CLH report

Proposal for Harmonised Classification and Labelling

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

International Chemical Identification:

2-methoxyethyl acrylate

EC Number: **221-499-3**

CAS Number: **3121-61-7**

Index Number: **-**

Contact details for dossier submitter:

Unit of chemical assessment, Risk Assessment Directorate, French agency for food, environmental and occupational health & safety (ANSES) on behalf of French Competent Authority

Postal adress: 94701 Maisons-Alfort cedex, France

Contact : Reach@anses.fr

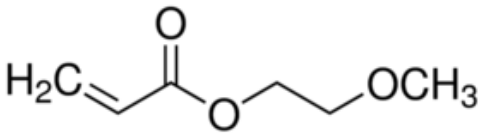
Version number: 3 **Date: February 2017**

CONTENTS

1 IDENTITY OF THE SUBSTANCE

1.1 Name and other identifiers of the substance

Table 1: Substance identity and information related to molecular and structural formula of the substance

Name(s) in the IUPAC nomenclature or other international chemical name(s)	2-methoxyethyl acrylate 2-methoxyethyl prop-2-enoate Ethylene glycol methyl ether acrylate
Other names (usual name, trade name, abbreviation)	2-MEA
ISO common name (if available and appropriate)	Not relevant
EC number (if available and appropriate)	221-499-3
EC name (if available and appropriate)	2-methoxyethyl acrylate
CAS number (if available)	3121-61-7
Other identity code (if available)	Not relevant
Molecular formula	$C_6H_{10}O_3$
Structural formula	
SMILES notation (if available)	COCCOC(=O)C=C
Molecular weight or molecular weight range	130.14 g/mol
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	Not relevant
Description of the manufacturing process and identity of the source (for UVCB substances only)	Not relevant
Degree of purity (%) (if relevant for the entry in Annex VI)	98% (w/w)

1.2 Composition of the substance

Table 2: Constituents (non-confidential information)

Constituent (Name and numerical identifier)	**Concentration range (% w/w minimum and maximum in multi-constituent substances)**	**Current CLH in Annex VI Table 3.1 (CLP)**	**Current self-classification and labelling (CLP)***
2-methoxyethyl acrylate EC no.: 221-499-3	98% (w/w)	None	See table below

*As published in ECHA website on February 2016

Classification		Number of notifiers
Hazard class and category code	Hazard statement code	
Flam. Liq. 3	H226	62
Acute Tox. 4	H302	43
Acute Tox. 3	H311	43
Acute Tox. 3	H331	41
Acute Tox. 4	H332	12
Skin Corr. 1C	H314	38
Skin Irrit. 2	H315	22
Skin Sens. 1	H317	40
Eye Dam. 1	H318	40
Eye Irrit. 2	H319	21
Repr. 1 B	H360	40
STOT RE 2	H373	29
STOT SE 3	H335	10
Aquatic Chronic 3	H412	38
Aquatic chronic 2	H411	11

Table 3: Impurities (non-confidential information) if relevant for the classification of the substance

Confidential. No impurity is considered relevant for the classification of 2-MEA.

Table 4: Additives (non-confidential information) if relevant for the classification of the substance

Additive (Name and numerical identifier)	**Function**	**Concentration range (% w/w minimum and maximum)**	**Current CLH in Annex VI Table 3.1 (CLP)**	**Current self-classification and labelling (CLP)**	**The additive contributes to the classification and labelling**
Mequinol EC no.: 205-769-8	Stabiliser	50 – 100 ppm	Acute tox 4*, H302 Skin sens 1, H317 Eye Irrit. 2 , H319	-	-

2 PROPOSED HARMONISED CLASSIFICATION AND LABELLING

2.1 Proposed harmonised classification and labelling according to the CLP criteria

Table 5:

	Index No	International Chemical Identification	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors	Notes
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Pictogram, Signal Word Code(s)	Hazard statement Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	No existing Annex VI entry										
Dossier submitters proposal	tbd	2-methoxyethyl acrylate	221-499-3	3121-61-7	Flam Liq. 3 Acute Tox. 4 Acute Tox. 3 Skin Corr. 1C Eye Dam. 1 Skin Sens. 1 Muta. 2 Repr. 1B	H226 H302 H331 H314 H318 H317 H341 H360FD	Dgr GHS 02 GHS 05 GHS 06 GHS 08	H226 H302 H331 H314 H317 H341 H360FD	EUH071		
Resulting Annex VI entry if agreed by RAC and COM	Tbd	2-methoxyethyl acrylate	221-499-3	3121-61-7							

Tbd: to be determined

Table 6: Reason for not proposing harmonised classification and status under public consultation

Hazard class	Reason for no classification	Within the scope of public consultation
Explosives	Hazard class needs not to be applied based on chemical structure of the substance	No
Flammable gases (including chemically unstable gases)	Hazard class not applicable	No
Oxidising gases	Hazard class not applicable	No
Gases under pressure	Hazard class not applicable	No
Flammable liquids		Yes
Flammable solids	Hazard class not applicable	No
Self-reactive substances	Hazard class not applicable	No
Pyrophoric liquids	Hazard class needs not to be applied based on chemical structure of the substance	No
Pyrophoric solids	Hazard class not applicable	No
Self-heating substances	Hazard class needs not to be applied based on chemical structure of the substance	No
Substances which in contact with water emit flammable gases	Hazard class needs not to be applied based on chemical structure of the substance	No
Oxidising liquids	Hazard class needs not to be applied based on chemical structure of the substance	No
Oxidising solids	Hazard class not applicable	No
Organic peroxides	Hazard class not applicable	No
Corrosive to metals	Data conclusive but not sufficient for classification	No
Acute toxicity via oral route		Yes
Acute toxicity via dermal route	Hazard class not assessed in this dossier	No
Acute toxicity via inhalation route		Yes
Skin corrosion/irritation		Yes
Serious eye damage/eye irritation		Yes
Respiratory sensitisation	Data lacking	Yes
Skin sensitisation		Yes
Germ cell mutagenicity		Yes
Carcinogenicity	Hazard class not assessed in this dossier	No
Reproductive toxicity		Yes
Specific target organ toxicity-single exposure	Hazard class not assessed in this dossier	No
Specific target organ toxicity-repeated exposure	Conclusive but not sufficient for classification	Yes
Aspiration hazard	Hazard class not assessed in this dossier	No
Hazardous to the aquatic environment	Hazard class not assessed in this dossier	No
Hazardous to the ozone layer	Hazard class not assessed in this dossier	No

3 HISTORY OF THE PREVIOUS CLASSIFICATION AND LABELLING

2-methoxyethyl acrylate (2-MEA) has not previously been assessed for harmonised classification by RAC or TC C&L.

4 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

A substance with CMR classification is normally subject to harmonised classification (Art. 36 CLP regulation). 2-methoxyethyl acrylate is currently not classified according to Annex VI of CLP. However, based on a screening developmental reproductive toxicity study, it is warranted to classify 2-methoxyethyl acrylate as Repr. 1B. Although data were insufficient for classification, respiratory sensitisation is also discussed in the dossier. Moreover, following submission of a new *in vivo* study after decision no. TPE-D-2114300801-66-01/F to investigate Germ cell mutagenicity, this endpoint has been assessed in the dossier and it is concluded that 2-MEA warrant to be classified Muta. 2.

Furthermore, differences in self classifications for acute toxicity by oral or inhalation route, skin sensitisation, skin irritation/corrosion, seirous eye damage/eye irritation and STOT RE justify the need for action at Community level since:

- Based on the local lymph node assay performed with 2-methoxyethyl acrylate, classification as Skin Sens. 1 is warranted.
- Based on available animal data, 2-methoxyethyl acrylate shall be classified for skin corrosion, serious eye damage.
- Based on the available data, classification for acute toxicity by oral and inhalation route are warranted

Physico-chemical hazards have been assessed and is thus reported in the dossier.

5 IDENTIFIED USES

The substance is manufactured and used at industrial sites only. The sectors of end-uses are: manufacture of bulk, fine chemicals, rubber, plastics products, printing and reproduction of recorded media.

6 DATA SOURCES

The data sources used for this report include the aggregated dataset of the REACH registration dossier as available on 08 January 2016. A litterature search on pubmed and science direct was conducted for relevant studies up to February 2016. Subject words were used for the literature search including “2-methoxyethyl acrylate”, “ethylene glycol monomethyl ether acrylate”.

