	<p>Test results:</p> <p>positive for genotoxic damage in E. coli, other: PQ37 ; met. act.: with and without ; cytotoxicity: unknown</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA purity 97%</p>	<p>Hude (1990)</p> <p>OECD (1999)</p>
<p>bacterial reverse mutation assay (e.g. Ames test)</p> <p>Salmonella typhimurium TA97, TA98, TA100, TA1535 (met. act.: with and without)</p> <p>Test concentrations: 0, 10, 33, 100, 333, 1000 microgram/plate</p> <p>Positive control substance(s): With metabolic activation: 2-aminoanthracene (all strains) Without metabolic activation: sodium azide (TA 100, TA 1535), 9aminoacridine (TA 97), 4-nitro-o-phenylenediamine (TA 98)</p> <p>bacterial reverse mutation assay</p>	<p>Test results:</p> <p>positive for gene mutations in Salmonella typhimurium TA97, TA100, TA1535 but not in TA98 ; met. act.: with and without ; cytotoxicity: unknown</p>	<p>2 (reliable with restrictions)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA purity 98%</p>	<p>Dorothy (1986)</p> <p>OECD (1999)</p>
<p>bacterial reverse mutation assay (e.g. Ames test)</p> <p>S. typhimurium TA 1535, TA 1537, TA 98 and TA 100 (met. act.: with and without)</p> <p>Test concentrations: With metabolic activation: 32, 100,</p>	<p>Test results:</p> <p>positive for gene mutations in S. typhimurium TA 1535, TA 1537 and TA 100 but not in TA98 ; met. act.: with and without ; cytotoxicity: unknown</p>	<p>2 (reliable with restrictions)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Goodyear (1981)</p> <p>OECD (1999)</p>

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
<p>320, 1,000 µg/plate</p> <p>Without metabolic activation: 100, 320, 1,000, 2,000 µg/plate</p> <p>Positive control substance(s): With metabolic activation: 2-aminofluorene, 2-aminoanthracene, dimethylbenz(a)anthracene Without metabolic activation: 2-nitrofluorene, sodium azide, quinacrine mustard</p> <p>Health, Safety and Government Compliance test method, 79-10</p>			
<p>Analysis of the phenotype and the restriction enzyme mapping level of mutations</p> <p>E. coli, other: HB101 (met. act.: not applicable)</p> <p>Test concentrations: 0.1, 0.3 and 1.0 mmol/L</p>	<p>Test results:</p> <p>positive gene mutations in E. coli, other: HB101 ; met. act.: not applicable ; cytotoxicity: unknown</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Xie (1990a)</p> <p>OECD (1999)</p>
<p>sister chromatid exchange assay in mammalian cells</p> <p>Chinese hamster lung fibroblasts (V79) (met. act.: without)</p> <p>Test concentrations: 0, 0.02, 0.039, 0.078, 0.16, 0.31 mM</p>	<p>Test results:</p> <p>positive for chromosomal aberrations in Chinese hamster lung fibroblasts (V79) ; met. act.: without ; cytotoxicity: unknown</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA purity 97%</p>	<p>Hude (1991)</p> <p>OECD (1999)</p>
<p>DNA damage and repair assay, unscheduled DNA synthesis in mammalian cells <i>in vitro</i></p> <p>human lymphocytes</p> <p>Test concentrations: no data</p>	<p>Test results:</p> <p>positive for DNA damage in human lymphocytes</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Xie (1990b)</p> <p>OECD (1999)</p>
<p>DNA damage and repair assay, unscheduled DNA synthesis in mammalian cells <i>in vitro</i></p> <p>rat lymphocytes</p> <p>Test concentrations: no data</p>	<p>Test results:</p> <p>positive for DNA damage in rat lymphocytes</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Xie (1990b)</p> <p>OECD (1999)</p>
<p>DNA replication</p>	<p>Test results:</p>	<p>2 (reliable with</p>	<p>Xie (1989)</p>

Method	Results	Remarks	Reference
human lymphocytes Test concentrations: no data	positive for non-reverse type inhibition of DNA replication in human lymphocytes	restrictions) supporting study experimental result Test material: GMA	OECD (1999)
DNA replication rat lymphocytes Test concentrations: no data	Test results: positive for non-reverse type inhibition of DNA replication in rat lymphocytes	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Xie (1989) OECD (1999)
<i>in vitro</i> mammalian cell transformation assay Golden Syrian hamster embryo cells (diploid) Test concentrations: 0.9 - 14.2 mg/L	Test results: positive for transformation in Golden Syrian hamster embryo cells (diploid)	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Yang (1996) OECD (1999)
<i>in vitro</i> mammalian cell transformation assay Syrian hamster embryonic cells Test concentrations: no data	Test results: positive for transformation in Syrian hamster embryonic cells (diploid)	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Xie (1992) OECD (1999)
DNA binding study Calf thymus DNA (met. act.: not applicable) Test concentrations: no data	Test results: Positive for DNA binding to Calf thymus DNA ; met. act.: not applicable	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Xie (1990b) OECD (1999)

Bacterial tests

GMA was mutagenic to *Salmonella typhimurium* TA97, TA100, TA1535 with and without metabolic activation but not to TA98 (Dorothy et al.: 1986, The Goodyear Tire & Rubber Company: 1981 (key study), OuYang et al.: 1988).

GMA was mutagenic to *Klebsiella pneumoniae* without metabolic activation (Voogd et al.: 1981). In *Escherichia coli*, this substance induced SOS repair with and without metabolic activation (von

der Hude et al.: 1990). This substance was shown to react with the DNA of the gene governing tetracycline resistance in the plasmid pBR322. The modified DNA was transferred to a receptor cell (Escherichia coli HB 101) to screen for mutations based on alterations in phenotypic changes. Results showed the mutations caused by reactions of GMA with the plasmid were stable and heritable (Xie et al.: 1990a).

Non-bacterial tests *in vitro*

Key studies

Ministry of Health and Welfare, Japan (1997)

Structural chromosomal aberrations (including gap) and polyploidy were induced. Structural chromosome aberrations were induced by GMA in CHL/ IU cells with and without metabolic activation (Table 20b and c). However, a trend test showed no dose-dependency for the induction of polyploidy with the 24 hours continuous treatment and the short-term treatment with the metabolic activation system.

Table 20b: Summary of *in vitro* mutagenicity studies with GMA (Ministry of Health and Welfare, Japan, 1997)

Table 1 Chromosome analysis of Chinese hamster cells (CHL/IU) continuously treated with 2, 3- epoxypropyl methacrylate (EPMA)** without S9 mix

Group	Concentration (mg/ml)	Time of exposure (h)	No. of cells analysed	No. of structural aberrations						Others ³⁾	No. of cells with aberrations		Polyploid ⁴⁾ (%)	Trend test ⁵⁾		Concurrent ⁶⁾ cytotoxicity (%)	
				gap	ctb	cte	csb	cse	mul		total	TAG (%)		TA (%)	SA		NA
Control ¹⁾	0	24	200	0	0	3	5	0	0	8	0	2 (1.0)	2 (1.0)	0.38			—
Solvent	0.0063	24	200	1	1	0	0	0	0	2	0	2 (1.0)	1 (0.5)	0.25			100.0
EPMA	0.013	24	200	1	2	1	0	0	0	4	0	4 (2.0)	3 (1.5)	0.26 ⁷⁾			50.0
EPMA	0.025	24	200	1	5	1	0	0	0	7	1	7*(3.5)	6 (3.0)	1.38*	+	-	66.1
EPMA	0.050 ***	24	—	—	—	—	—	—	—	—	—	—	—	0.50			41.9
MC	0.00005	24	200	5	35	118	1	0	20	179	0	95 (47.5)	95 (47.5)	0.25			—
Solvent ¹⁾	0	48	200	0	0	0	0	0	0	0	5	0 (0.0)	0 (0.0)	0.00 ⁸⁾			100.0
EPMA	0.0063	48	200	1	0	1	2	0	0	4	0	4 (2.0)	3 (1.5)	0.38			62.8
EPMA	0.013	48	200	0	2	2	2	0	0	6	0	4 (2.0)	4 (2.0)	0.52 ⁹⁾	+	+	30.2
EPMA	0.025	48	200	8	56	141	5	0	80	290	0	97*(48.5)	95 (47.5)	9.40 ¹⁰⁾ *			32.6
EPMA	0.050 ***	48	—	—	—	—	—	—	—	—	—	—	—	—			—
MC	0.00005	48	200	4	38	88	1	4	0	135	5	66 (33.0)	64 (32.0)	0.25			—

Abbreviations, gap : chromatid gap and chromosome gap, ctb : chromatid break, cte: chromatid exchange, csb : chromosome break, cse : chromosome exchange (dicentric and ring), mul : multiple aberrations, TAG : total no. of cells with aberrations, TA : total no. of cells with aberrations except gap, SA : structural aberration, NA : numerical aberration, MC : mitomycin C.

1) Dimethylsulfoxide was used as solvent. 2) More than nine aberrations in a cell were scored as 10. 3) Others, such as attenuation and premature chromosome condensation, were excluded from the no. of structural aberrations. 4) Eight hundred cells were analysed in each group. 5) Cochran • Armitage's trend test was done at p<0.05. 6) Relative metaphase frequency to the solvent control, representing cytotoxicity, was calculated. 7) Seven hundred and eighty one cells were analysed. 8) Seven hundred and ninety one cells were analysed. 9) Seven hundred and sixty three cells were analysed. 10) Seven hundred and sixty six cells were analysed.

* : Significantly different from historical solvent control data at p<0.05 by Fisher's exact test using a Bonferroni correction for multiple comparisons. ** : Purity of test substance was 99.93 %. 2- Methyl- 3- methoxypropanoic acid 2, 3- epoxypropyl ester (0.07 %) and hydroquinone monomethyl ether (46 ppm) were contained as impurities. *** : Chromosome analysis was not performed because there were small number of metaphase due to cytotoxicity.

