Comments from the Glycol Ethers REACH consortium on the proposal for a harmonised classification and labelling of 2-butoxyethanol (EC 203-905-0, CAS 111-76-2)

End point: Acute toxicity (oral, dermal and inhalation)

We would like to submit comments on the proposal for the harmonised classification and labelling for 2-butoxyethanol (or ethylene glycol butyl ether, EGBE) that has been submitted by the German Competent Authority to amend the harmonised classification of the above substance. We request that the RAC take these comments into account in their discussions. This document specifically addresses the end point of acute toxicity. The evidence to support what we believe to be the correct classification based on the scientific data available is underpinned by the difference in species sensitivity to haemolysis, the effect which is the basis for the STOT-RE classification in the CLH proposal. Some of the evidence we have submitted to demonstrate that the STOT-RE exposure is unwarranted is also presented here as it is key to the justification for the selection of appropriate species to assess the acute toxicity hazard in humans.

Introduction

The harmonised classification of 2-butoxyethanol (EGBE) has been reviewed at EU level on two occasions. On both occasions, the acute toxicity classification according to the dangerous substance directive (67/548) criteria was retained as 'harmful' by all three routes of exposure (inhalation, oral and dermal). During the discussions of the first review following a proposal made in 1999 by France (ECBI/35/99) and covering a number of end points, the specific concentrations limit of 12.5% (lower than the default) was removed in recognition of the fact that the critical effect seen in rodents and rabbits (haemolysis) is a toxic effect that is not seen in humans[1]. The classification was again reviewed when EGBE was examined under the EU Existing Substances regulation (EU, 2006) when the prevailing acute toxicity classification was retained.

This classification proposal has arisen not because there is new data available but because under the CLP regulation (1272/2008) the thresholds for classification for acute toxicity have reduced significantly for the inhalation and dermal routes of exposure.

The toxicology of this substance is well understood. The main and most significant substance specific toxic effect of EGBE is that it causes haemolysis, in some cases at quite low doses. However, it is also a highly species-specific effect and of critical importance is that rodents and rabbits are particularly susceptible to haemolysis caused by EGBE exposure whilst humans are remarkably resistant. This difference has been accepted by a wide number of regulatory bodies and taken into account when interpreting the toxicological data on the substance. For instance:

- In 1994, based on evidence to show that the acute toxicity of EGBE is better represented by data from species that are resistant to haemolysis, such as the guinea pig, the United Nations (UN) Committee of Experts deleted EGBE from its list of substances requiring special toxicity labelling. As a result, 2BE (UN2369) was not included in the Ninth Revised Edition of the UN Recommendations and since this date has not been classified as hazardous for transport (UN, 1994a, 1994b).

[1] As a point of note, the R37 risk phrase was also deleted as there was found to be no evidence to support it.

- The OECD under their Existing Chemicals Review Programme have reviewed EGBE on a number of occasions. It was first reviewed at SIAM 3 in 1997 with Australia as the dossier sponsor. The substance was re-reviewed as part of the monoethylene glycol ether series at SIAM 19 in 2004 with the USA as the rapporteur. The results of the EU risk assessment were presented by France to the OECD at SIAM 23 in 2007. These reviews consistently recognised the big species differences in sensitivity to humans. The 2004 review states "Red blood cells of humans are many-fold more resistant to toxicity from EGBE in vitro than those of rat." (OECD, 1997, 2004, 2007)
- In 1999, The US EPA in their IRIS risk assessments of EGBE recognised that the lack of sensitivity of humans to haemolysis allowed lower assessment factors to be justified when deriving the Rfd values (US EPA, 1999). The relative differences in species response were still recognised in the update carried out in 2010 (US EPA, 2010).
- In their 1999 Priority List Assessment of the substance, the Canadian EPA concluded that the available data supported at least a 10-fold difference in sensitivity between rats and humans for the haemolytic effect of butoxyacetic acid, the metabolite of EGBE (CEPA, 1999).
- In 2006, IARC recognised that rats and mice are more sensitive to EGBE-induced haemolysis than other experimental species and that humans appear to be much less sensitive (IARC, 2006).
- In the 2006 EU risk assessment of EGBE, an interspecies toxicodynamic factor of 0.1 rather than the default 2.5 was used to derive the margin of safety for human risk assessment in recognition that humans are far less sensitive to the haemolytic effects of EGBE than rats, mice and rabbits (EU, 2006).