7 PHYSICOCHEMICAL PROPERTIES

Table 7: Summary of physicochemical properties

Property	Value	Reference	Comment (e.g. measured or estimated)
Physical state at 20°C and 101,3 kPa	Colourless transparent liquid	Chemicals Evaluation and Research Institute, Japan, 2005 (Registration dossier, IUCLID 5)	Visual inspection Purity: 99.81%
Melting/freezing point	- 45°C	Chemicals Evaluation and Research Institute,	Measured OECD Guideline 102 (DSC)

Property	Value	Reference	Comment (e.g. measured or estimated)
		Japan, 2005 (Registration dossier, IUCLID 5)	Purity: 99.81%
Boiling point	164°C	Chemicals Evaluation and Research Institute, Japan, 2005 (Registration dossier, IUCLID 5)	Measured OECD Guideline 103 (Siwoloboff method) Purity: 99.81%
Relative density	1.012 g/cm^3 at 20°C	CRC Handbook of Chemistry and Physics, 86th edition, 2005 (Registration dossier, IUCLID 5)	Handbook data
Vapour pressure	399 Pa at 30°C 931 Pa at 40°C 1660 Pa at 50°C 281 Pa at 25°C	Chemicals Evaluation and Research Institute, Japan, 2005 (Registration dossier, IUCLID 5)	Measured OECD Guideline 104 (static method) Purity: 99.81% Extrapolated value OECD Guideline 104 (static method) Purity: 99.81%
Surface tension	Based on the chemical structure, surface activity is not expected.	Registration dossier, IUCLID 5	
Water solubility	144 g/L at 20°C (pH=5.3)	Chemicals Evaluation and Research Institute, Japan, 2005 (Registration dossier, IUCLID 5)	Measured OECD Guideline 105 (flask method) Purity: 99.9%
Partition coefficient n-octanol/water	Log Pow=0.9 at 25°C	Chemicals Evaluation and Research Institute, Japan, 2005 (Registration dossier, IUCLID 5)	Measured OECD Guideline 117 (HPLC method) Purity: 99.81%
Flash point	59°C at 101.3 kPa	Tremain, S.P., 2012 (Registration dossier, IUCLID 5)	Measured EU Method A.9 (closed cup method) Purity: 99.9%
Flammability	Flammable liquid	Registration dossier, IUCLID 5	Based on flash point.
Explosive properties	There are no chemical groups associated with explosive properties present in the molecule.	Registration dossier, IUCLID 5	Statement
Self-ignition temperature	246°C at 101.1 kPa	Tremain, S.P., 2012	Measured

Property	Value	Reference	Comment (e.g. measured or estimated)
		(Registration dossier, IUCLID 5)	EU Method A.15 Purity: 99.9%
Oxidising properties	On the basis of the chemical structure the substance is incapable of reacting exothermically with combustible materials.	Registration dossier, IUCLID 5	Statement
Stability in organic solvents and identity of relevant degradation products	The stability of the substance is not considered to be critical.	Registration dossier, IUCLID 5	Statement
Dissociation constant	The substance has no dissociable groups.	Registration dossier, IUCLID 5	Statement
Viscosity	Study is ongoing.		The test will be conducted after a decision on the requirement to carry out the proposed test has been taken according to the procedure laid down in Regulation (EC) 1907/2006.
Corrosive to metals	Corrosion rate: Aluminium Test Piece: max. 0.06 mm/year Steel Test Piece: max. 0.03 mm/year	Shimbori, K., 2012 (Registration dossier, IUCLID 5)	Measured UN Test C.1 (UN RTDG, Manual of Tests and Criteria, Part III, Section 37, paragraph 37.4). Purity: ≥99.9%

8 EVALUATION OF PHYSICAL HAZARDS

8.1 Explosives

Table 8: Summary table of studies on explosive properties

Method	Results	Remarks	Reference
Statement	There are no chemical groups associated with explosive properties present in the molecule. Not explosive.		Registration dossier

8.1.1 Short summary and overall relevance of the information provided on explosive properties

The assessment of explosives properties of 2-MEA is based on a statement on the chemical structure of the substance. Data provided are considered as relevant.

8.1.2 Comparison with the CLP criteria

According to CLP criteria, a substance shall not be classified as explosive when there are no chemical groups present in the molecule associated with explosive properties as given in Table A6.1 in Appendix 6 of the UN RTDG, Manual of Tests and Criteria.

8.1.3 Conclusion on classification and labelling for explosive properties

Based on chemical structure, it is considered that the substance has no explosive properties according to the CLP criteria.

8.2 Flammable gases (including chemically unstable gases)

Not relevant.

8.3 Oxidising gases

Not relevant.

8.4 Gases under pressure

Not relevant.

8.5 Flammable liquids

Table 9: Summary table of studies on flammable liquids

Method	Results	Remarks	Reference
EU Method A.9 – Flash point (closed cup method)	59°C at 101.3 kPa	Measured Purity: 99.9%	Tremain, S.P., 2012 (Registration dossier)

8.5.1 Short summary and overall relevance of the provided information on flammable liquids

The assessment of flammability of 2-MEA is based on the flash point of the substance, determined according to the EU method A.9 – Flash Point (closed-cup method). Data provided are considered as relevant.

8.5.2 Comparison with the CLP criteria

According to CLP criteria, "Flammable liquids" means a liquid having a flash point of not more than 60°C, they are classified in three categories based on their boiling point and their flash point. The substance has a flash point of 59°C which corresponds to a Category 3 flammable liquid.

8.5.3 Conclusion on classification and labelling for flammable liquids

Based on the flash point, it is concluded that the substance is classified as Category 3 Flammable liquid (H226: Flammable liquid and vapour) according to the CLP criteria.

8.6 Flammable solids

Not relevant.

8.7 Self-reactive substances

Table 10: Summary table of studies on self-reactivity

Method	Results	Remarks	Reference
Statement	Not self-reactive substance There are no chemical groups		Registration dossier

Method	Results	Remarks	Reference
	present in the molecule associated with explosive or self- reactive properties		

8.7.1 Short summary and overall relevance of the provided information on self-reactive substances

The assessment of self-reactive properties of 2-MEA is based on a statement on the chemical structure of the substance. Data provided are considered as relevant.

8.7.2 Comparison with the CLP criteria

According to CLP criteria, the classification procedure for self-reactive substance does not need to be applied when there are no chemical groups present in the molecule associated with explosive or self- reactive properties as given in Table A6.1 and A6.2 in Appendix 6 of the UN RTDG, Manual of Tests and Criteria.

8.7.3 Conclusion on classification and labelling for self-reactive substances

Based on chemical structure, it is considered that the substance has no self- reactive properties according to the CLP criteria.

8.8 Pyrophoric liquids

Table 11: Summary table of studies on pyrophoric liquids

Method	Results	Remarks	Reference
Statement	Not pyrophoric substance. Regarding the experience in handling and use, pyrophoric properties are not to be expected.		Registration dossier

8.8.1 Short summary and overall relevance of the provided information on pyrophoric liquids

The assessment of pyrophoric properties of 2-MEA is based on a statement on experience in handling and use of the substance. Data provided are considered as relevant.

8.8.2 Comparison with the CLP criteria

According to CLP criteria, the classification procedure for pyrophoric liquids does not need to be applied when experience in manufacture or handling shows that the substance or mixture does not ignite spontaneously on coming into contact with air at normal temperatures.

8.8.3 Conclusion on classification and labelling for pyrophoric liquids

Based on the experience in use, it is concluded that the substance has no pyrophoric properties according to the CLP criteria.

8.9 Pyrophoric solids

Not relevant.

8.10 Self-heating substances

Table 12: Summary table of studies on self-heating substances

Method	Results	Remarks	Reference
Statement	Not self-heating substance. As the substance is a liquid, no self-heating properties is expected.	-	Registration dossier

8.10.1 Short summary and overall relevance of the provided information on self-heating substances

The assessment of self-heating properties of 2-MEA is based on a statement on the physical state of the substance. Data provided are considered as relevant.

8.10.2 Comparison with the CLP criteria

According to CLP criteria, self-heating substances are classified in two categories following the results of the test described in Part III, Sub-section 33.3.1.6 of the UN RTDG, Manual of Tests and Criteria.

The Guidance on the Application of the CLP Criteria states that in general, the phenomenon of self-heating applies only to solids. The surface of liquids is not large enough for reaction with air and the test method is not applicable to liquids. Therefore liquids are not classified as self-heating.

Self-heating properties of liquid should be considered only if the substance is absorbed on a large surface.

8.10.3 Conclusion on classification and labelling for self-heating substances

As the substance is a liquid, it is concluded that the substance is not classified as self-heating.

8.11 Substances which in contact with water emit flammable gases

Table 13: Summary table of studies on substances which in contact with water emit flammable gases

Method	Results	Remarks	Reference
Statement	Not a substance which in contact with water emits flammable gases. Regarding the chemical structure and the experience in handling and use, the substance is not expected to emit flammable gases in contact with water.		Registration dossier

8.11.1 Short summary and overall relevance of the provided information on substances which in contact with water emit flammable gases

The assessment of flammability on contact with water of 2-MEA is based on a statement on experience in handling and use and on the chemical structure of the substance. Data provided are considered as relevant.

8.11.2 Comparison with the CLP criteria

According to CLP criteria, the classification procedure for substances which in contact with water emit flammable gases does not need to be applied when the chemical structure of the substance or mixture does

not contain metal or metalloids or experience in handling and use shows that the substance does not react with water or if the substance or mixture is known to be soluble in water to form a stable mixture.

8.11.3 Conclusion on classification and labelling for substances which in contact with water emit flammable gases

Based on chemical structure and on the experience in handling and use, it is concluded that the substance is not classified as substance which in contact with water emit flammable gases.

8.12 Oxidising liquids

Table 14: Summary table of studies on oxidising liquids

Method	Results	Remarks	Reference
Statement	Not oxidising On the basis of the chemical structure the substance is incapable of reacting exothermically with combustible materials.		Registration dossier

8.12.1 Short summary and overall relevance of the provided information on oxidising liquids

The assessment of oxidising properties of 2-MEA is based on a statement on the chemical structure of the substance. Data provided are considered as relevant.