Table 20c: Summary of *in vitro* mutagenicity studies with GMA (Ministry of Health and Welfare, Japan, 1997)

Table 2 Chromosome analysis of Chinese hamster cells (CHL/IU) treated with 2, 3- epoxypropyl methacrylate (EPMA)** with and without S9 mix

Group	Concentration (mg/ml)	S9 mix	Time of exposure (h)	No. of cells analysed	No. of structural aberrations							Others ³⁾	No. of cells with aberrations		Polyploid ⁴⁾ (%)	Trend test ⁵⁾		Concurrent ⁶⁾ cytotoxicity (%)
					gap	ctb	cte	csb	cse	mul ²⁾	total		TAG (%)	TA (%)		SA	NA	
Control ¹⁾	0	-	6-(18)	200	1	0	0	0	0	0	1	0	1 (0.5)	0 (0.0)	0.13			—
Solvent	0	-	6-(18)	200	1	0	0	0	0	0	1	0	1 (0.5)	0 (0.0)	0.25			100.0
EPMA	0.011	-	6-(18)	200	0	1	1	0	0	0	2	0	2 (1.0)	2 (1.0)	0.13			72.1
EPMA	0.022	-	6-(18)	200	0	0	4	0	0	0	4	0	4 (2.0)	4 (2.0)	0.75	+	+	56.6
EPMA	0.044	-	6-(18)	200	6	45	59	0	2	10	122	0	57*(28.5)	53 (26.5)	0.88 *			24.6
EPMA	0.088 ***	-	6-(18)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	
CPA	0.005	-	6-(18)	200	0	0	0	0	0	0	0	1	0 (0.0)	0 (0.0)	0.13			—
Solvent ¹⁾	0	+	6-(18)	200	0	0	1	0	0	0	1	0	1 (0.5)	1 (0.5)	0.13			100.0
EPMA	0.044	+	6-(18)	200	0	1	0	0	0	0	1	0	1 (0.5)	1 (0.5)	0.38			117.3
EPMA	0.088	+	6-(18)	200	0	5	4	2	0	0	11	0	7 (3.5)	7 (3.5)	0.88 *	+	-	94.2
EPMA	0.18	+	6-(18)	200	1	5	24	0	0	0	30	0	19*(9.5)	18 (9.0)	0.38			80.8
EPMA	0.35 ***	+	6-(18)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	
CPA	0.005	+	6-(18)	200	17	30	129	0	0	0	176	0	102 (51.0)	98 (49.0)	0.39 ⁷⁾			—

Abbreviations, gap : chromatid gap and chromosome gap, ctb : chromatid break, cte: chromatid exchange, csb : chromosome break, cse : chromosome exchange (dicentric and ring), mul : multiple aberrations, TAG : total no. of cells with aberrations, TA : total no. of cells with aberrations except gap, SA : structural aberration, NA : numerical aberration, CPA : cyclophosphamide.

1) Dimethylsulfoxide was used as solvent. 2) More than nine aberrations in a cell were scored as 10. 3) Others, such as attenuation and premature chromosome condensation, were excluded from the no. of structural aberrations. 4) Eight hundred cells were analysed in each group. 5) Cochran-Armitage's trend test was done at p<0.05. 6) Relative metaphase frequency to the solvent control, representing cytotoxicity, was calculated. 7) Seven hundred and seventy nine cells were analysed. *: Significantly different from historical solvent control data at p<0.05 by Fisher's exact test using a Bonferroni correction for multiple comparisons. **: Purity of test substance was 99.93 %. 2-Methyl- 3- methoxypropanoic acid 2, 3- epoxypropyl ester (0.07 %) and hydroquinone monomethyl ether (46 ppm) were contained as impurities. *** : Chromosome analysis was not performed because there were small number of metaphases due to cytotoxicity.

In cell cultures, GMA induced hypoxanthine-guanine-phosphoribosyl transferase forward gene mutation with metabolic activation in Chinese hamster ovary cell (Linscombe and Engle: 1995), very slight increase of unscheduled DNA synthesis in lymphocytes of human and/or rat (Xie et al.: 1990b), non-reverse type inhibition of the DNA replication in lymphocytes of human and/or rat (Xie et al.: 1989), sister-chromatid exchange without metabolic activation in Chinese hamster V79 cells (von der Hude et al.: 1991), transformation of Syrian hamster embryonic cells (SHE) (Xie et al., 1992) and transformation in diploid golden Syrian hamster embryo (SHE) cells (Yang et al.; 1996). This substance was strongly and covalently bound with calf thymus DNA *in vitro* (Xie et al.: 1990b).

4.9.1.2 *In vivo* data

Table 21a: Summary table of relevant *in vivo* mutagenicity studies

Method	Results	Remarks	Reference
<p>Micronucleus assay</p> <p>mouse (BDF1)</p> <p>5 male + 5 female/group</p> <p>oral: gavage</p> <p>Male: 188, 375 and 750 mg/kg. Female 250, 500 and 1000 mg/kg (nominal in diet)</p> <p>Positive control substance(s): yes, cyclophosphamide (50 mg/kg)</p> <p>OECD Guideline 474 (Mammalian Erythrocyte Micronucleus Test)</p>	<p>Test results:</p> <p>Mutagenicity: positive (male/female)</p> <p>Micronucleus test in mice is positive after 48 h but only at the highest doses.</p>	<p>1 (reliable without restriction)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA purity 99.93%</p>	<p>Ministry of Health and Welfare, Japan (1997)</p> <p>OECD (1999)</p>
<p>Transgenic animal mutagenicity assay</p> <p>rat (transgenic Big Blue(R) Fischer 344 rats) male</p> <p>inhalation: vapour</p> <p>6 hours per day, 5 days per week for 2 weeks followed by 4 weeks for fixation.</p> <p>1, 10, 25 ppm (5.82, 58.2, 145.5 mg/m³. Calculated daily dose: 0.71, 7.08, 17.70 mg/kg/day) (nominal conc.)</p>	<p>Test results:</p> <p>Mutagenicity: negative (male) in olfactory and respiratory epithelium</p>	<p>2 (reliable with restrictions)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Gollapudi et al. (1999)</p> <p>OECD (1999)</p>
<p>Micronucleus assay</p> <p>mouse male</p> <p>intraperitoneal (2 treatments, 24 h</p>	<p>Test results:</p> <p>Mutagenicity: ambiguous (male)</p> <p>There was an increase in the</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>Test material:</p>	<p>Ou-Yang (1988)</p> <p>OECD (1999)</p>

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
<p>apart)</p> <p>25, 50 and 100 mg/kg bw</p> <p>Positive control substance(s): methyl pterine</p>	<p>number of cells with micronuclei 6h after last treatment, but this change was very slight and inversed dose-response.</p>	<p>GMA purity 92%</p>	
<p>Micronucleus assay</p> <p>mouse (CD-1 (ICR) BR);</p> <p>5 male + 5 female per group</p> <p>intraperitoneal (single)</p> <p>75, 150 and 300 mg/kg</p> <p>Positive control substance(s): Cyclophosphamide (120 mg/kg)</p> <p>At 24, 48 and 72 hours after exposure.</p> <p>40 CFR 798.5395 (The Dow Chemical Company)</p>	<p>Test results:</p> <p>Mutagenicity: negative (male/female)</p>	<p>1 (reliable without restriction)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA purity 99.5%</p>	<p>Lick (1995)</p> <p>OECD (1999)</p>
<p>Micronucleus assay</p> <p>Mouse</p> <p>intraperitoneal</p> <p>42.2, 133, 422, 464 mg/kg bw</p>	<p>Test results:</p> <p>Mutagenicity: negative</p>	<p>4 (not assignable)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>INBIFO (1979)</p> <p>OECD (1999)</p>
<p>Unscheduled DNA synthesis</p> <p>5 Kummimg hybrid male mice/group</p>	<p>Test results:</p> <p>Genotoxicity: ambiguous (male)</p> <p>Unscheduled DNA synthesis was increased in the germ cells, but this change was very slight and not dose related.</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Xie (1990b)</p> <p>OECD (1999)</p>

In vivo tests

In a micronucleus assay, mice were administered by gavage with GMA at a single dose of 188, 375 and 750 mg/kg in males and 250, 500 and 1000 mg/kg in females. The frequency of micronucleated polychromatic erythrocytes in both sexes (Table 21c and 21d) was significantly increased only at the highest doses 48 hour after administration. (Ministry of Health and Welfare, Japan: 1997).

Table 21b: Summary of *in vivo* mutagenicity study with GMA (Ministry of Health and Welfare, Japan, 1997)

Table 7. Results of micronucleus test in BDF₁ male mice after single administration of 2,3-epoxypropyl methacrylate by gavage

Group	Animal No.	a		b	
		MNPCE / PCE		PCE / ERY	
Solvent control Olive oil 10 ml/kg	1	5 / 2000		599 / 1000	
	2	6 / 2000		542 / 1000	
	3	4 / 2000		569 / 1000	
	4	4 / 2000		548 / 1000	
	5	1 / 2000		609 / 1000	
	Total	20 / 10000		2867 / 5000	
	%(Mean±S.D.)	(0.20 ± 0.09)		(57.3 ± 3.0)	
EPMA 188 mg/kg	6	3 / 2000		521 / 1000	
	7	1 / 2000		585 / 1000	
	8	6 / 2000		578 / 1000	
	9	6 / 2000		569 / 1000	
	10	1 / 2000		555 / 1000	
	Total	17 / 10000		2808 / 5000	
	%(Mean±S.D.)	(0.17 ± 0.13)		(56.2 ± 2.5)	
EPMA 375 mg/kg	11	2 / 2000		568 / 1000	
	12	5 / 2000		571 / 1000	
	13	1 / 2000		495 / 1000	
	14	3 / 2000		527 / 1000	
	15	4 / 2000		543 / 1000	
	Total	15 / 10000		2704 / 5000	
	%(Mean±S.D.)	(0.15 ± 0.08)		(54.1 ± 3.1)	
EPMA 750mg/kg	16	27 / 2000		474 / 1000	
	17	12 / 2000		523 / 1000	
	18	14 / 2000		412 / 1000	
	19	7 / 2000		394 / 1000	
	20	20 / 2000		414 / 1000	
	Total	80 / 10000		2217 / 5000	
	%(Mean±S.D.)	(0.80 ± 0.39)***		(44.3 ± 5.4)**	
Positive control CPA 50 mg/kg	21	56 / 2000		407 / 1000	
	22	40 / 2000		482 / 1000	
	23	22 / 2000		365 / 1000	
	24	38 / 2000		401 / 1000	
	25	62 / 2000		437 / 1000	
	Total	218 / 10000		2092 / 5000	
	%(Mean±S.D.)	(2.18 ± 0.79)***		(41.8 ± 4.4)***	

a: Number of micronucleated polychromatic erythrocytes / total number of polychromatic erythrocytes observed

b: Number of polychromatic erythrocytes / total number of erythrocytes observed

CPA: Cyclophosphamide

EPMA: 2,3-epoxypropyl methacrylate

Purity was 99.93 wt % and 0.07 wt % 2,3-epoxypropyl 2-methyl-3-methoxypropanoate and 46 ppm hydroquinone monomethyl ether were contained as impurities.