Meek et al (2003), then of the Existing Substances Division of Health Canada, in a publication on an initiative of the IPCS on the derivation of Chemical Specific Assessment Factors, used EGBE as an example of a substance where it is justified to reduce the toxicodynamic assessment factor from 2.5 to 0.1 due to the resistance of humans to the haemolytic effect of EGBE.

Michael Cunningham, then of the NIEHS in the USA, in his paper of 2002 used EGBE and its selective species specific haemolytic effects as an example of the differences between species and how care therefore is required in extrapolating the results from animal studies to humans.

There has been no new data published since these reviews were completed that would change the validity of these decisions.

2-butoxyethanol and haemolysis

The haemolytic effects of EGBE in sensitive species are seen following both acute and repeated exposure and can be demonstrated using in vitro approaches as well. The lack of effects in humans, including of potentially sensitive sub-populations, has been shown using in vitro approaches and also from the documentation of numerous accidental and deliberate ingestion incidents in humans as well as in human volunteer inhalation exposures.

There is a large database of acute toxicity data for 2-butoxyethanol (EGBE) by all three routes of exposure, covering multiple species. An extensive review of this data and a recommendation of how it should be interpreted under the GHS classification and labelling criteria was recently published by Boatman (2014). As this is a free access publication, a copy of it is included in this response as a separate attachment and therefore the detail of its contents is not repeated here, although a summary is presented for convenience.

The haemolysis of red blood cells is actually caused by butoxy acetic acid (BAA), the primary metabolite of EGBE (Carpenter, 1956; Ghanayem, 1987, 1989, 1993). This metabolite is produced by the alcohol and aldehyde dehydrogenase enzyme system in the liver (Ghanayem, 1987). Rodents, guinea pigs and humans are therefore all capable of converting EGBE to BAA. BAA appears to increase the fragility of red blood cells in some species, leading them to rupture when passing through the vascular system (Ghanayem, 1989). It also produces more haemolysis in older animals compared with younger animals. This was demonstrated to be due to the increased fragility of older red blood cells (present in higher numbers in old animals). Animals that had been bled, triggering the increased production of new red blood cells, were less susceptible to the haemolytic activity of BAA; in these animals the LD50 of EGBE was higher than animals of the same age that had not been bled indicating that haemolysis is a critical component of the acute toxicity of EGBE (Ghanayem , 1990, 1992; Sivarao, 1995).

There is a clear species difference in susceptibility to the haemolysis caused by BAA. Acute toxicity studies in rats and rabbits show clear evidence of haemolysis, whereas it is not seen in guinea pigs. In vitro studies of red blood cell haemolysis using rodents, rabbits, guinea pigs, cats, dogs, pigs and primates (including humans) demonstrated that the red blood cells of rodents and rabbits are considerably more sensitive to this effect whereas guinea pigs and humans are significantly less sensitive (Ghanayem, 1993; Udden 1994a, 2000). Udden (1994b) also looked at potentially susceptible sub-populations of humans, including the elderly and young and those with diseases of the red blood cells (hereditary spherocytosis and sickle cell disease); none of these populations showed any susceptibility to BAA induced haemolysis.