8.12.2 Comparison with the CLP criteria

According to CLP criteria, for organic substance or mixture containing oxygen in their chemical structure, the classification for oxidizing liquids does not need to be applied if oxygen is chemically bonded only to carbon or hydrogen.

The chemical structure of 2-MEA contains oxygen which is chemically bonded only to carbon.

8.12.3 Conclusion on classification and labelling for oxidising liquids

Based on chemical structure, it is considered that the substance has no oxidising properties according to the CLP criteria.

8.13 Oxidising solids

Not relevant

8.14 Organic peroxides

Not relevant.

8.15 Corrosive to metals

Table 15: Summary table of studies on the hazard class corrosive to metals

Method	Results	Remarks	Reference

Method	Results	Remarks	Reference
UN Test C.1 (UN RTDG, Manual of Tests and Criteria, Part III, Section 37, paragraph 37.4).	Not corrosive to metal Corrosion rate: Aluminium Test Piece: max. 0.06 mm/year Steel Test Piece: max. 0.03 mm/year	Measured Purity: ≥99.9%	Shimbori, K., 2012

8.15.1 Short summary and overall relevance of the provided information on the hazard class corrosive to metals

The assessment of the hazard class corrosive to metals of 2-MEA is based on the corrosion rate on aluminium test piece and steel test piece immersed in the liquid substance at 55 °C for 7 days, following the test described in Part III, Section 37, paragraph 37.4 of the UN RTDG, Manual of Tests and Criteria. Data provided are considered as relevant.

8.15.2 Comparison with the CLP criteria

According to CLP criteria, substances of hazard class corrosive to metals are classified in a single hazard category on the basis of the outcome of the test described in Part III, Section 37, paragraph 37.4 of the UN RTDG, Manual of Tests and Criteria, if corrosion rate on either steel or aluminium surfaces exceeding 6.25 mm per year at a test temperature of 55 °C when tested on both materials.

Corrosion rate on aluminium test piece and steel test piece are max. 0.06 mm/year and max. 0.03 mm/year respectively, meaning the substance is not corrosive to metal according to CLP criteria.

8.15.3 Conclusion on classification and labelling for corrosive to metals

Based on the corrosion rate, it is concluded that the substance is not corrosive to metal.

9 TOXICOKINETICS (ABSORPTION, METABOLISM, DISTRIBUTION AND ELIMINATION)

9.1 Short summary and overall relevance of the provided toxicokinetic information on the proposed classification(s)

There are no experimental studies available in which the toxicokinetic properties of 2-methoxyethyl acrylate (2-MEA) were investigated.

2-MEA is highly soluble in water (144 g/L), has a high vapour pressure of 281 Pa at 25 °C and a molecular weight of 130.14g/mol.

The low partition coefficient (log K_{ow}) of 0.9 suggests a low potential to accumulate in biological systems. Based on the physico-chemical properties and the systemic toxicity observed in toxicity studies performed by oral and inhalation routes of administration, 2-MEA is expected to be bioavailable.

There are no experimental data available concerning the metabolism of 2-MEA.

Ester hydrolysis to acrylic acid and an alcohol has been shown to be the principal metabolic pathway of acrylates (Silver and Murphy, 1981, Millers *et al*., 1981, Ghanayem *et al*., 1987). This is the case also for methacrylate such as methylmetacrylate (Borak *et al*., 2009).

QSAR estimation using the OECD Toolbox v.3.4 Rat liver S9 metabolism simulator results in eight metabolites reported in the table 16 below.

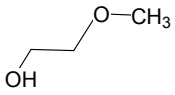
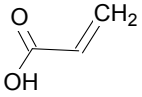
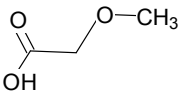
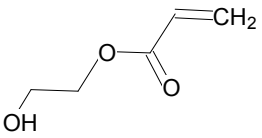
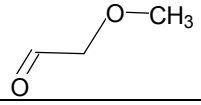
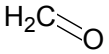
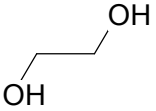
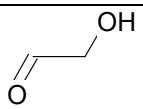
Based on the expected enzymatic cleavage of the ester bond, it is anticipated that acrylic acid and 2-methoxyethanol will be the main primary metabolites of 2-MEA.

Based on the known metabolite pathway of 2-methoxyethanol, methoxyacetic acid, methoxyacetaldehyde ethylene glycol and formaldehyde are expected to be degradation products of 2- methoxyethanol (See figure 1 below from WHO, 2009 on 2-methoxyethanol).

According to the EU RAR of 2002 on acrylic acid, acrylic acid is rapidly metabolised by oxidative pathways to carbon dioxide which is formed via acrylyl-CoA by the non-vitamin-B12-dependent pathway of mammalian propionate. About 80% of an ingested dose of acrylic acid is exhaled as carbon dioxide within 24 hours. In urine poorly characterised substances of a higher polarity than those of acrylic acid are detected. Unmetabolised acrylic acid was not found in urine. However, a small proportion of 3-hydroxypropionic acid as major urinary metabolite of absorbed acrylic acid was identified.

Based on the OECD QSAR toolbox, three other acrylates (2 unknown compounds and 2-hydroxyethyl acrylate) may also be formed.

Table 16: Summary table of predicted metabolites of 2-MEA

Simulated metabolites	**Structure**	**Harmonised classification (CMR and sensitising properties)**	**Self-classification (CMR and sensitising properties)**
2- methoxyethanol CAS no 109-86-4		Repr. 1B H360 FD	Repr. 1B, H360 FD
Acrylic acid CAS no 79-10-7		No classification for CMR or sensitising properties	No self-classification for CMR or sensitising properties
Methoxyacetic acid CAs no 625-45-6		Repr. 1B , H360 FD	Repr. 1B , H360 FD
2-hydroxyethyl acrylate CAS no 818-61-1		No classification for CMR or sensitising properties	No self-classification for CMR or sensitising properties
Methoxyacetaldehyde CAS no 10312-83-1		No Harmonised classification	Skin sens 1
Formaldehyde CAS no 50-00-0		Carc. 1B Muta 2 Resp. Sens 1 Skin sens 1A	Carc. 1B Muta 2 Resp. Sens 1 Skin sens 1A
Ethylene glycol CAS no 107-21-1		No classification for CMR or sensitising properties	No self-classification for CMR or sensitising properties
Glycollaldehyde CAS no 141-46-8		No Harmonised classification	Skin sens 1

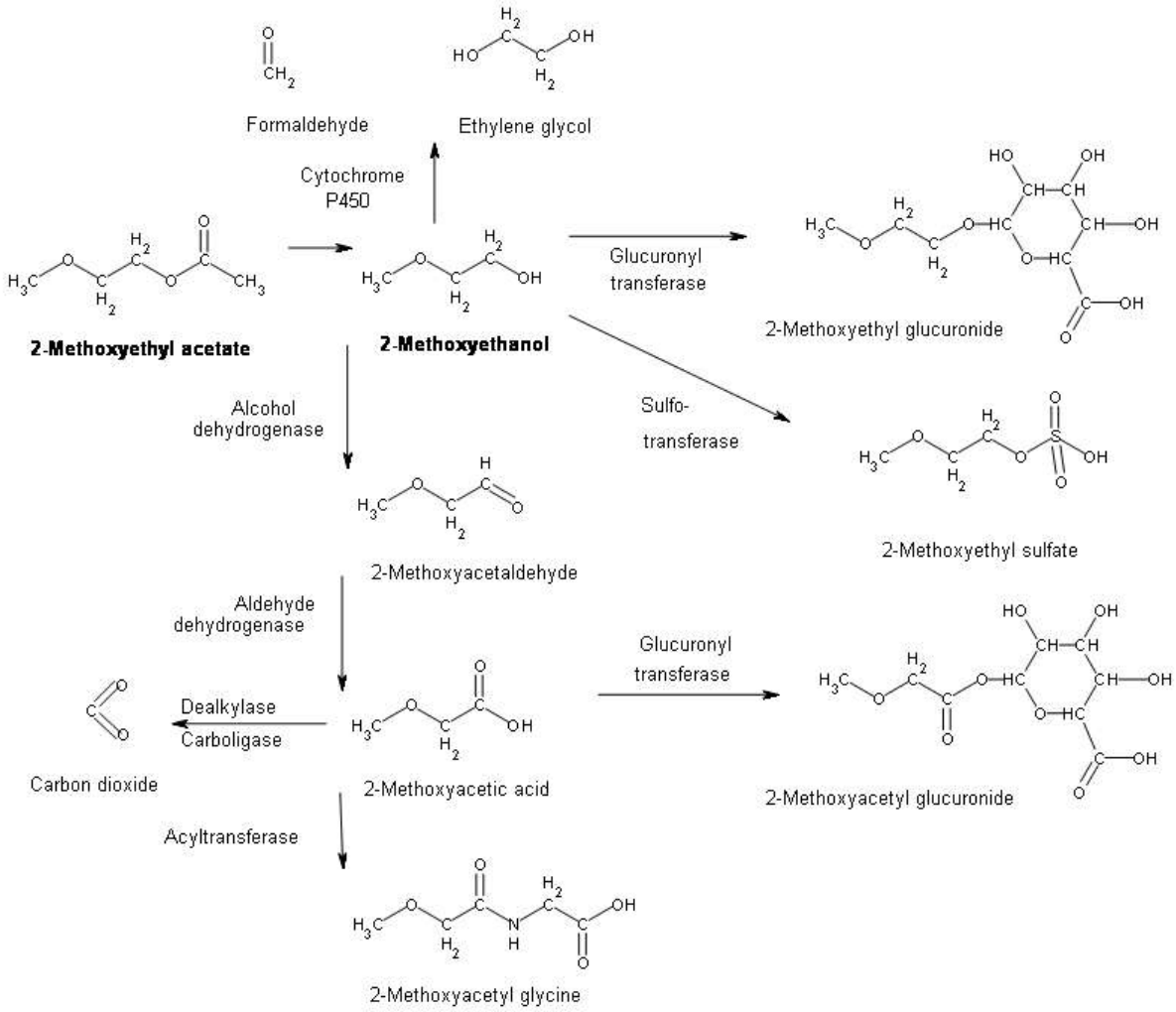


Figure 1: Metabolic pathways of 2-methoxyethanol (WHO, 2-Methoxyethanol, 2009)