** : Data significantly different from the solvent control at 1 % level

*** : Data significantly different from the solvent control at 0.1 % level

Table 21c: Summary of *in vivo* mutagenicity study with GMA (Ministry of Health and Welfare, Japan, 1997)

 Results of micronucleus test in BDF₁ female mice after single administration of 2,3-epoxypropyl methacrylate by gavage

Group	Animal No.	a		b	
		MNPCE / PCE		PCE / ERY	
Solvent control Olive oil 10 ml/kg	51	5 / 2000		614 / 1000	
	52	1 / 2000		602 / 1000	
	53	5 / 2000		639 / 1000	
	54	5 / 2000		616 / 1000	
	55	3 / 2000		573 / 1000	
	Total		19 / 10000		3044 / 5000
	%(Mean±S.D.)	(0.19 ± 0.09)		(60.9 ± 2.4)	
EPMA 250 mg/kg	56	6 / 2000		572 / 1000	
	57	2 / 2000		648 / 1000	
	58	4 / 2000		617 / 1000	
	59	5 / 2000		554 / 1000	
	60	4 / 2000		581 / 1000	
	Total		21 / 10000		2972 / 5000
	%(Mean±S.D.)	(0.21 ± 0.07)		(59.4 ± 3.8)	
EPMA 500 mg/kg	61	9 / 2000		621 / 1000	
	62	5 / 2000		634 / 1000	
	63	10 / 2000		582 / 1000	
	64	5 / 2000		572 / 1000	
	65	3 / 2000		580 / 1000	
	Total		32 / 10000		2989 / 5000
	%(Mean±S.D.)	(0.32 ± 0.15)		(59.8 ± 2.8)	
EPMA 1000mg/kg	66	15 / 2000		467 / 1000	
	67	9 / 2000		599 / 1000	
	68	13 / 2000		503 / 1000	
	69	20 / 2000		527 / 1000	
	70	6 / 2000		556 / 1000	
	Total		63 / 10000		2652 / 5000
	%(Mean±S.D.)	(0.63 ± 0.27)***		(53.0 ± 5.0)*	
Positive control CPA 50 mg/kg	71	66 / 2000		547 / 1000	
	72	71 / 2000		622 / 1000	
	73	55 / 2000		563 / 1000	
	74	45 / 2000		548 / 1000	
	75	33 / 2000		515 / 1000	
	Total		270 / 10000		2795 / 5000
	%(Mean±S.D.)	(2.70 ± 0.77)***		(55.9 ± 3.9)	

a: Number of micronucleated polychromatic erythrocytes / total number of polychromatic erythrocytes observed

b: Number of polychromatic erythrocytes / total number of erythrocytes observed

CPA: Cyclophosphamide

EPMA: 2,3-epoxypropyl methacrylate

Purity was 99.93 wt % and 0.07 wt % 2,3-epoxypropyl 2-methyl-3-methoxypropanoate and 46 ppm hydroquinone monomethyl ether were contained as impurities.

*: Data significantly different from the solvent control at 5 % level

***: Data significantly different from the solvent control at 0.1 % level

Three additional mouse bone marrow micronucleus tests by intraperitoneal (ip) administration have been reported. In one study, this substance produced a 2-3 fold increase in micronuclei at all administered doses relative to the control although it showed an inverse dose response (Ou-Yang et al., 1988). On the other hand, this substance did not cause an increase in the number of cells containing micronuclei in two other ip injection studies with GMA doses of 75, 150, and 300 mg/kg (Lick et al., 1995) or doses of 42.2, 133, 422, and 464 mg/kg (INBIFO: 1979). The study by Lick (1995) fulfils the OECD criteria. The highest dose of 300 mg/kg bw was based on a range finding in

which 100% mortality occurred at 500 mg/kg bw and no mortality at 250 mg/kg bw. At the highest dose tested there was no change in the PCE%.

Lick (1995)

Glycidyl Methacrylate (GMA) was evaluated in the mouse bone marrow micronucleus test. The micronucleus test is capable of detecting agents causing chromosomal aberrations and spindle malfunction. The test material was administered to CD-1 mice by single intraperitoneal injection at dose levels of 0 (negative control), 75, 150, and 300 mg/kg body weight (BW). The highest dose level of 300 mg/kgBW was based upon the results of a range-finding test. The concentrations of the test material in the dosing solutions were verified by either gas chromatography with flame ionization detection or high performance liquid chromatography. Groups of animals were sacrificed at 24, 48, or 72 h after treatment. Mice treated orally with 120 mg/kgBW cyclophosphamide and sacrificed at 24 h served as positive controls. There were five animals per sex per dose level per sacrifice time. One thousand polychromatic erythrocytes (PCE) were evaluated from each surviving animal and the frequencies of micronucleated polychromatic erythrocytes (MN-PCE) were recorded. There were no statistically significant increases in the frequencies of MN-PCE in groups treated with the test material as compared to negative controls. The positive control mice showed significant increases in MN-PCE. Hence, under the experimental conditions used, the test material was considered to be negative in the mouse bone marrow micronucleus test.

Xie (1990b)

Sperm abnormality tests and assays of unscheduled DNA synthesis in germ cells of male mice were conducted to study the *in vivo* genotoxicity of GMA. The results revealed that GMA could damage DNA, increase sperm abnormality frequency, and reduce the number of sperm cells. This substance increased unscheduled DNA synthesis in germ cell of male mice but this effect was very slight (~25% above controls for all doses administered) and not dose-related (Xie et al.: 1990b).

Assay of UDS in the Germ Cell of Male Mice Exposed in Vivo to the GMA

Kunming hybrid male mice, 10–14 weeks old at the start of the experiment, were used. GMA was dissolved in peanut oil, and the concentration of GMA was adjusted so that a standard 30-g mouse would receive an ip injection of 0.5 ml. The animals were randomly divided into five groups with five mice in each group. The negative control animals were injected ip with peanut oil. Those in the GMA experiments were injected ip with 25, 50, and 100 mg of GMA per kg body wt, respectively. Those in the positive control group were injected ip with 200 mg of cyclophosphamide (CP) per kg body wt. Each mouse was immediately anesthetized with ether after the injections were given. Each of these animals received a dose of 72 μ Ci of [³H]TdR divided into equal parts and injected separately into testis in 0.05 ml of 2% ethanol. All animals were killed 16 days after the treatments. The caudal epididymides were removed and washed with physiological saline. After filtration and centrifugation, the sperm cells were recovered for hemacytometer counts. Thereafter the sperm cells were transferred onto glass fiber membrane and dried and a toluene:PPO:POPOP scintillation mixture was added. The radioactivity was measured in a Beckman liquid scintillation counter. Results were expressed as mean values of cpm/10⁶ sperm cells.

Table 21d: Unscheduled DNA synthesis induced by GMA in germ cells of male mice (Xie et al. 1990b)

UDS Induced by GMA in Germ Cells of Male Mice			
Group	Dose (mg/kg)	No. of animals	cpm/10 ⁶ Sperms ($\bar{X} \pm SD$)
Control	0	6	3.77 ± 0.09
GMA	25	5	5.07 ± 0.34*
	50	5	4.98 ± 0.15*
	100	5	4.99 ± 0.19*
	200	5	5.30 ± 0.34*
CP ^b	200	5	5.30 ± 0.34*

* Control vs GMA group, *P* < 0.01.

^b Cyclophosphamide.

GMA was evaluated in an *in vivo* assay for the induction of gene mutations at the lacI locus of transgenic Big Blue Fischer 344 (F-344) rats. The rats (15 males/group) were exposed to GMA vapours by inhalation at targeted concentrations of 0 (negative control), 1, 10, and 25 ppm for 2 weeks, 6 hrs/day, 5 days/week followed by a 4 week fixation period. There were no statistically significant increases in the frequencies of lacI mutants in either the olfactory or respiratory epithelium of rats exposed to 25 ppm (145.5 mg/cm³) (17.7 mg/kg/day) when compared to the corresponding negative control values. Based upon these results, the authors concluded that GMA was not mutagenic to the nasal epithelium of rats under the conditions of the study (Gollapudi et al., 1999).

4.9.2 Human information

No relevant information available

4.9.3 Other relevant information

Table 21e: Additional mutagenicity data on Glycidol for read-across analysis

Method	Results	Remarks	Reference
micronucleus assay (chromosome aberration) mouse (B6C3F1) male intraperitoneal 2 times at 24h intervals Positive control substance(s): Mitomycin C (1 mg/kg) OECD Guideline 474 (Mammalian Erythrocyte Micronucleus Test)	Evaluation of results: positive Test results: Genotoxicity: positive (male); toxicity: no effects ; vehicle controls valid: yes; negative controls valid: yes; positive controls valid: yes	1 (reliable without restrictions) supporting study experimental result Test material: 2,3-epoxypropan-1- ol (glycidol) purity 94% Form: liquid	Irwin et al. (1990)

Metabolism of GMA to glycidol

In view of the fact that GMA is metabolised to glycidol, information is being provided below on the genotoxicity of glycidol, as supporting data for an evaluation of the genotoxic potential of GMA.

Glycidol was evaluated for the potential to cause micronuclei in mouse bone marrow *in vivo* (NTP, 1990). Male mice were given two intraperitoneal injections of 37.5, 75 and 150 mg/kg, 24 hours apart, with the glycidol dissolved in phosphate-buffered saline; the total dose volume was 0.4 ml. Solvent control animals were injected with 0.4 ml phosphate-buffered saline only. The positive control mice received injections of mitomycin C. Twenty-four hours after the second injection, the mice were killed by cervical dislocation, and smears were prepared of the bone marrow cells obtained from the femurs. Air-dried smears were fixed and stained; 2,000 PCEs were scored for the number of micronucleated cells in each of five animals per dose group. The results were tabulated as the mean of the pooled results from all animals within a dose group +/- standard error of the mean. Preliminary range-finding studies were performed to determine appropriate doses for the *in vivo* micronucleus test. Dose selection in this study was based on animal lethality; no decrease in the percentage of polychromatic erythrocytes (PCEs) in the bone marrow was observed in any of the dose groups. In the first trial, the incidence of micronuclei was increased statistically above concurrent phosphate-buffered saline controls. Micronuclei incidence was increased at doses of 75 and 150 mg/kg. In a second trial, incidences of micronuclei were statistically increased above concurrent control values at 37.5 and 150 mg/kg but not at 75 mg/kg. (It is noted that the control for the second trial was lower than the control in the first trial.)