10 EVALUATION OF HEALTH HAZARDS

10.1 Acute toxicity - oral route

Table 17: Summary table of animal studies on acute oral toxicity

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance	Dose levels, duration of exposure	Value LD_{50}	Reference
Equivalent to OECD 401 (Acute Oral toxicity) 2 (reliable with restriction) Oral: gavage Limitations: only dead animals were necropsied; no	SD Rats 5/sex/dose	2-MEA	Acute single exposure 252, 353.5, 505, 555.5, 606 mg/kg bw	404 mg/kg bw (95% CL =343.4-464.6)	Rhône-Poulenc Inc., 1980

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance	Dose levels, duration of exposure	Value LD_{50}	Reference
histopathology, prior to GLP					
Equivalent to OECD 401 (Acute oral toxicity) 2 (reliable with restriction) oral: gavage Limitations: No details on analytical purity of the test substance; limited details on test animals and environmental conditions; prior to GLP	Wistar male rats	2-MEA	Single exposure 505, 1010, 2020 mg/kg bw	818 mg/kg bw/d (95%CL = 596-1131)	Union Carbide Corporation study, 1968

CL: confidence limits

Detailed study summaries are available in Annex I of the CLH report.

10.1.1 Short summary and overall relevance of the provided information on acute oral toxicity

In an acute oral toxicity study, rats were administered 2-MEA via gavage (Rhône-Poulenc Inc., 1980). Five rats per sex and dose received the following dose levels: 252.5, 353.3, 505.0, 555.5, 606.0 mg/kg bw. The mortality was 0, 2, 2, 4 and 5 for males and 0, 2, 3, 4 and 5 for females, respectively, listed by increasing dose levels. Autopsy of dead animals revealed pulmonary haemorrhages. No clinical signs were noted. Based on the results, the oral LD_{50} in rats was 404 mg/kg bw.

The acute toxicity of the test substance was also assessed in a study similar to OECD 401, in which 5 male rats per group received the test substance via oral gavage at dose levels of 252.5, 1010 and 2020 mg/kg bw (Union Carbide Corporation, 1968). Mortalities were observed in 4/5 animals and 5/5 animals treated with 1010 and 2020 mg/kg bw, respectively. No mortalities were observed in animals administered the lowest dose (252.5 mg/kg bw). However, at this dose level, sluggish behaviour was observed in the animals during the 14-day observation period. In all surviving animals of the 252.5 and 1010 mg/kg bw/day, no effects on body weights were noted. At necropsy, congestion was observed in the lungs and the abdominal viscera of treated animals. Based on the probit method, the oral LD_{50} value in rats was calculated to be 818 mg/kg bw.

10.1.2 Comparison with the CLP criteria

The LD_{50} values are within the range (300-2000 mg/kg bw) established for classification as Acute tox. 4 – H302 under regulation (EC) 1272/2008 criteria.

10.1.3 Conclusion on classification and labelling for acute oral toxicity

Based on the available acute oral toxicity studies, 2-MEA needs to be classified **Acute tox. 4 "Harmful if swallowed" – H302**

10.2 Acute toxicity - dermal route

Not evaluated.

10.3 Acute toxicity - inhalation route

Table 18: Summary table of animal studies on acute inhalation toxicity

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance, form and particle size (MMAD)**	**Dose levels, duration of exposure**	**Value** LC_{50}	**Reference**
Similar to OECD 403 2 (reliable with restriction) Limitations: prior to GLP and OECD guideline, no details on analytical purity of the test substance; limited details on inhalation exposure as well as on test animals and environmental conditions	Male Wistar rats 6/group	2-MEA, no data on MMAD	Whole body exposure 4h exposure Vapour 1.4; 2.7; 5.4 mg/L	2.7 mg/L (95% CL = 1.9-3.8)	Union Carbide Corporation study, 1968

Detailed study summary is available in Annex I.

10.3.1 Short summary and overall relevance of the provided information on acute inhalation toxicity

The acute inhalation toxicity of 2-methoxyethyl acrylate was investigated in male rats using a whole body exposure system (Union Carbide Corporation, 1968). In a preliminary test, 6 animals per group were exposed to the test substance at target concentrations of 10.4, 9.6 and 8.4 mg/L for periods of 15 min, 30 min and 1 h, respectively. Since mortalities already occurred at 9.6 mg/L, concentrations used in the main study were lowered to 5.3, 2.7 and 1.3 mg/L and animals (6 per concentration) were exposed to the test substance for 4 h. At 2.7 and 5.3 mg/L, mortalities were observed between Days 1 and 3 in 3/6 and 6/6 animals, respectively. No mortality occurred in animals treated with 1.3 mg/L up to the end of the 14-day observation period. Clinical signs observed in the animals involved swollen abdomen, laboured breathing and gasping. Furthermore, irritation of the eyes, nose and extremities was noted during exposure to the test substance. Necropsy of rats dying during the study revealed slight haemorrhage of lungs and blood in intestines. In two of the three surviving rats at 2.7 mg/L areas of focal consolidation scattered throughout the lungs were observed at necropsy. All others showed nothing remarkable. Body weights in all surviving animals were not affected by treatment. Based on the results, the LC_{50} value in rats was 2.7 mg/L.

10.3.2 Comparison with the CLP criteria

The LC_{50} value for 2-MEA as vapour are in range (2-10 mg/L) for classification as Acute tox. 3 –H331 under regulation (EC) no. 1272/2008 criteria.

10.3.3 Conclusion on classification and labelling for acute inhalation toxicity

Based on the available acute inhalation toxicity study, 2-MEA is classified **Acute tox. 3 "Toxic if inhaled" – H331**

10.4 Skin corrosion/irritation

Table 19: Summary table of animal studies on skin corrosion/irritation

Method, guideline, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels duration of exposure	Results -Observations and time point of onset -Mean scores/animal -Reversibility	Reference
Equivalent to OECD 404, non GLP 4(not assignable) Deviations: 24h instead of 4h, open condition, no postexposure period, only 0.01 mL instead of 0.5 mL, only short summary available	5 Rabbits	2-MEA	0.01 mL 24-h exposure	Immediately after exposure to the test substance, very slight to slight irritation was observed in 1/5 and 4/5 animals, respectively	Union Carbide Corporation study, 1968
Equivalent to OECD 404 Non GLP 2(reliable with restriction) Deviations: 24h instead of 4h, occlusive test condition on both abraded and intact skin, only two reading points, the study was terminated at 72h	6 NZ rabbits	2-MEA	0.5mL 24-h exposure	Mean Skin irritation scores on intact skin: at 24h: Erythema: 3 Edema: 3 at 72h: Erythema: 3.17 Edema: 2.5 No differences between intact and abraded application sites	Rhône-Poulenc Inc., 1980a
Equivalent to OECD 404 Non GLP 2(reliable with restriction) Deviations: 1mL instead of 0.5 mL, only 4 and 48h readings. The study was terminated at 48h, individual scores not given	6 NZ rabbits	2-MEA	1mL 4-h exposure	No corrosive effects at 4h readings. Skin Corrosion in 5/6 animals at 48h exposure	Rhône-Poulenc Inc., 1980b

Detailed study summaries are available in Annex I of the CLH report.

10.4.1 Short summary and overall relevance of the provided information on skin corrosion/irritation

The Union Carbide Corporation study (1968) is not considered suitable for classification purposes. Indeed, only 0.01mL of test material was applied under open condition.

In the study of Rhone-Poulenc Inc (1980a), a 48-hour observation time was not included and involve a 24-hour test material exposure followed by observations at 24 hour and 72 hours. The test material was patched both on abraded and on intact skin of six rabbits. Twenty four hours instead of 4h were used under occlusive dressing condition. Pronounced responses at the 72 hours time point was observed. Reversibility of the effects was not studied.

In the skin irritation study of Rhone-Poulenc Inc (1980b), corrosive effects have been observed at 48h post-exposure but not after 4-h exposure.

10.4.2 Comparison with the CLP criteria

Visible necrosis was seen at 48h after 4-hour exposure in rabbits (Rhone-Poulenc Inc., 1980b). As the responses were observed after exposure longer than 1 hour, skin Corr. 1A and 1B are not appropriate. According to the CLP criteria 2-MEA has to be classified Skin Corr. 1C, H314.

10.4.3 Conclusion on classification and labelling for skin corrosion/irritation

2-MEA is a corrosive substance to skin and classification **Skin Corrosion category 1C, H314 "Causes severe skin burns and eye damage"** is warranted.

Due to 2-MEA high vapour pressure, 2-MEA may be inhaled and since 2-MEA is classified for skin corrosivity, the supplementary hazard statement **EUH071 " Corrosive to respiratory tract"** is considered warranted.

10.5 Serious eye damage/eye irritation

Table 20: Summary table of animal studies on serious eye damage/eye irritation

Method, guideline, Klimish score, deviations if any	**Species, strain, sex, no/group**	**Test substance,**	**Dose levels duration of exposure**	**Results** **- Observations and time point of onset** **- Mean scores/animal** **- Reversibility**	**Reference**
Similar to OECD 405 4(not assignable) Deviations: Original report not available and documentation insufficient for assessment.	Albino rabbits,	2-MEA	6 animals/dose 0.001, 0.005, 0.02, 0.1, and 0.5 mL 24h exposure	24h reading: severe corneal injury was observed in 3 eyes treated with 0.02 mL of the undiluted test substance. Minor to moderate injury was observed in the eyes after treatment with 0.005 mL of the undiluted test substance (no further details)	Union Carbide Corporation, 1968
Similar to OECD 405 2(reliable with restriction) Deviations: Study termination at day 7	NZ rabbits	2-MEA	0.1mL 6 animals Single exposure without washing or 30s exposure	Mean 24-72h score/6 animals: Conjunctivae redness: 2.67 Conjunctivae oedema: 3.88 Iris: 0.2 Cornea: 1.7 Only iris effects were fully reversible at day 7.	Rhône-Poulenc Inc., 1980c

Detailed study summaries are available in Annex I of the CLH report.

10.5.1 Short summary and overall relevance of the provided information on serious eye damage/eye irritation

The eye irritation potential of 2-methoxyethyl acrylate was investigated in New Zealand Albino rabbits according to EPA guideline 40 CFR 163.81.4 (Rhône-Poulenc Inc., 1980c). The undiluted test substance (0.1 mL) caused serious and irreversible damage to the eyes on conjuctivea and cornea. The reversibility of the effects was not shown at the end of the observation period (day-7).

The union Carbide Corporation study (1968) is not considered suitable for classification. Nevertheless, the study gives supporting evidence that the undiluted test substance (0.02 mL) caused severe corneal injury to the eyes after an exposure period of 24 h in all tested albino rabbits. Even minor to moderate injury was observed in the eyes of the animals after treatment with 0.005 mL of the undiluted test substance after 24 h.

10.5.2 Comparison with the CLP criteria

Severe eye effects were observed in conjunctivae and cornea in rabbits in the Rhône-Poulenc Inc., 1980c study. The mean scores of the 6 rabbits meet the criteria for eye irritation category 2. The reversibility of the effects were not studied until 21 day post exposure period. Nevertheless, eye scores of 3 to 4 were still observed in 5/6 rabbits after the 7 days post-exposure period in conjunctivae.
Therefore, 2-MEA is considered to cause irreversible damage to the eyes and support classification as Eye dam. 1 – H318 "Causes serious eye damage" according to the CLP criteria.

10.5.3 Conclusion on classification and labelling for serious eye damage/eye irritation

2-MEA is a severe eye irritant. As 2-MEA needs to be classified as Skin corr. 1C, the risk of severe damage to eyes is considered implicit. Therefore, the substance is classified for **Eye damage, category 1, H318 "Causes serious eye damage"** but will not be labelled for serious eye damage.

Since 2-MEA was assessed as corrosive to skin and eyes, a potential for respiratory tract irritation is considered to be very likely. According to Regulation (EC) 1272/2008, classification for corrosivity is considered to implicitly cover the potential to cause respiratory tract irritation and so the additional classification is considered to be superfluous.

10.6 Respiratory sensitisation

No specific animal or human data available on 2-MEA.

10.6.1 Short summary and overall relevance of the provided information on respiratory sensitisation

Table 21: Summary table of other studies relevant for respiratory sensitisation

Type of study/data	Test substance,	Relevant information about the study (as applicable)	Observations	Reference
SAR (structural alert)	2-MEA	OECD QSAR Toolbox v.3.4 Profiler: Respiratory sensitisation	2-MEA hit the alert : acrylates Proposed mechanism: A Michael addition mechanism has been suggested to be responsible for the ability of chemicals containing this structural alert to react with proteins in the lung.)	Enoch, S.J., et al., Development of Mechanism-Based Structural Alerts for Respiratory Sensitization Hazard Identification. Chemical Research in Toxicology, 2012. 25(11): p. 2490-2498
Danish QSAR database (Requested on February	2-MEA	(Q)SAR predicted profile for respiratory sensitisation in humans Software used are : CASE Ultra,	Leadscope predict positive results and the prediction was inside the applicability domain of the model, CASE Ultra and SciQSAR give positive prediction but the	-

Type of study/data	Test substance,	Relevant information about the study (as applicable)	Observations	Reference
2016)		Leadscope, SciQSAR	prediction was outside the applicability domain. Overall the battery of test predict positive results but outside applicability domain.	
SAR (structural alert)	2-MEA	DEREK v5.0.2	No alert flagged. This is expected as DEREK v5.0.2 does not contain respiratory sensitisation structural alerts referring to Acrylates.	-

According to the OECD QSAR database, acrylates have been suggested to be capable of reacting with proteins in the lung *via* a direct Michael addition mechanism. Leadscope also predict positive results for this substance. DEREK nexus do not predict respiratory sensitisation for 2-MEA as no structural alerts for acrylates were developed in the model.

With regard to the predicted metabolites only formaldehyde has an harmonised classification for respiratory sensitisation. Furthermore, respiratory sensitisation has not been reported with the two expected main metabolites 2-methoxyethanol or acrylic acid.

No human or animal data are available specifically on 2-MEA on respiratory sensitisation in the literature. Furthermore, since data to get a clear understanding of the sensitising properties of members within the group of acrylate are currently not available, no classification is proposed for 2-MEA.

10.6.2 Comparison with the CLP criteria

No data are available in both human and animals.

10.6.3 Conclusion on classification and labelling for respiratory sensitisation

No classification for respiratory sensitisation is warranted based on insufficient data.

10.7 Skin sensitisation

Table 22: Summary table of animal studies on skin sensitisation

Method, guideline, Klimish score, deviations if any	Species, strain, sex, no/group	Test substance,	Dose levels duration of exposure	Results	Reference
Local lymph node assay OECD 429, GLP 1(reliable without restriction)	CBA/Ca Mice 4 females/group Vehicle: acetone/olive oil 4:1	2-MEA	0, 25, 50, 100 %	Sensitising Stimulation index results: 25%: 9.20 50%: 12.84 100%: 11.55	Study report, 2012a

10.7.1 Short summary and overall relevance of the provided information on skin sensitisation

The local lymph node assay (LLNA) performed with 2-MEA was positive (SI > 3 at 25% and above). The dossier and literature do not contain human data on 2-MEA. Nevertheless, acrylates is a class of chemical known to be contact allergens.

10.7.2 Comparison with the CLP criteria

The results of local lymph node assay demonstrate the sensitising properties of 2-MEA. A classification Skin Sens 1, H317 "may cause an allergic reaction" is considered warranted since positive data are available.

The criteria for subcategorisation of skin sensitizers based on LLNA study is an estimated concentration to produce a stimulation index of 3 (EC_3) ≤ 2% for sub-category 1A and EC_3 value > 2% for sub-category 1B.

An EC_3 value could not be derived adequately as all stimulation index values exceed 3 and were not linear. Thus, a derivation of an EC_3 value may be associated with great uncertainty.

Therefore a classification Skin Sens. 1, H317 without sub-categorisation is proposed.

10.7.3 Conclusion on classification and labelling for skin sensitisation

Based on a LLNA assay, 2-MEA has to be classified as **Skin Sensitiser, Category 1, H317 "May cause an allergic skin reaction"** according to the CLP criteria.