Table 21f: Summary of micronucleus study with Glycidol (NTP, 1990)

TABLE H7. INCIDENCE OF MICRONUCLEI IN BONE MARROW POLYCHROMATIC ERYTHROCYTES OF MICE ADMINISTERED GLYCIDOL (a)

	Dose (mg/kg)	Micronucleated Cells/1,000 Cells (b)	
		Trial 1	Trial 2
Vehicle controls (c)	0	1.5 ± 0.4	0.6 ± 0.2
Glycidol	37.5	1.5 ± 0.3	1.3 ± 0.3
	75	2.4 ± 0.4	0.7 ± 0.3
	150	4.4 ± 0.8	1.9 ± 0.6
		P < 0.001	0.01 < P < 0.05
Mitomycin C (d)	1	37.7 ± 4.6	30.2 ± 2.7

(a) Study performed at Environmental Health Research and Testing, Inc. Glycidol, dissolved in phosphate-buffered saline, was administered by intraperitoneal injection to male B6C3F₁ mice two times, at 24-hour intervals; bone marrow smears were prepared 24 hours after the second injection. For each trial, 2,000 polychromatic erythrocytes were scored for the number of micronuclei in each of five animals per dose group.

(b) Mean ± standard error of the mean

(c) Vehicle control animals received injections of 0.4 ml phosphate-buffered saline.

(d) Positive control material was dissolved in phosphate-buffered saline and administered by intraperitoneal injection.

Glycidol has a harmonised classification as a germ cell mutagen category 2 under CLP.

4.9.4 Summary and discussion of mutagenicity

Summary

Genotoxicity studies on GMA *in vitro* showed positive results. In micronucleus tests *in vivo*, oral administration of GMA increased the frequency of micronucleated polychromatic erythrocytes at the highest dose only, while mostly negative results were shown in other *in vivo* genotoxicity studies including micronucleus tests by intraperitoneal administration and including the gene mutation study using transgenic Big Blue Fischer 344 rats. The negative i.p. micronucleus test (Lick, 1995) was performed with lower exposure levels compared to the positive oral study. This could be explained by a higher mortality after i.p. exposure of the unprotected peritoneal compared

to the stomach after oral exposure. The dose exposure levels applied in the i.p. studies were also negative in the oral study. Also no decrease in PCE% was observed in the i.p study at the highest dose level which was observed at the highest dose level in the oral study. Further, *in vitro* and *in vivo* studies indicate carboxylesterase mediated hydrolysis of GMA to glycidol. Glycidol, a metabolite of GMA, is classified as a Category 2 germ cell mutagen under CLP. Based on the available studies for GMA itself and read across data for glycidol, GMA is considered to be a substance mutagenic towards somatic cells. There is only one *in vivo* study on germ cells which showed a significant increase in UDS in sperm cells. However, the increase was small and not dose related.

4.9.5 Comparison with criteria

Substances which cause concern for humans owing to the possibility that they may induce heritable mutations in the germ cells of humans.

The classification in Category 2 is based on:

- positive evidence obtained from experiments in mammals and/or in some cases from *in vitro* experiments, obtained from:
 - somatic cell mutagenicity tests *in vivo*, in mammals; or
 - other *in vivo* somatic cell genotoxicity tests which are supported by positive results from *in vitro* mutagenicity assays.

Note: Substances which are positive in *in vitro* mammalian mutagenicity assays, and which also show chemical structure activity relationship to known germ cell mutagens, shall be considered for classification as Category 2 mutagens.

This proposal is based on a positive somatic cell mutagenicity test *in vivo* in mammals (micronuclei tests, Ministry of Health and Welfare, 1997)) although a comparable test was negative. This could be explained by the lower dose level in the negative test. However, the positive tests are supported by positive tests *in vitro* and by the formation of the metabolite glycidol, which is already classified as a category 2 germ cell mutagen.

There is only very limited information regarding germ cell mutagenicity in which only a slight and not dose-related increase in unscheduled DNA synthesis was observed in the germ cells. This is not sufficient for category 1B.

Category 1A is not applicable as there are no human data.

4.9.6 Conclusions on classification and labelling

According to the available studies GMA must be classified according to CLP for Germ cell Mutagenicity Category 2 (H341).

4.10 Carcinogenicity

4.10.1 Non-human information

4.10.1.1 Carcinogenicity: oral

In a very limited one-year study (Hadidian et al., 1968), rats (3 males and 3 females) were dosed 5 days/week by gavage at 0.1 mg/kg. Groups of 15 male and 15 female rats were also dosed at 0.3 mg/kg. The authors concluded that GMA gave a tumor incidence pattern similar to that of

controls. However the doses applied are considered to be too low and the number of animals was very small.

Table 22: Summary of carcinogenicity study (Hadidian et al. 1968)

Compound	(Number)	Dose† (mg)	Number and sex	Average‡		Average body weight (g)			Liver (g)	Neoplastic lesions, No. of rats with lesions	Non-neoplastic lesions
				Number Rx	Survival (days)	Initial	Week 18	Final			
Glycidyl methacrylate.....	(26)	0.3	3 M	260	557	65	302	447	17(16-17)		
			3 F	260	451	67	182	305	12(10-14)	Ad-Lu 1; FAd-Ma 1	
		0.1	15 M	257	509	64	275	416	17(10-22)	Io-Ts 4	Hep-Tox 1
			15 F	260	526	64	176	288	11 (8-15)	Ly 1; Ad-Br 1	Pol-Ut 1
		0.001 -0.03	12 M	260	541	62	287	430	18(14-21)	Io-Ts 5	Hep-Tox 1
			12 F	260	564	48	175	306	12 (8-16)	FAd-Ma 2	Pol-Ut 2

*For abbreviations see table 3. †Dose refers to mg per day per animal. ‡Number Rx = number of individual doses; Survival = survival period for group.

Organs				Lesions			
Adr = Adrenal	Ma = Mammary	AdCa = Adenocarcinoma	He = Hepatoma				
Br = Bronchus	Mo = Mouth	Ad = Adenoma	Hyp = Hypertrophy				
Bl = Bladder	Pan = Pancreas	At = Atrophy	Hypl = Hyperplasia				
Ce = Cecum	Ph = Pharynx	Ba = Basal cell	Io = Interstitial cell tumor				
Co = Colon	Pi = Pituitary	Ca = Carcinoma	Lei = Leiomyoma				
Duo = Duodenum	Pr = Prostate	Ci = Cirrhosis	Lp = Lipoma				
Ea = Ear	Seb = Sebaceous gland	Ch = Cholangioma	Ly = Lymphoma				
En = Endometrium	Sk = Skin	Cy = Cyst	Me = Mesothelioma				
Es = Esophagus	Sp = Spleen	FAd = Fibroadenoma	Met = Metastasis				
Hep = Liver	St = Stomach	Fi = Fibroma	Pa = Papilloma				
Il = Ileum	Th = Thyroid	FSA = Fibrosarcoma	Poi = Polyp				
Int = Intestine	Tn = Tongue	Gr = Granulosa cell tumor	Sa = Sarcoma				
Ki = Kidney	Tr = Trachea	Gran = Granuloma	Sq = Squamous cell				
La = Larynx	Ts = Testes		Tox = Toxic				
Li = Lip	Ut = Uterus						
Lu = Lung							

4.10.1.2 Carcinogenicity: inhalation

In a 26-weeks inhalation toxicity study at concentrations of 15.3 and 206 mg/m³ in rats and rabbits (Ouyang Guoshun et al., 1990), a wide range of toxic effects were observed in both species at both concentrations. A wide range of chronic toxic effects, such as changes of liver and spleen weight, and enzyme (transaminase) levels in blood or tissue, and lesion in central nervous system, cardiovascular system, liver and kidney, were observed. At 206 mg/m³, all the changes were more pronounced and the pathological lesions only got worsened after the exposure was ceased. On the other hand, the changes at 15.3 mg/m³ were sparse and slight, and almost all vanished one month after the exposure had ended. Because of the higher vapor pressure and lower purity, the author suggested that the test material used in this study contained components other than GMA, which may have contributed to the toxicity observed. Therefore, the systemic toxicity observed in the studies are questionable.

4.10.1.3 Carcinogenicity: dermal

No relevant information available

4.10.2 Human information

No relevant information available

4.10.3 Other relevant information

There are few reports on chronic exposure studies with GMA, but each one has significant methodological deficiencies (low doses, few animals and low GMA purity) which make their results questionable. Consequently, a read-across for GMA was used with the rationale that although the kinetics of carboxylesterase-mediated hydrolysis of GMA appear to be species dependent, the primary metabolite of GMA found in humans, rats and rabbits is glycidol. Chronic bioassay data were located for glycidol in rats and mice.

Table 23a: Summary table of relevance for carcinogenic assessment of GMA

Method	Results	Remarks	Reference
rat (Fischer 344) male/female: 0, 37.5 and 75 mg/kg/d (nominal) Mouse (B6C3F1) male/female: 0, 25 or 50 mg/kg bw/day (nominal) 50 animals/sex/group oral: gavage Vehicle: water Exposure: 103 weeks (5 days per week)	Positive. An increase in tumors at multiple sites in both sexes of both species.	1 (reliable without restrictions) read-across from supporting substance (structural analogue or surrogate) Test material: 2,3-epoxypropan-1-ol (glycidol) purity 94% See endpoint summary for justification of read-across Form: liquid	Irwin et al. (1990) IARC (2000)

The results of studies on carcinogenicity after oral administration of glycidol are summarized as follows:

Groups of 50 male and 50 female Fischer 344 rats, eight weeks of age, were administered 0, 37.5 or 75 mg/kg bw of glycidol (purity, 94%) in distilled water by gavage on five days per week for 103 weeks. Survival of rats was significantly lower in the treated groups in both males and females than in the control groups, with the mean survival being 92, 82 and 66 weeks for the control, mid- and high-dose males, respectively, and 97, 85 and 78 weeks for the female dose groups. As shown in Table 3 of the IARC monograph, respectively in the main table in the abstract of the NTP report (Irwin, 1990), there was a statistically significant increase in the incidence of mesotheliomas of the tunica vaginalis/peritoneum in males at both 37.5 and 75 mg/kg bw. There was a statistically significant increase in the incidence of fibroadenoma and adenocarcinoma of the mammary gland in female rats, and of mammary fibroadenoma in male rats at both doses in each case. The incidences in the forestomach of squamous-cell papilloma and of squamous papilloma or carcinoma combined were significantly increased in female rats at both doses and in male rats at the high dose. Gliomas of the brain were significantly increased in both sexes at the high dose. Other tumour types with increased incidence included squamous-cell papillomas or carcinomas of the mouth or tongue, adenomas or carcinomas of the clitoral gland and leukaemia in female rats; and Zymbal gland carcinomas, thyroid follicular-cell adenomas or carcinomas, skin tumours and adenomatous polyps or adenocarcinomas of the intestine combined in male rats. According to the

National Toxicology Program Review Panel, there was clear evidence of carcinogenicity in male and female Fischer 344 rats. (Irwin, 1990; IARC, 2000).