10.8 Germ cell mutagenicity

Table 23: Summary table of mutagenicity/genotoxicity tests *in vitro*

Method, guideline, Klimish score, deviations if any	**Test substance,**	**Relevant information about the study including rationale for dose selection (as applicable)**	**Observations**	**Reference**
Bacterial reverse mutation Similar to OECD 471 2 (reliable with restriction) Limitations: - 4 strains instead of 5 recommanded -non GLP - limited data on test system and conditions - dose rationale not specified - no analytical purity, - positive controls not specified	2-MEA	*S. typhimurium* TA 100, TA 1535, TA 97, TA98 With and without rat or hamster S9mix Pre-incubation method and plate test with vapour from the test liquid	Negative with and without metabolic activation	Confidential report available in REACH registration IUCLID file, 1991
Bacterial reverse mutation OECD 471, GLP 1 (Reliable without restriction)	2-MEA	*S. typhimurium* TA 1535, TA 1537, TA 98 and TA 100 and *E. coli* WP2 uvrA Test concentrations: 5-5000 µg/plate	Negative with and without metabolic activation	Confidential report available in REACH registration IUCLID file, 2012

Method, guideline, Klimish score, deviations if any	Test substance,	Relevant information about the study including rationale for dose selection (as applicable)	Observations	Reference
		With and without rat S9 mix		
In vitro mammalian cell gene mutation OECD 476, GLP 1 (Reliable without restriction)	2-MEA	L5178Y lymphoma cells: mouse (with and without rat met. Act.) Test concentrations: 4h treatment (-S9 mix): 0.63, 1.25, 2.5, 5, 10, 20, 30, 40 µg/mL 4h treatment (+S9 mix): 20.25, 40.5, 81, 162, 324, 432, 540 and 648 µg/mL	Positive with and without metabolic activation. The increase is mainly due to small colony formation ±S9	Confidential report available in REACH registration IUCLID file, 2013
Mammalian chromosomal aberration test OECD 473, GLP 1 (Reliable without restriction)	2-MEA	Cultured peripheral human lymphocytes With and without rat S9mix Test concentrations: - 4h treatment (-S9 mix): 10, 20, 40 µg/mL - 4h treatment (+S9 mix): 320, 480, 640 µg/mL	Positive with metabolic activation. Negative without S9 (short exposure period only performed)	Confidential report available in REACH registration IUCLID file, 2013

Table 24: Summary table of mutagenicity/genotoxicity tests in mammalian somatic or germ cells *in vivo*

Method, guideline, deviations if any	Test substance	Relevant information about the study (as applicable)	Observations	Reference
In vivo alkaline comet assay OECD 489, GPL 2(reliable with restriction) Limitations: - Negative controls were slightly below historical control data but the relevance of this observation is questionable as Only very low number of animals were included in the historical control data, - No historical control data for non-glandular stomach.	2-MEA	2 single treatment within 24-h Sacrifice 4-h after final treatment Doses: 120, 240, 480 mg/kg bw Positive control: N-methyl-N-nitrosurea 7 animals/group except 5 in the positive control group Tissues: liver, non-glandular and glandular stomach Vehicle: PBS	Negative in liver. Equivocal in glandular stomach and positive in non-glandular stomach. Histopathological findings: Degenerative changes in the epithelium of the non-glandular stomach and glandular stomach was noted, with dose-related increased severity of effects in the non-glandular stomach. These are signs of cytotoxicity at the site of contact. Positive control: positive.	Confidential study report, 2016

Method, guideline, deviations if any	Test substance	Relevant information about the study (as applicable)	Observations	Reference

Detailed study summaries are available in Annex I of the CLH report.

10.8.1 Short summary and overall relevance of the provided information on germ cell mutagenicity

- *In vitro*

Two gene mutation assays in bacteria (Ames test) were conducted with 2-MEA. No increase in the mean revertant number of colonies was observed at any of the concentrations tested in both experiments with or without rat or hamster S9.

2-MEA was also tested for its potential to cause gene mutations in the mouse lymphoma assay according to OECD 476. The potential mutagenicity of the test substance on the thymidine kinase, TK +/- locus of the L5178Y mouse lymphoma cell line was investigated after 4 h exposure. The concentration range of the test substance was 0.63 to 40 µg/mL in the absence of metabolic activation and 20.25 to 648 µg/mL in the presence of metabolic activation. The test substance induced toxicologically significant dose-related increases in the mutant frequency at the TK +/- locus in L5178Y cells both with and without metabolic activation. The increases in mutant frequency observed were mainly due to small colony formation, indicating clastogenic activity resulting in structural chromosome damage. 2-MEA is, therefore, considered to be mutagenic under the conditions of the test. 2-MEA was more cytotoxic without S9 than in presence of S9 as shown by the higher tested concentrations with S9. The clastogenic potential observed in the chromosome aberration test was taken as confirmatory evidence for the mutagenicity of the test substance under *in-vitro* test conditions.

The potential of 2-MEA to induce chromosomal aberrations was tested in cultured peripheral human lymphocytes according to OECD 473. The lymphocytes were exposed to 2-MEA for 4h with or without metabolic activation followed by 20h culture in treatment-free media prior to cell harvest. The concentration range of the test substancewas 10 to 40µg/mL in the absence of S9 mix and 320 to 640 µg/mL in the presence of S9 mix. The test substance did not induce any statistically significant increases in the frequency of cells with aberrations in the absence of S9 mix (4-h exposure). In the presence of metabolic activation, the test substance induced a statistically significant increase in the frequency of cells with aberrations, at a dose level of 640 µg/mL. The test substance was therefore considered to be clastogenic to human lymphocytes under the conditions of the test. The substance appeared around 10 times more cytotoxic without S9 than in presence of S9. Nevertheless, a positive result without S9 cannot be excluded since long-term treatment (e.g. 24-h) was not performed.

Overall, 2-MEA is considered genotoxic *in vitro* with and without metabolic activation.

- *In vivo*

An *in vivo* mammalian alkaline comet assay was performed with 2-MEA on male rats, according to OECD guideline 489 and under GLP conditions. Male rats (7/dose) were administered 120, 240 and 480 mg/kg bw of the test substance for 2 consecutive days (at 0 and 24 h). The animals were sacrificed 4 hours after the second dose administration and samples of the liver, glandular stomach and non-glandular stomach tissues were taken from each animal. 1/7 animals in the high-dose group died within 24 h; no reason for the mortality was given in the report. The remaining 6/7 rats had a hunched posture for approximately 1 h after each dosing. The positive control substance produced a marked increase in the % tail intensity value in all the investigated tissues. The negative control was slightly below the historical control values observed in glandular stomach. However, only a very low number of animals were included in the historical control data (11 animals). No significant change in the percentage tail intensity in the liver tissue was observed between the treatment groups and control group. A dose-related significant increase in the mean of median percentage tail intensity in the glandular stomach tissue was noted in all dose groups, and in the mean percentage tail

intensity in the mid- and high dose group, compared with the control group respectively. However, the increase fell within the range of the historical negative control data. But the limited dataset of historical control data (only 11 animals) question the adequacy of using these values. A significant increase in the percentage tail intensity was also observed in the non-glandular stomach tissue of the mid- and high-dose groups, compared to the control group (mean percentage tail intensity and mean of median percentage tail intensity).
In this study, the results of the histopathological examination of the non-glandular stomach showed that 2-MEA had a dose-related cytotoxic effect at the site of contact: inflammation and degeneration of the glandular- and non-glandular stomach tissues in the mid- and high dose animals (See table 21 of Annex I of the CLH report). The inflammation and degeneration effects are considered to be a result of the corrosive properties of the test substance and were more severe in non-glandular stomach than in glandular stomach. However, statistically significant increase in the mean percentage tail intensity in the non-glandular stomach was already observed at the lowest dose showing only minimal concomitant histopathological findings in the non glandular-stomach. Moreover, in the non-glandular stomach, the increase in cytotoxicity was clearly dose-related at the mid and high dose level but was not correlated with an increased genotoxic response. This result suggests that the genotoxic response cannot only be explained by a cytotoxic response. Therefore, the results in non-glandular stomach are considered true intrinsic genotoxic response. Based on the results of the comet assay, the test substance 2-MEA is considered positive *in vivo* under the conditions of this test at the site of contact in non-glandular stomach.

10.8.2 Comparison with the CLP criteria

Germ cell mutagens category 1 in the CLP regulation is dedicated to "Substances known to induce heritable mutations or to be regarded as if they induce mutations in the germ cells of humans. The classification in Category 1A is based on positive evidence from human epidemiological studies.

No human data are available with 2-MEA, therefore Muta. 1A is not appropriate.

The classification in Category 1B is based on:
– "positive results from *in vivo* heritable germ cell mutagenicity tests in mammals; or
– positive results from *in vivo* somatic cell mutagenicity tests in mammals, in combination with some evidence that the substance has potential to cause mutations to germ cells. It is possible to derive this supporting evidence from mutagenicity/genotoxicity tests in germ cells *in vivo*, or by demonstrating the ability of the substance or its metabolites to interact with the genetic material of germ cells; or
– positive results from tests showing mutagenic effects in the germ cells of humans, without demonstration of transmission to progeny; for example, an increase in the frequency of aneuploidy in sperm cells of exposed people".

According to the CLP criteria 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."

In the ECHA guidance on the application of CLP criteria (v.4.1, June 2015), it is also stated that "It is also warranted that where there is evidence of only somatic cell genotoxicity, substances are classified in cat. 2. This holds true especially for those genotoxicants which are incapable of causing heritable mutations because they cannot reach the germ cells (e.g. genotoxicants only acting locally, 'site of contact' genotoxicants). **This means that if positive results *in vitro* are supported by at least one positive local *in vivo*, somatic cell test, such an effect should be considered as enough evidence to lead to classification in Category 2.**"

The equivocal and positive results obtained in glandular and non-glandular stomach, respectively, give evidence that 2-MEA may react at the site of contact at all doses tested and induce local genotoxicity. As

human do not have a forestomach, the extrapolation to humans may be questionable. However, humans have comparable squamous epithelial tissues in the oral cavity and the upper two-thirds of the oesophagus (CLP guidance 2015, page 375). Therefore, the substance is considered to have genotoxic potential that can be evidenced in humans at the route of entry.