Table 23b: Tumor incidence at each dose level of Glycidol (Irwin, 1990)

NEOPLASMS ASSOCIATED WITH THE TWO-YEAR GAVAGE ADMINISTRATION OF GLYCIDOL (a)

Site/Neoplasm	Male			Female		
	Veh. Control	37.5 mg/kg	75 mg/kg	Veh. Control	37.5 mg/kg	75 mg/kg
RATS						
Tunica vaginalis/peritoneum						
Mesothelioma	3/49	34/50	39/47			
Mammary gland						
Fibroadenoma	3/45	8/39	7/17	14/49	32/46	29/44
Adenocarcinoma				1/50	11/48	16/48
Brain						
Glioma	0/46	5/50	6/30	0/49	4/46	4/46
Oral mucosa						
Papilloma or carcinoma				1/46	3/37	7/26
Forestomach						
Papilloma or carcinoma	1/46	2/50	6/32	0/47	4/38	11/30
Intestine						
Adenomatous polyp or adenocarcinoma	0/47	1/50	4/37			
Skin						
Sebaceous gland adenoma, basal cell tumor, or sebaceous gland adenocarcinoma	0/45	5/41	4/18			
Zymbal gland						
Carcinoma	1/49	3/50	6/48			
Clitoral gland						
Adenoma, adenocarcinoma, or carcinoma				5/49	9/47	12/45
Thyroid gland						
Follicular cell adenoma or carcinoma	1/46	4/42	6/19	0/49	1/38	3/35
Hematopoietic system						
Leukemia				13/49	14/44	20/41
MICE						
	Veh. Control	25 mg/kg	50 mg/kg	Veh. Control	25 mg/kg	50 mg/kg
Harderian gland (b)						
Adenoma or adenocarcinoma	8/46	12/41	22/44	4/46	11/43	17/43
Mammary gland						
Adenoma, fibroadenoma, or adenocarcinoma				2/50	6/50	15/50
Forestomach						
Squamous cell papilloma or carcinoma	1/50	2/50	10/50			
Uterus						
Carcinoma or adenocarcinoma				0/50	3/50	3/50
Subcutaneous tissue						
Sarcoma or fibrosarcoma				0/50	3/50	9/50
Skin						
Squamous cell papilloma or carcinoma	0/50	0/50	4/50	0/50	0/50	2/50
Liver						
Adenoma or carcinoma	24/50	31/50	35/50			
Lung						
Alveolar/bronchiolar adenoma or carcinoma	13/50	11/50	21/50			

(a) A blank space indicates that the tumor incidence at that site and in that sex was not increased by chemical exposure. Tumor incidence is expressed as the number of tumor-bearing animals divided by the number of animals alive in each group at the time the first tumor was observed in any of the three groups.

(b) The denominators for the incidence of harderian gland tumors are the actual number of harderian glands available for microscopic examination.

Groups of 50 male and 50 female B6C3F1 mice, nine weeks of age, were administered 0, 25 or 50 mg/kg bw of glycidol (94% purity, with the primary impurity, as determined by gas chromatography, being diglycidyl ether at a concentration of 2.8%, and 3-methoxy-1,2-propanediol (1.2%), 2,6-dimethanol-1,4-dioxane (1.1%), 3-chloro-1,2-propanediol (0.4%) and methanol (0.1%) as lesser impurities) in distilled water by gavage on five days per week for 103 weeks. The survival of female mice at the high dose was significantly lower after week 101 than in the controls. As shown in Table 2 of the IARC monograph, there was a significantly increased incidence of

Harderian gland adenomas and adenocarcinomas combined in the high-dose males and in the high- and mid-dose females, and of Harderian gland adenocarcinomas in the high-dose males. In male mice only, the incidences of adenomas and carcinomas of the liver, squamous-cell papillomas of the forestomach and skin and alveolar/bronchiolar adenomas of the lung were significantly increased at the high dose; in females only, the incidences of mammary gland adenocarcinomas and of subcutaneous sarcomas and fibrosarcomas combined were significantly increased at the high dose. There was also a slight increase in uterine glandular carcinomas in female mice (Irwin, 1990; IARC, 2000).

Glycidol has a harmonised classification as Carc 1B (entry 603-063-00-8).

4.10.4 Summary and discussion of carcinogenicity

There are reports on chronic exposure studies with GMA, but each one has significant methodological deficiencies such that the conclusion is that there are no acceptable chronic studies with GMA. Consequently a Read Across for GMA was used. Rationale: although the kinetics of carboxylesterase-mediated hydrolysis of GMA appear to be species dependent, the primary metabolite of GMA found in humans, rats and rabbits is glycidol. Therefore, glycidol is the breakdown product of GMA as required for read-across according to REACH Annex XI paragraph 1.5 option (2). Read-across is also supported by the comparable effects on fertility of both substances. Chronic bioassay data were located for glycidol in rats and mice. These were clearly positive and have resulted in a harmonised classification of glycidol as Carc. 1B. Therefore, the comparable induction of tumors is also expected for GMA.

4.10.5 Comparison with criteria

No data in humans exist and limited animal studies, but the study results from a read-across to glycidol which is the main (if not the only) metabolite of GMA clearly support a category 1B classification for which the requirements are (quote from the CLP Regulation):

3.6.2.2. *‘A substance that has not been tested for carcinogenicity may in certain instances be classified in Category 1A, Category 1B or Category 2 based on tumour data from a structural analogue together with substantial support from consideration of other important factors such as formation of common significant metabolites, e.g. for benzidine congener dyes.’*

This classification is further supported by the *in vivo* mutagenicity to somatic cells as shown in chapter 4.9.

4.10.6 Conclusions on classification and labelling

According to the available studies on glycidol, GMA must be classified according to CLP as Carcinogenic Category 1B (H350).

4.11 Toxicity for reproduction

4.11.1 Effects on fertility

4.11.1.1 Non-human information

Table 24a: Summary table of relevant reproductive toxicity studies (fertility)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
<p>rat (Crj: CD(SD))</p> <p>12 male/12 female</p> <p>oral: gavage</p> <p>10, 30, 100 mg/kg/day (nominal in diet)</p> <p>Vehicle: corn oil</p> <p>Exposure: 40-47 days (daily)</p> <p>OECD Guideline 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)</p>	<p>NOAEL (P): 30 mg/kg bw/day (nominal) (male/female) (At 100 mg/k the fertility index (number of delivered animals/ number of mated animals) dropped to 16.7 %, compared to 81.8 %, 100 % and 91.7 % at 0, 10 and 30 mg/kg, respectively. There were no effects on the oestrous cycle, copulation index, or gestation length. No significant changes in the numbers of corpora lutea, implants, pups born and live pups as well as the implantation and delivery indices were observed. There were no significant differences in the gestation index, live birth index or viability index on day 4.</p> <p>Histopathological analysis of the gonads showed no significant effect. No change in the number of gonocyte per Sertoli cell was observed in epithelium of seminiferous tubules (stage VIII) of all survival males at 100 mg/kg.)</p> <p>NOEL (F1): 100 mg/kg bw/day (nominal) (male/female) (Toxicity to offspring:</p> <p>No abnormalities were noted in the body weights of live pups or on necropsy of pups of any treated group).</p>	<p>1 (reliable without restriction)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA purity 99.93%</p>	<p>Ministry of Health and Welfare, Japan (1997)</p> <p>OECD (1999)</p>
<p>5 mouse male (CD-1)/group</p> <p>sperm abnormality test</p> <p>intraperitoneal</p> <p>0, 1, 5, 25 and 100 mg/kg/day</p> <p>Exposure: Daily for 5 consecutive days</p> <p>Necropsy was performed on Day 36</p>	<p>NOEL (P): 5 mg/kg bw/day (nominal) (male) (At 100 mg/kg mice had decreased caudal epididymal weights, slightly lower testicular weights, decreased sperm counts and increased abnormal sperm. Mice given 25 mg/kg/day showed decreased sperm counts and increased abnormal sperm. The NOAEL for spermatotoxicity was 5 mg/kg/day).</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Vedula (1994)</p> <p>OECD (1999)</p>

Method	Results	Remarks	Reference
5 male mice (Kunming hybrid)/group Sperm abnormality test Intraperitoneal 0, 25, 50, 100 mg/kg Exposure: Daily for 5 consecutive days Necropsy was performed on Day 36	LOAEL (P): 25 mg/kg (male) (At 25 mg/kg mice, the number of sperm cells decreased and there was an increase in sperm abnormalities) There was no NOAEL in this study.	2 (reliable with restrictions) supporting study experimental result Test material: GMA	Xie (1990b)

Oral toxicity study was performed in SD (Crj: CD) rats by an OECD combined repeated dose and reproductive/developmental toxicity screening test (OECD TG 422). Administration was conducted by gavage at doses of 10, 30 and 100 mg/kg/day from 14 days before mating to 14 days after mating in males and from 14 days before mating to day 3 of lactation in females (Ministry of Health and Welfare, Japan: 1997).

Maternal toxicity was mainly limited to effects on the stomach at all dose levels due to the local irritating/corrosive properties of GMA (see 4.7). Many animals were infertile apparent in the 100 mg/kg group, however, morphological abnormalities were not apparent in the epididymis, seminal vesicles, prostate, uterus, or pituitary gland. Moreover, counts of Stage VIII seminiferous tubules in the testes of the 100 mg/kg group did not reveal any effects attributed to GMA exposure. The fertility index decreased significantly in the 100 mg/kg group, presumably due to the low sperm mobility. No sperm analysis was performed in the original investigation as this is not required according to OECD 422. Secondary investigations showed reduced motility in sperm but no further details were provided in the report. There were no effects on the estrous cycle, copulation index, gestation length or parturition. Slight decreases in the numbers of corpora lutea, implants, pups born and live pups as well as the implantation and delivery indices were observed in the 100 mg/kg group. However, clear effects attributable to the administration of GMA could not be concluded due to the few cases. There were no significant differences in gestation index, liver birth index or viability index on day 4. No abnormalities attributable to GMA were noted in body weights of live pups or on necropsy of pups in any GMA-treated group. Therefore, NOAELs for reproductive performance of parents (males and females) and pup development were considered to be 30 mg/kg/day and 100 mg/kg/day, respectively (Ministry of Health and Welfare, Japan: 1997).

Table 24b: Copulation and fertility results in rats (Ministry of Health and Welfare, 1997)

Dose level (mg/kg)	0	10	30	100
No. of pairs mated	11	12	12	12
No. of pairs copulated	11	12	12	12
No. of pregnant females	9	12	11	2
Copulation index (%) 1)	100.0	100.0	100.0	100.0
Fertility index (%) 2)	81.8	100.0	91.7	16.7**
Estrus cycle (days) (Mean ± S.D.)	4.5 ± 0.5 (12)	4.5 ± 0.6 (12)	4.4 ± 0.5 (12)	4.6 ± 0.7 (12)

1) (No. of animals with successful copulation / no. of animals mated) x 100
 2) (No. of pregnant animals / no. of animals with successful copulation) x 100
 Values in parentheses are expressed no. of animals observed
 Significant difference from control group; ** : p ≤ 0.01

Table 24c: Reproductive and foetal findings in rats in rats (Ministry of Health and Welfare, 1997)

Dose level (mg/kg)	0	10	30	100
No. of dams observed	10	12	11	2
No. of dams delivered live pups	10	12	11	2
Duration of gestation (Mean ± S.D.)	22.6 ± 0.5	22.8 ± 0.4	22.7 ± 0.6	23.0 ± 0.0
No. of total corpora lutea (Mean ± S.D.)	195(19.5 ± 3.6)	233(19.4 ± 2.4)	212(19.3 ± 2.2)	31(15.5 ± 0.7)
No. of total implants (Mean ± S.D.)	166(16.6 ± 2.7)	200(16.7 ± 1.9)	184(16.7 ± 1.6)	24(12.0 ± 2.8)
No. of total pups born (Mean ± S.D.)	152(15.2 ± 3.3)	176(14.7 ± 2.6)	160(14.5 ± 1.6)	18(9.0 ± 1.4)
No. of total live pups born (Mean ± S.D.)	151(15.1 ± 3.3)	174(14.5 ± 2.9)	160(14.5 ± 1.6)	18(9.0 ± 1.4)
Male	66(6.6 ± 2.5) a)	85(7.1 ± 3.0)	79(7.2 ± 2.5) a)	9(4.5 ± 0.7)
Female	85(8.5 ± 2.7) a)	89(7.4 ± 2.5) a)	81(7.4 ± 2.5)	9(4.5 ± 2.1)
Sex ratio (Mean ± S.D.)	0.88 ± 0.46	1.10 ± 0.57	1.15 ± 0.65	1.17 ± 0.71
No. of total live pups on day 4 (Mean ± S.D.)				
Male	61(6.1 ± 2.6)	82(6.8 ± 2.7)	76(6.9 ± 2.5)	9(4.5 ± 0.7)
Female	81(8.1 ± 2.4)	83(6.9 ± 2.8)	77(7.0 ± 2.8)	8(4.0 ± 1.4)
No. of total dead pups born (Mean ± S.D.)	1(0.1 ± 0.3)	2(0.2 ± 0.6)	0(0.0 ± 0.0)	0(0.0 ± 0.0)
stillbirth	0(0.0 ± 0.0)	0(0.0 ± 0.0)	0(0.0 ± 0.0)	0(0.0 ± 0.0)
cannibalism	1(0.1 ± 0.3)	2(0.2 ± 0.6)	0(0.0 ± 0.0)	0(0.0 ± 0.0)
Gestation index (%) 1)	100.0	100.0	100.0	100.0
Implantation index (% Mean ± S.D.) 2)	86.0 ± 12.1	86.5 ± 10.2	87.5 ± 10.0	77.9 ± 21.8
Delivery index (% Mean ± S.D.) 3)	91.0 ± 7.8	88.0 ± 11.3	87.0 ± 5.0	75.7 ± 6.1
Live birth index (% Mean ± S.D.) 4)	99.3 ± 2.1	98.5 ± 5.2	100.0 ± 0.0	100.0 ± 0.0
Viability index on day 4 (% Mean ± S.D.) 5)				
Male	90.2 ± 17.8	97.6 ± 4.5	96.3 ± 6.4	100.0 ± 0.0
Female	96.1 ± 5.1	92.3 ± 13.7	93.5 ± 12.9	91.7 ± 11.8

1) (No. of females with live pups / no. of pregnant females) x 100
 2) (No. of implants / no. of corpora lutea) x 100
 3) (No. of pups born / no. of implants) x 100
 4) (No. of live pups born / no. of pups born) x 100
 5) (No. of live pups on day 4 after birth / no. of live pups born) x 100
 a) Includes live pups died before observations

Male mice injected i.p. with 5 consecutive daily doses of 0, 25, 50 or 100 mg/kg/day showed an increase in the percentage of abnormal sperm and decrease in the number of sperm (Table 24d) (Xie *et al.* 1990b).

Induction of Sperm Abnormalities in Mice by GMA

Male mice of Kunming hybrid, 11–15 weeks old, were used. GMA was dissolved in maize oil and administered to the animals by five consecutive daily ip injections of 25, 50, and 100 mg/kg body wt. Groups of five mice were used in each dose level of the chemical. The negative control group received only maize oil, while the positive control group received ip injections of cyclophosphamide (40 mg/kg body wt) daily for 5 days. The mice were killed by cervical dislocation 35 days after the first injection. The caudal epididymides were removed. Sperm suspensions, smears, and slides were made and sperm abnormalities were evaluated according to Wyrobek and Bruce (1975). The morphological abnormalities included sperm with a banana-like form, without the usual hook, swollen head, or amorphous.

Table 24d: Sperm abnormalities of male mice induced by GMA (Xie et al. 1990b)

Sperm Abnormality of Male Mice Induced by GMA			
Group	Dose (mg/kg)	Sperm abnormality (%)	No. of sperm × 10 ⁶ ($\bar{X} \pm SD$)
Control	0	6.15	1.48 ± 0.11
GMA	25	16*	1.08 ± 0.15*
	50	18*	0.66 ± 0.08*
	100	17*	0.66 ± 0.08*
CP	200	25*	0.72 ± 0.08*

* Control vs GMA and CP groups, *P* < 0.01.

These results were confirmed in a subsequent study where mice were dosed i.p. with 0, 1, 5, 25 or 100 mg/kg for five consecutive days and on day 36 complete necropsy was performed (Vedula *et al.*, 1994). At 100 mg/kg, mice had decreased caudal epididymal weights and slightly lower testicular weights (Table 24e), decreased sperm counts (Table 24g) and increased abnormal sperm (Table 24h). Mice given 25 mg/kg/day showed decreased sperm counts and increased abnormal sperm.

Table 24e: Summary of male reproductive organ weights (Vedula et al. 1994)

ORGAN AND ORGAN/BODY WEIGHTS - MALES

DOSE MG/KG/DAY		FINAL BODY WT. (G)	EPIDIDYMIS		TESTES		CAUDA	
			(G)	(G/100)	(G)	(G/100)	(G)	(G/100)
0	MEAN	38.6	0.111	0.286	0.258	0.668	0.034	0.087
	S.D.	1.4	0.010	0.021	0.022	0.059	0.003	0.007
	N=	5	5	5	5	5	5	5
1	MEAN	36.4	0.109	0.300	0.254	0.701	0.034	0.092
	S.D.	0.8	0.014	0.035	0.039	0.116	0.005	0.012
	N=	4	4	4	4	4	4	4
5	MEAN	37.5	0.098	0.262	0.243	0.648	0.030	0.080
	S.D.	1.3	0.011	0.026	0.011	0.042	0.006	0.015
	N=	5	5	5	5	5	5	5
25	MEAN	37.8	0.103	0.273	0.243	0.644	0.030	0.079
	S.D.	1.3	0.009	0.022	0.030	0.073	0.006	0.017
	N=	5	5	5	5	5	5	5
100	MEAN	38.6	0.111	0.292	0.212	0.550	0.026\$	0.068\$
	S.D.	1.9	0.030	0.095	0.035	0.097	0.004	0.009
	N=	5	5	5	5	5	5	5
40 (CP)	MEAN	36.5	0.095	0.259	0.228	0.626	0.027\$	0.075
	S.D.	2.3	0.009	0.008	0.035	0.091	0.002	0.007
	N=	5	5	5	5	5	5	5

\$ STATISTICALLY DIFFERENT FROM CONTROL MEAN BY WILCOXON'S TEST, ALPHA = 0.05.

Table 24f: Summary of gross pathologic observations in male reproductive organs (Vedula et al. 1994)

SEX	DOSE IN MG/KG/DAY	NUMBER OF MICE EXAMINED	GROSS PATHOLOGIC OBSERVATIONS#					
			MALES					
			0	1	5	25	100	40\$
TESTES								
WITHIN NORMAL LIMITS.			4	4	4	5	4	4
CYST, UNILATERAL:			0	0	1	0	0	0
DECREASED SIZE, UNILATERAL:			1	0	1	0	1	1
FOCUS - ELEVATED, UNILATERAL:			0	0	0	0	0	1

Table 24g: Summary of sperm count effects induced by GMA (Vedula et al. 1994)

CAUDAL EPIDIDYMAL SPERM COUNT

DOSE MG/KG/DAY		TOTAL SPERM (X 10 ⁶)	SPERM COUNT/G CAUDA EPIDIDYMIS (X 10 ⁹)
0	MEAN	54.2	1.63
	S.D.	12.4	0.44
	N=	5	5
1	MEAN	65.4	1.98
	S.D.	6.2	0.29
	N=	4	4
5	MEAN	48.9	1.59
	S.D.	19.7	0.52
	N=	5	5
25	MEAN	39.7	1.37
	S.D.	6.5	0.32
	N=	5	5
100	MEAN	29.8§	1.11
	S.D.	8.4	0.22
	N=	5	5
40 (CP)	MEAN	33.8	1.23
	S.D.	11.5	0.37
	N=	5	5

§ STATISTICALLY DIFFERENT FROM CONTROL MEAN BY WILCOXON'S TEST,
ALPHA = 0.05.