There is neither *in vivo* heritable germ cell mutagenicity test nor tests in human germ cells available with 2-MEA. Some evidence are available on the ability of 2-MEA or most probably its metabolites (e.g. 2-ethoxyexanol is classify Repr. 1B, H360FD) to interact with the genetic material of germ cells as effects on the spermatogenesis were observed in the combined repeated dose toxicity study with reproduction /developmental toxicity screening test (OECD 422) (Study report, 2012b). Detoxification with regard to the genotoxic potential of the substance may occur in liver as shown by the negative result in this organ from the comet assay *in vivo* and the decreased cytotoxicity in presence of metabolic activation *in vitro*. But, with regard to the genotoxicity potential of the test substance, this is not supported by the *in vitro* assays as positive results were observed with and without metabolic activation in the MLA. In addition, the test substance was positive with S9 and negative without S9 in the Mammalian chromosomal aberration test. However, a positive result without S9 cannot be excluded since long-term treatment according to OECD guideline was not performed.

Overall, 2-MEA fulfils the criteria for category 2. Positive local *in vivo* genotoxic response was supported by the positive *in vitro* gene mutation assay and *in vitro* chromosomal aberration assay.

Due to the absence of mutagenicity test on germ cells, a category 1B cannot be judged adequate at this time. Therefore, further mutagenicity test on germ cells would be need to conclude if category 1B is fulfilled

10.8.3 Conclusion on classification and labelling for germ cell mutagenicity

Category 2 for germ cell mutagenicity is warranted based on the positive *in vivo* data on somatic cells supported by the *in vitro* data.

10.9 Carcinogenicity

Not evaluated. No data.

10.10 Reproductive toxicity

10.10.1 Adverse effects on sexual function and fertility

Table 25: Summary table of animal studies on adverse effects on sexual function and fertility

Method, guideline, Klimish score deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results	Reference

Method, guideline, Klimish score deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results	Reference
Combined repeated dose toxicity study with reproduction/developmental toxicity screening test OECD 422, GLP 1(reliable without restriction) Oral (gavage) Wistar rats 10/sex/group	2-MEA 0, 40, 100, 250/150 mg/kg bw (250 mg/kg bw/day: from Day 1 to 11; 150 mg/kg bw/day: from Day 12 to study termination) Males: 2 weeks prior to mating, during mating, and up to termination Females: during 2 weeks prior to mating, during mating, during post-coitum, and during at least 4 days of lactation Vehicle: propylene glycol	Parental effects: **250/150 mg/kg bw per day** 30% mortality in males (euthanised on days 2 and 8) Hunch posture, piloerection, pale and lean appearance (f/m) Red vagina or bleeding from vaginal in 2 females Bw loss (m/f) Hematology: ↓haemoglglobin, MCHC, MCH, Platelets (m+f), MCV (f), ↑prothrombin time (f) Reduced relative organ weight: thymus, prostate (m) Reduced absolute organ weight: testis, epididymides (m) Histopathology: degeneration of seminiferous tubular epithelium, edema, inflammation and enlarged ampholitic cells, impairment of the spermatogenetic cycle in testes. Sperm degeneration, atrophy and inflammation in epididymides. Hepatocellular necrosis in liver (m/f). Atrophy and haemorrhage in thymus (m/f) **100 mg/kg bw per day** 1 female died on study day 21 post-coitum Red vagina or bleeding from vaginal in 1 female Bw loss during gestation in females and reduced bw gain in males Hematology: ↓haemoglglobin, MCHC, MCH, Platelets, MCV, ↑ prothrombin time (f) Reduced relative organ weight: thymus, prostate (m) Reduced absolute organ weight: testis, epididymides (m) Histopathology: degeneration of seminiferous tubular epithelium, edema, necrosis, inflammation and enlarged ampholitic cells, impairment of the spermatogenetic cycle in testes. Sperm granuloma, degeneration, atrophy and inflammation in epididymides. Haemorrhage and apoptosis in thymus (m/f). **40 mg/kg bw per day** Histopathology: necrosis, enlarged ampholitic cells, impairment of the spermatogenetic cycle in testes. Sperm granuloma in one male in epididymides. Atrophy, haemorrhage and apoptosis in thymus. **A LOAEL for parental toxicity of 40 mg/kg bw was derived from this study.** Reproductive effects: **250/150 mg/kg bw per day** ↑precoital time ↓fertility index (20% vs 100% in control) ↓number of corporea lutea and implantation sites **100 mg/kg bw per day** ↓fertility index (90%) ↓number of corporea lutea and implantation sites **40 mg/kg bw per day**	Study report, 2012b

Method, guideline, Klimish score deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results	Reference
		↑precoital time ↑ duration of gestation **A LOAEL for reproductive toxicity of 40 mg/kg bw was derived from this study.** <u>Developmental effects</u> **250/150 mg/kg bw per day** ↓ number of live pups (at day 1): 0% vs 100% in control **100 mg/kg bw per day** ↓ number of live pups (at day 1): 0% vs 100% in control **40 mg/kg bw per day** ↓ number of live pups (at day 1): 70% vs 100% in control ↓ viability index (66.7% vs 99% in control) Sligh decrease in the bw of pups Lean and pale appearances of surviving pups Absence of milk in the stomach and blue discoloration of the snout. In addition autolysis was noted for pups found dead. **A LOAEL for developmental toxicity of 40 mg/kg bw was derived from this study.**	

10.10.2 Short summary and overall relevance of the provided information on adverse effects on sexual function and fertility

A combined oral repeated dose toxicity study with the reproduction/developmental toxicity screening test was performed with 2 -methoxyethyl acrylate (2 -MEA) according to OECD 422 (Study report, 2012b). The LOAEL for parental toxicity was 40 mg/kg bw based on histopathological changes on testis, epididymides and thymus at all dose levels. Mortality and severe bw effects occurred in parental animals at 100 mg/kg bw onward. At 100 and 250/150 mg/kg bw no live litters were observed. The LOAEL for reproductive effects was 40 mg/kg bw based on dose-related increase precoital time and reduced fertility at all dose levels. The LOAEL for developmental toxicity was 40 mg/kg bw based on decreased live litters and decrease viability index.

In addition, there are data available on effects on fertility for the expected primary metabolite 2-methoxyethanol (CAS no. 109-86-4) which showed effects in reproduction toxicity studies as observed for 2-MEA. Studies on 2-methoxyethanol with respect to effects on fertility show consistent toxicity to the male reproductive system in multiple species (mice, rats, guinea-pigs, rabbits and dogs) exposed by all routes of administration (subcutaneous, dermal, oral or inhalation) (CICAD, 2009). Effects on reproductive ability as well as reproductive organs have been observed, often from the lowest dose or concentration tested. Single or repeated oral administration of 2-methoxyethanol induced adverse effects on the testes (including weight and histopathological changes or biochemical indicators of testicular damage, such as urinary creatinine)

and/or various sperm parameters in every identified studies in which these endpoints were examined (CICAD, 2009).

10.10.3 Comparison with the CLP criteria

Reproductive toxicity category 1 in the CLP Regulation is dedicated to "substances which are known or presumed human reproductive toxicant". "Substances are classified in category 1 for reproductive toxicity when they are known to have produced an adverse effect on sexual function an fertility or when there is evidence from animal studies possibly supplemented with other information, to provide as strong presumption that the substance has the capacity to interfere with reproduction with humans. The classification of a substance is further distinguished on the basis of whether the evidence for classification is primarily from human data (category 1A) or from animal data (category 1B). "

Reproductive toxicity category 2 in the CLP Regulation is dedicated to substances which are "suspected human reproductive toxicants". "Substances are classified in category 2 for reproductive toxicity when there is some evidence from humans or experimental animals, possibly supplemented with other information, of an adverse effect on sexual function or fertility, and where the evidence is not sufficiently convincing to place the substance in category 1."

No human data were provided, therefore Repr. 1A is not appropriate.

In the reproductive/developmental screening study (Study report, 2012b), weight and histopathology effects on reproductive organs were observed (including testis, epididymis) from 40 mg/kg bw (oral gavage). In this study, fertility effects were observed at all dose levels including increase precoital time and dose-related decreased fertility index.

These effects may be considered secondary to the high parental toxicity observed at 100 and 250/150 mg/kg bw (body weight loss, mortality). However, at 40 mg/kg bw/d, no indication of marked general toxicity has been observed. Indeed, at this dose only changes in hematological parameters were observed in females (decreased MCV and MCH). The adversity of these findings is not clear as no change in haematocrit and haemoglobin was reported at this dose level.

In conclusion, the available data on reproductive toxicity present clear evidence of adverse effects on fertility. Because the effects are severe and not considered secondary to maternal or parental toxicity at the low dose level, the available data support classification for reproductive toxicity category 1B. There is no information that the effects may not be relevant to human and the quality of the study is good, therefore, category 2 according to the CLP criteria is not considered appropriate.