Table 24h: Summary of abnormal sperm effects induced by GMA (Vedula et al. 1994)

CAUDAL EPIDIDYMAL SPERM MORPHOLOGY

DOSE MG/KG/DAY		% ABNORMAL
0	MEAN	4.6
	S.D.	1.9
	N=	5
1	MEAN	4.3
	S.D.	1.7
	N=	4
5	MEAN	5.4
	S.D.	1.1
	N=	5
25	MEAN	7.0
	S.D.	1.5
	N=	5
100	MEAN	9.6*
	S.D.	3.7
	N=	5
40	MEAN	14.0*
	S.D.	4.4
	N=	5

* STATISTICALLY DIFFERENT FROM CONTROL MEAN BY DUNNETT'S TEST, ALPHA = 0.05.

These results might support the decreased fertility index of rat study at 100 mg/kg/day.

4.11.1.2 Human information

No relevant information available

4.11.2 Developmental toxicity

4.11.2.1 Non-human information

Table 24i: Summary table of relevant reproductive toxicity studies (developmental toxicity)

Method	Results	Remarks	Reference
rat (Wistar) oral: gavage 5.38, 10.76, 21.52 and 108.0 mg/kg/day Exposure: Day 5 to day 15 of gestation (Daily)	NOAEL (maternal toxicity): 21.52 mg/kg bw/day (nominal). NOAEL (teratogenicity): 108 mg/kg bw/day (nominal). Pregnancy/litter data: At 108.0 mg/kg, a statistically significant increase in the fetal resorption rate (12.7 %,	2 (reliable with restrictions) key study experimental result Test material: GMA purity 92%	Ou-Yang (1988) OECD (1999)

CLH REPORT FOR 2,3-EPOXYPROPYL METHACRYLATE (GLYCIDYL METHACRYLATE)

Method	Results	Remarks	Reference
no data	<p>compared to 5.18 % of control group)</p> <p>Foetal data: No external, skeletal or organ abnormalities.</p> <p>No significant difference in body weight from the control.</p> <p>Developmental toxicity in rats by oral administration is not observed at the highest dose, 108 mg/kg/day which induces maternal toxicity.</p>		
<p>rabbit (New Zealand White)</p> <p>inhalation: vapour</p> <p>0.5, 2, and 10 ppm (2.91, 11.6 and 58.2 mg/m³. Calculated daily dose: 0.31, 1.22 and 6.11 mg/kg/day (nominal conc.)</p> <p>Exposure: Day 7 to 19 of gestation (7 hours/day, daily)</p> <p>yes</p>	<p>NOAEL (maternal toxicity): 2.91 mg/m³ air (nominal) (The principal indication of maternal toxicity was inflammation of the nasal olfactory and respiratory epithelium at the 11.6 and 58.2 mg/m³.)</p> <p>NOAEL (teratogenicity): 58.2 mg/m³ air (nominal) (There was no teratogenic effect)</p>	<p>2 (reliable with restrictions)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA purity 99.5%</p>	<p>Vedula (1996)</p> <p>OECD (1999)</p>
<p>rat (Crj: CD(SD)) male/female</p> <p>oral: gavage</p> <p>10, 30, 100 mg/kg/day (nominal in diet)</p> <p>Vehicle: corn oil</p> <p>Exposure: 40-47 days (daily)</p> <p>OECD Guideline 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)</p>	<p>NOEL (F1): 100 mg/kg bw/day (nominal) (male/female) (Toxicity to offspring.</p> <p>No abnormalities were noted in the body weights of live pups or on necropsy of pups of any treated group).</p>	<p>1 (reliable without restriction)</p> <p>key study</p> <p>experimental result</p> <p>Test material: GMA purity 99.93%</p>	<p>Ministry of Health and Welfare, Japan (1997)</p> <p>OECD (1999)</p>
<p>rabbit (New Zealand White)</p> <p>inhalation: vapour</p> <p>5, 10 and 50 ppm (29.1, 58.2 and 291 mg/m³). Calculated daily dose 2.62, 5.24 and 26.2 mg/kg/day (nominal conc.)</p> <p>Exposure: From day 7 through day 19 of gestation (6 hours/day, daily)</p>	<p>LOAEL (maternal toxicity): 29.1 mg/m³ air (nominal) (At 29.1 mg/m³ histopathologic alterations of the nasal respiratory and olfactory epithelium (hyperplasia, necrosis, etc.) in all animals.</p> <p>At 58.2 mg/m³ reddened eyes, swollen eyes and mucus discharge from eyes, and wet muzzle and sneezing after</p>	<p>2 (reliable with restrictions)</p> <p>supporting study</p> <p>experimental result</p> <p>Test material: GMA</p>	<p>Vedula (1995)</p> <p>OECD (1999)</p>

Method	Results	Remarks	Reference
	<p>exposure.</p> <p>Histopathologic alterations of the nasal respiratory and olfactory epithelium (hyperplasia, necrosis, etc.) in all animals</p> <p>At 291 mg/m³ decrease in feed consumption and faecal output during the exposure period in all animals (group removed from study after third exposure).</p> <p>Laboured breathing, reddened eyes and nares, swollen eyelids, squinting, decreased activity, nasal congestion, lacrimation, dorsal extension of the head, wet muzzle, excessive sneezing after exposure and coloured nasal discharge</p> <p>Histopathologic alterations of the nasal respiratory and olfactory epithelium (hyperplasia, degeneration, etc.) in all animals.</p> <p>Pregnancy/litter data: No adverse effect on any reproductive parameters at 29.1 and 58.2 mg/m³)</p> <p>NOAEL (teratogenicity): 58.2 mg/m³ air (nominal) (Foetal data: No adverse effect on any embryo/foetal parameters at 29.1 and 58.2 mg/m³)</p>		

GMA was administered by oral gavage to rats during day 5 to day 15 of gestation at doses of 5.38, 10.76, 21.52 and 108.0 mg/kg/day. The animals were sacrificed on day 19 of pregnancy. (OuYang et al.: 1988). As maternal toxicity, there was significant decrease in body weight gain at 108.0 mg/kg. There was a statistically significant increase in the fetal resorption rate at the 108.0 mg/kg. The percentage of pups stillborn was somewhat higher than control at all dose levels (0 % for control, and 1.35 %, 7.58 %, 1.26 % and 6.03 % for treated group at 5.38, 10.76, 21.52 and 108.0 mg/kg/day, respectively). However, because this change was not dose-dependent and statistically significant change only at 10.76 mg/kg, this was not considered to be chemical-related change. Neither birth defects nor fetal abnormalities were noted in rats treated with GMA. There was also no significant difference in fetal body weight from the control. Therefore, NOAELs were considered to be 21.52 mg/kg/day for maternal toxicity and 108.0 mg/kg/day for teratogenicity.

Table 24j: Fetal resorption data as provided by OuYang et al. (1988)

表5 GMA大鼠致畸试验结果

组别	剂量 (mg/kg)	孕鼠数 (只)	平均体重增长值 (g)	平均活胎数 (只)	平均窝重 (g)	死胎率 (%)	吸收胎率 (%)
阴性对照		14	76.78	9.14	32.63	0	5.18
GMA1/100LD ₅₀	5.38	17	66.94	8.70	35.40	1.35	2.60
GMA1/50LD ₅₀	10.76	15	65.40	8.80	37.36	7.58**	3.40
GMA1/20LD ₅₀	21.52	18	68.10	8.80	33.38	1.26	1.23
GMA1/5LD ₅₀	108.0	14	47.14 *	8.30	36.93	6.03	12.70*
敌枯双	1.0	15	35.80 *	2.40**	16.50**	0	73.10**

In an oral toxicity study performed in SD (Crj: CD) rats by an OECD combined repeated dose and reproductive/developmental toxicity screening test (OECD TG 422), administration was conducted by gavage at doses of 10, 30 and 100 mg/kg/day from 14 days before mating to 14 days after mating in males and from 14 days before mating to day 3 of lactation in females. No abnormalities attributable to the administration of GMA were noted in the body weights of live pups or on necropsy of pups in any treated group. Therefore, in this study there were no teratogenic effects induced by GMA in rats with a NOAEL of 100 mg/kg/day (Ministry of Health and Welfare, Japan, 1997).

There were two inhalation tests on developmental toxicity. In the first, rabbits were exposed to GMA at concentrations of 29.1, 58.2 and 291 mg/m³ (daily intake is calculated as 2.62, 5.24 and 26.2 mg/kg/day, respectively), 6 hours/day, daily during day 7 to day 19 of gestation (Vedula et al.: 1995). Respiratory distress and decrease in feed consumption was observed at 291 mg/m³. Less severe signs of ocular and respiratory irritation consisting of reddened eyes, wet muzzle and sneezing after exposure were observed at 58.2 mg/m³. Treatment-related histopathologic alterations of the nasal tissues (hyperplasia, necrosis, etc.) were present in all animals treated with GMA. Because of respiratory distress, animals at 291 mg/m³ were removed early from study after the third exposure. Therefore, evaluation of reproductive and embryonal/fetal parameter was precluded. There was no adverse effect on any reproductive and embryo/fetal parameter at 29.1 and 58.2 mg/m³. LOAEL for maternal toxicity was 29.1 mg/m³ (2.62 mg/kg/day) and NOAEL for teratogenicity was 58.2 mg/m³ (5.24 mg/kg/day).

In the second, rabbits were exposed to GMA at concentrations of 2.91, 11.6 and 58.2 mg/m³ (daily intake 0.31, 1.22, 6.11 mg/kg/day, respectively) 7 hours/day, daily during day 7 to day 19 of gestation (Vedula et al.: 1995). The principal indication of maternal toxicity was inflammation of the nasal olfactory and respiratory epithelium at the 11.6 and 58.2 mg/m³. There was no adverse effect on any reproductive and embryo/fetal parameter at any doses. Therefore, NOAEL for maternal toxicity was 2.91 mg/m³ (0.31 mg/kg/day) and NOAEL for teratogenicity was 58.2 mg/m³ (6.11 mg/kg/day).

4.11.2.2 Human information

No relevant information available

4.11.3 Other relevant information

As shown in chapter 4.1, GMA is metabolised into glycidol. Glycidol has a harmonised classification for reproductive toxicity in category 1B with H360F indicating an effect on fertility.

Summaries of the main studies showing effects of **glycidol** on fertility are provided below and were copied from the C&L proposal of glycidol (ECBI-92/95-add.3). In addition, a summary of the 13-week study by the NTP in rats and mice is also summarized below.

Thirteen-Week Studies with glycidol: Doses for groups of 10 rats ranged from 25 to 400 mg/kg, and doses for groups of 10 mice ranged from 19 to 300 mg/kg; vehicle controls received distilled water. All rats that received 400 mg/kg died by week 2; three males and one female that received 200 mg/kg died during weeks 11-12. Final mean body weights of male rats that received 50, 100, or 200 mg/kg were 96%- 85% that of vehicle controls; final mean body weights of female rats receiving the same doses were 94%-89% that of vehicle controls. **Sperm count and sperm motility were reduced in male rats that received 100 or 200 mg/kg.** Necrosis of the cerebellum, demyelination in the medulla of the brain, tubular degeneration and/or necrosis of the kidney, lymphoid necrosis of the thymus, and testicular atrophy and or degeneration occurred in rats that received 400 mg/kg (NTP, 1990).

All mice that received 300 mg/kg died by week 2; deaths of mice that received 150 mg/kg occurred during weeks 4-8 for males and weeks 1-5 for females. Mean body weights of chemically exposed mice surviving to the end of the studies were generally 90%-94% those of vehicle controls. Sperm count and sperm motility were reduced in dosed male mice. Compound-related histopathologic lesions included demyelination of the brain in males and females that received 150 or 300 mg/kg, **testicular atrophy in males at all doses**, and renal tubular cell degeneration in male mice that received 300 mg/kg (NTP, 1990).

Table 24k: Summary table of relevant reproductive toxicity studies (fertility) with Glycidol

Species	Route	Dose	Exposure time	Observations and remarks
Rat (Sprague-Dawley)	Oral	15mg/kg	12 days	<p>In a single generation study, fertile male rats were given glycidol and at different times during treatment 2 pre-oestrous females were placed in the cage of each male. After 7 days in each case, fertility was evaluated on the basis of the number of uterine implantations.</p> <p>Within one week, the males became infertile, although this effect was reversible within one week.</p> <p>Neither libido nor ejaculation ability was adversely affected by treatment.</p> <p>Histological examination of testes, epididymides, prostate and seminal vesicles on day 12 of treatment showed no difference from controls.</p> <p>The study was only briefly reported and no comments on systemic toxicity were recorded (Hahn, 1970).</p> <p>The study provides evidence for impaired fertility in male rats.</p>
Rat (Wistar)	Oral	20, 40, 100, 200mg/kg	5 days	<p>In a single generation study, male rats were given glycidol and mated with females of proven fertility.</p> <p>At 20mg/kg there were no treatment related effects. When mating was commenced on day 3 after the first dose of 40mg/kg, preimplantation losses were 40% in that week, rising to 95% in the second week. At 100mg/kg, sterility was observed in the first 2 weeks with no effect on sperm motility or mating activity. At 200mg/kg, glycidol induced epididymal spermatoceles.</p> <p>This study was briefly reported and made no specific reference to systemic toxicity (Jackson <i>et al</i>, 1970).</p> <p>The data demonstrate impaired fertility in male rats.</p>

Rat (Wistar)	Oral	200mg/kg	1-5 days	<p>Male rats were examined histologically between 1 and 12 weeks after dosing.</p> <p>Single oral doses of glycidol had no adverse effect on male rat fertility or on histology of the testis and its ducts. After five daily doses, males were sterile for 3 weeks, although histology was normal. In one animal, a large spermatocele occupied the cauda epididymidis, and in another, a small spermatocele was seen after 5 weeks in one ductulus efferens near its origin. Normal spermatogenic activity was evident in the testes at both 5 and 8 weeks and the epididymides were filled with spermatozoa. More prolonged treatment failed to produce sustained adverse effects on fertility and histology was normal.</p> <p>The methodological details in this paper were only briefly described (Cooper <i>et al.</i>, 1974).</p> <p>The study indicates that glycidol can cause impaired fertility in male rats.</p>
Mouse (B6C3F ₁)	Oral	0, 19, 75, 150mg/kg	5 days per week for 13 weeks	<p>Group size: 10 male mice and 10 male rats. Treatment-related deaths occurred: 4/10 mice given 150 mg/kg and 3/10 rats receiving 200 mg/kg died before study completion.</p> <p>Subsequently, sperm analysis was conducted on 5 animals from each group.</p>
Rat (F344/N)		0, 25, 100, 200mg/kg		<p>In mice a reduction in sperm count was observed in treated animals, achieving statistical significance at 75 and 150mg/kg. Sperm motility, recorded on a scale of 0-4, in mice given 0, 19, 75, 150mg/kg, was respectively, 3.6, 3.2, 2.8, 1.6. A treatment related lesion of the testis (atrophy and or degeneration) was observed but failed to achieve statistical significance.</p> <p>Rats showed a reduction in sperm counts which reached statistical significance in all treatment groups. On a scale of 0-4 sperm motility at doses of 0, 25, 100 and 200mg/kg was respectively, 3.4, 3.0, 2.0, 0.2. Testicular atrophy showed a significantly greater incidence in rats given 200mg/kg (NTP, 1990).</p> <p>The data show that glycidol reduces sperm count and sperm motility in mice and rats.</p>

The effects observed with glycidol resemble the effects observed in the combined repeated dose toxicity and reproductive screening study in that effects were observed on the fertility without clear effects on the reproductive organs.

4.11.4 Summary and discussion of reproductive toxicity

In the OECD combined study (TG 422), the NOAEL for reproductive toxicity was considered to be 30 mg/kg/day, based on a decrease in the fertility index (number of delivered animals/ number of mated animals) at 100 mg/kg. No effects on the reproductive organs were observed. Comparable effects on the fertility were observed for glycidol, a metabolite of GMA, supporting the relevance of the effect in the screening study. The effects on fertility were observed in the presence of maternal toxicity which was limited to local irritation of the forestomach.

As three reliable developmental studies by two different routes, oral and inhalation, and the screening study indicated no teratogenicity even at the highest doses which showed maternal toxicity. The only concern is the significant increase in fetal resorption rate at the highest dose tested. This dose also induced maternal toxicity in the form of reduced body weight gain.

4.11.5 Comparison with criteria

No data in humans exist (a Category 1A requirement) but the study results support a category 1B classification for which the requirements are (quote from the CLP Regulation):

Presumed human reproductive toxicant:

The classification of a substance in Category 1B is largely based on data from animal studies. Such data shall provide clear evidence of an adverse effect on sexual function and fertility or on development in the absence of other toxic effects, or if occurring together with other toxic effects the adverse effect on reproduction is considered not to be a secondary non-specific consequence of other toxic effects. However, when there is mechanistic information that raises doubt about the relevance of the effect for humans, classification in Category 2 may be more appropriate.

These criteria are fulfilled because a clear and statistically significant reduction in the number of pregnant rats was observed at 100 mg/kg bw/day. At this dose level also other toxic effects were observed in the parental rats. However, these effects were limited to changes in absolute and relative kidney and adrenal weights without histopathological changes or increases in total protein and albumin in the blood and not considered adverse. Local effects in the stomach were observed due to the irritating properties of GMA. However, these effects were limited to local effects as no general effect on body weight was observed. Therefore, the reduced pregnancy rate is not considered secondary to the mainly local paternal effects.

The mechanism by which GMA induces the reduced pregnancy is not clear as no effects were observed on the reproductive organs in the male and female rats. The oral developmental study in rats shows an increase in fetal absorption at 108 mg/kg bw/day, indicating that the reduced pregnancy may be due to a developmental effect. However, an ip study showed in mice that 25 and 100 mg/kg bw/day for 5 days resulted in an effect on testes weight and sperm counts and abnormal sperm. This would indicate an effect on the sexual function. However, such effects were determined in the rat oral screening study but not observed. GMA is metabolised into glycidol which has a harmonised classification including Repr. 1B H360F. This was based on studies in which males exposed to glycidol became infertile within one week without clear effects on libido, ejaculation or male reproductive organ histology (C&L proposal for glycidol U033, 1995). The reduction in fertility was observed in studies in which only males were exposed confirming that this was an effect on fertility. The observed effects with glycidol confirm the effects on fertility without effects on the reproductive organs for GMA and justify classification for effects on fertility in category 1B.

The significant increase in fetal resorption was considered for classification for developmental toxicity. However, as this effect was not observed in the comparable OECD 422 study (same route and dose levels), only observed in the presence of maternal toxicity and the main metabolite glycidol has no classification for development, no classification is proposed.

4.11.6 Conclusions on classification and labelling

Based on the available studies classification under CLP of GMA for developmental toxicity is not required.

However based on the same studies the substance must be classified under this Regulation as Toxic to Reproduction Category 1B (H360F).

4.12 Other effects

4.12.1 Non-human information

4.12.1.1 Neurotoxicity

Table 25: Studies on neurotoxicity

Method	Results	Remarks	Reference
rat (Fischer 344) (inhalation: vapour) 0.5, 2 and 15 ppm Exposure: 13 weeks (6 hours per day, 5 days per week) 13-wk inhalation neurotoxicity study in Fisher 344 rats	NOAEL : > 15 ppm (nominal) (13-week inhalation neurotoxicity study was performed in Fischer 344 rats. At week 4, there was a low incidence of nasal discharge and enlarged nostrils at 3.9 and 12 mg/m ³ . There were no treatment-related effects in any of the other measures. There was no evidence of neurotoxic effects at any exposure level.	2 (reliable with restrictions) key study experimental result Test material: GMA	Mattsson (1996) OECD (1999)

4.12.1.2 Immunotoxicity

No relevant information available

4.12.1.3 Specific investigations: other studies

No relevant information available

4.12.1.4 Human information

No relevant information available

4.12.2 Summary and discussion

Neurotoxicity

Fischer 344 rats were exposed by inhalation to GMA at approximately 0.5, 2 or 15 ppm (2.9, 12, 87 mg/m³), 6 hours/day, 5 days/ week for 13 weeks (calculated daily dose: 0.35, 1.46, 10.59 mg/kg/day). At week 4, there was a low incidence of rat with nasal discharge and enlarged nostrils at 2 and 15 ppm. There were no other treatment-related effects. A functional observation battery (FOB) and motor activity (MA) were conducted pre-exposure and at the end of each month of exposure. In addition, the post-exposure neurotoxicity evaluation focused on evoked potential testing of the visual (FEP), auditory (ABR), somatosensory system (SEP), and caudal nerves (CNAP), and a comprehensive neuropathological examination.

There was no evidence of neurotoxic effects at any exposure level (Mattsson et al.: 1996).

4.12.3 Comparison with criteria

Based on the available studies and based on the other classifications already presented, no further classification of GMA for STOT RE is required under CLP.

4.12.4 Conclusions on classification and labelling

Based on the available studies classification under CLP of GMA is not required.

5 ENVIRONMENTAL HAZARD ASSESSMENT

Not assessed.

6 OTHER INFORMATION

No further information available

7 REFERENCES

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8 ANNEXES

None