10.10.4 Adverse effects on development

Table 26: Summary table of animal studies on adverse effects on development

Method, guideline, Klimish score deviations if any, species, strain, sex, no/group	**Test substance, dose levels duration of exposure**	**Results**	**Reference**
Combined repeated dose toxicity study with reproduction/developmental toxicity screening test OECD 422, GLP 1(reliable without restriction) Oral (gavage)	2-MEA 0, 40, 100, 250/150 mg/kg bw (250 mg/kg bw/day: from Day 1 to 11; 150 mg/kg bw/day: from Day 12 to study termination) Males: 2 weeks prior to mating, during mating, and	See results in table 25.	Study report, 2012b

Method, guideline, Klimish score deviations if any, species, strain, sex, no/group	Test substance, dose levels duration of exposure	Results	Reference
Wistar rats 10/sex/group	up to termination Females: during 2 weeks prior to mating, during mating, during post-coitum, and during at least 4 days of lactation Vehicle: propylene glycol		
Non guideline Non-GLP 3 (unreliable) CD-1 mouse 50 mice/group Only one dose level, short treatment period, short reporting, dose above the maximum tolerable dose, pups were not examined for malformations	Oral : gavage GD6-13 (daily, 7 days/week) 0, 650 mg/kg bw Vehicle: distilled water	Maternal toxicity: 30% mortality in dams Developmental toxicity: 100% intrauterine death	Hardin et al., 1987

10.10.5 Short summary and overall relevance of the provided information on adverse effects on development

A developmental toxicity study evaluating 60 chemicals in mice including 2-methoxyethyl acrylate (2-MEA) was published by Hardin et al. (1987). Fifty pregnant mice were dosed by gavage with 650 mg/kg bw/day of the test substance on gestation days 6 -13. The mice were then permitted to deliver litters. The test substance produced 30% maternal mortality and 100% intrauterine death. Therefore, the test substance adversely affected all measures of reproductive success since no liveborn pups were recorded. Dead pups were not examined for malformations. However, it should be pointed out that maternal mortality was 30% and that the dose tested was too high to be suitable for evaluating developmental toxicity.

In the combined screening study (Confidential report, 2012b), implantation sites were only noted for nine females at 100 mg/kg bw/day and two females at 250/150 mg/kg bw/day. The remaining females were non pregnant or did not mate. No pups were born at 100 and 250/150 mg/kg bw/day. Out of the nine litters at 40 mg/kg bw/day, only six had live pups at first litter check. The number of pups per litter was decreased when compared to the control group. In addition, most of these pups did not survive the first days of lactation. At 40 mg/kg bw/day, lean and pale appearance was seen in the surviving pups and body weights were slightly, but not statistically significantly decreased when compared to the control. Macroscopic findings involved absence of milk in the stomach and blue discolouration of the snout. In addition, autolysis was noted for pups found dead. Based on the results of the study, the NOAEL for developmental toxicity in rats was considered to be lower than 40 mg/kg bw/day. High maternal toxicity was observed at 100 mg/kg bw and above.

In addition, there are data on developmental toxicity for the primary expected metabolite 2-methoxyethanol (CAS no.109-86-4) which showed similar effects in developmental toxicity studies as observed for 2-MEA. 2-methoxyethanol has consistently induced developmental toxicity in numerous oral studies in several species of laboratory animals, generally at doses lower than those that are maternally toxic, and often at the

lowest exposure level tested (CICAD, 2009). Decreased fetal body weights were noted in rats repeatedly exposed to 2-methoxyethanol doses of 16 mg/kg bw/day or more in the diet during gestation, with malformations being observed at doses of 31 mg/kg bw/day or greater, whereas maternal toxicity was present only at higher doses. Similar results were obtained in several other studies in rats exposed to 2 methoxyethanol in the diet or by gavage. In many of the studies, the cardiovascular system, kidney and skeletal system were the principal targets for 2-methoxyethanol-induced malformations; functional defects of the heart were also noted (CICAD, 2009).

10.10.6 Comparison with the CLP criteria

Reproductive toxicity category 1 in the CLP Regulation is dedicated to "substances which are known or presumed human reproductive toxicant". "Substances are classified in category 1 for reproductive toxicity when they are known to have produced an adverse effect on development in humans or when there is evidence from animal studies possibly supplemented with other information, to provide as strong presumption that the substance has the capacity to interfere with reproduction with humans. The classification of a substance is further distinguished on the basis of whether the evidence for classification is primarily from human data (category 1A) or from animal data (category 1B)."

Reproductive toxicity category 2 in the CLP Regulation is dedicated to substances which are "suspected human reproductive toxicants". "Substances are classified in category 2 for reproductive toxicity when there is some evidence from humans or experimental animals, possibly supplemented with other information, of an adverse effect on development, and where the evidence is not sufficiently convincing to place the substance in category 1."

No human data were available and therefore, Repr. 1A is not considered appropriate.

The developmental toxicity study published by Hardin et al., 1987 is not considered appropriate for classification as only one dose was tested and the dose was above the maximum tolerated dose.

In the combined repeated dose toxicity study with the reproduction/developmental toxicity screening test (Study report, 2012b) performed in rat, dose-related decrease in live birth and viability index was observed at all dose tested. At 100 and 150/250 mg/kg bw, where high maternal toxicity occured, no dam had live pups on day 1. At 40 mg/kg bw, decrease live birth index and viability index was observed without clear evidence of maternal toxicity.

As marked developmental effects were observed an OECD guideline developmental screening study, 2-MEA is considered to meet the criteria for classification as Repr. 1B (H360D) according to Regulation (EC) 1272/2008.

There are no information supporting that the effect could not be relevant for human and therefore Repr. 2 is not considered appropriate.

10.10.7 Adverse effects on or via lactation

No specific data available.

10.10.8 Short summary and overall relevance of the provided information on effects on or via lactation

No specific data available.

10.10.9 Comparison with the CLP criteria

According to the CLP criteria classification for lactation is recommended when "absorption, metabolism, distribution and excretion studies indicate the likelihood that the substance is present at toxic levels in breast milk. In the reproductive screening toxicity study, no milk was present in the stomach of the dead pups. There is no data on the presence of 2-MEA in the breast milk. Since most of these pups did not survive the first days of lactation, the reason of death is probably not related to lactation.Therefore, there is no sufficient information to propose a classification for effects on or via lactation.

10.10.10 Conclusion on classification and labelling for reproductive toxicity

In conclusion, 2-MEA has been found to induce both reproductive and developmental effects. These effects observed at 40 mg/kg bw could not be explained by maternotoxicity. Classification **Reproductive toxicity category 1B, H360FD "May damage fertility or the unborn child**" is thus warranted.

No specific concentration limit could be set for 2-MEA based on the available data as no NOAEL could be determined in the available screening study.

10.11 Specific target organ toxicity-single exposure

Not evaluated.

10.12 Specific target organ toxicity-repeated exposure

In the screening 28-day study described in table 25, at 250 mg/kg bw/day, 2 males died on day 2 (no cause of death could be determined), 1 male was killed on day 8 (showed ulcerative inflammation in the stomach with resultant peritonitis) and at 100 mg/kg bw/day, one female was killed *in extremis* on day 21 *post-coitum* (Study report, 2012b).

In the prenatal developmental toxicity study in mouse (Hardin et al., 1987), a mortality rate of 30% was observed at 650 mg/kg bw per day.

Table 27: Extrapolation of equivalent effective dose for toxicity studies of greater or lesser duration than 90 days

Study reference	**Effective dose (mg/kg/d)**	**Length of exposure**	**Extrapolated effective dose when extrapolated to 90-day exposure**	**Classification supported by the study**
Study report, 2012b	250	28-day	83 mg/kg bw	STOT RE 2
Hardin et al., 1987	650	8-day corresponding to exposure during GD 6-13	73 mg/kg bw	STOT RE 2

10.12.1 Comparison with the CLP criteria

According to Regulation (EC) No. 1272/2008 substances are classified as specific target organ toxicants following repeated exposure by the use of expert judgement on the basis of the weight of all available evidence. 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 repeated exposure are assigned to the STOT-RE categories.

Classification of 2-MEA as STOT RE 2 is justified by the following findings observed at dose values for STOT RE 2:

- In screening developmental toxicity studies in rats, after oral exposure during 1-month, high mortality of 30% was seen at doses of 250 mg/kg bw/d in males. These are within the guidance values of 30 < C ≤ 300 mg/kg bw for the 28 day repeated toxicity study for classification as STOT RE 2.
- In the prenatal developmental toxicity study in mice, a mortality rate of 30% was observed at 650 mg/kg bw per day. There are within guidance values of 100 < C ≤ 1000 mg/kg bw/d justifying classification as STOT RE 2.

2-MEA induces corrosive and acute effects. Furthermore, based on the hypothesized metabolism, it is not expected to be bioaccumulable. Furthermore, the factor between LD_{50} (404 mg/kg bw) and LOAEL (about 80 mg/kg bw/day) is about 5 supporting low cumulative potential. Moreover, lethality occurred during the 3 first days in the 28-day study suggesting that these effects are related to acute toxicity.

10.12.2 Conclusion on classification and labelling for STOT RE

Taking into account the low cumulative potential of 2-MEA, mortality observed in the sub-acute oral toxicity studies are considered to be related to acute toxicity. Thus, 2-MEA does not warrant classification as STOT RE for mortality**.**

10.13 Aspiration hazard

Not evaluated.

11 EVALUATION OF ENVIRONMENTAL HAZARDS

Not evaluated.

12 EVALUATION OF ADDITIONAL HAZARDS

Not evaluated.

13 REFERENCES

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14 ANNEXES

See separated annex I file for detailed study summaries.