Physiologically based pharmacokinetic models of EGBE demonstrate that it is not possible to achieve a high enough plasma concentration of BAA in humans by inhalation[2] or dermal routes[3] to trigger even a slight haemolysis of red blood cells (Corley, 1996; EU, 2006). This is consistent with the reported cases of acute human intoxication with EGBE where there was no evidence of haemolytic activity (Bauer, 1992; Butera, 1996; Burkhart, 1998; Dean, 1992; Gijsenbergh, 1989; Gualtieri, 1995, 2003; Hung, 2010[4]; McKinney, 2000; Rambourg-Schepens, 1988. In an old volunteers study 2 men and one woman were exposed to 0.98mg/L EGBE for 2x4hr exposures with a 30 minute break in between (The Dow Chemical Co, 1955). Subsequent haematology showed no signs of haemolysis[5]. Such an exposure in rodents or rabbits would cause profound haemolysis.

Boatman (2014) lists the results of the available acute toxicity studies with EGBE and, in particular, the clinical and toxic effects seen. The studies with mice, rats and rabbits consistently report observations that are coherent with severe haemolysis being the critical effect leading to mortality. Human red blood cells are significantly less susceptible to BAA mediated haemolysis, therefore the LD50 and LC50 values derived from mice, rat and rabbit studies are not reliable predictors of acute toxicity potential in humans; they will significantly over-predict the likely toxicity at a given dose. It is more appropriate to use a species that has similar resistance to BAA mediated haematotoxicity. Guinea pig red blood cells are similar to human red blood cells regarding their sensitivity to BAA

[2] Up to the saturated vapour concentration of 1160ppm.

[3] Using worse case permeability co-efficient, assuming 10% of skin surface exposed and no evaporative losses.

[4] Note that the Hung publication was not included in the CLH document – details are therefore shown in the appendix 1 to this document.

[5] The CLH dossier author notes that some haemolytic effects were noted in the documented poisonings. However, this statement does not seem to be supported by the evidence offered. Most reports noted no haemolysis. Anaemias were noted as non-haemolytic. The isolated incidents of haematuria could be due to other toxic events.

mediated haemolysis. This similarity in sensitivity to haemolysis makes the guinea pig a more appropriate model for assessing the acute toxicity potential of EGBE in humans (Gingell et al., 1998).

Species sensitive to BAA induced haemolysis	**Species resistant to BAA induced haemolysis**
Rat, mouse, rabbit, hamster, baboon	Human, guinea pig, dog, cat

Other toxic modes of action should be considered. In cases of human poisoning incidents involving exposure to EGBE, the primary toxic effect was metabolic acidosis, likely resulting from high concentrations of BAA in the blood (Bauer, 1992; Butera, 1996; Burkhart, 1998; Dean, 1992; Gijsenbergh, 1989; Gualtieri, 1995, 2003; Hung, 2010[4]; McKinney, 2000; Rambourg-Schepens, 1988). There were no reports of haemolysis in these cases. In the cases where haematuria and haemoglobinuria were reported, there were other severe toxicity effects that could have been a cause of the observations. It is notable that in the publication of Dean, no haemolytic effects were noted in any of the 24 cases of accidental ingestion by children of significant amounts of EGBE. Metabolic acidosis, if severe enough, can produce mortality and this mode of action is likely to be responsible for acute toxicity observed in guinea pigs following oral exposure to EGBE, but this effect only occurs at much higher doses than those which cause haemolysis in sensitive species. Metabolic acidosis is not a substance specific effect and can be triggered by any substance that is metabolised to acidic species if ingested in sufficient quantity. It is notable that the EU risk assessment of EGBE used the human data rather than the animal data as a starting point for deriving safe acute exposure levels.

Therefore, based on clear and compelling scientific evidence, the guinea pig is the most appropriate species for assessing the acute toxicity potential of EGBE to humans (Gingell et al., 1998). Reliable and robust studies exist for the guinea pig for all three routes of exposure.

Oral route of exposure

There are two studies available using the guinea pig, one of which is a modern guideline GLP study. The results are shown in the table below:

Doses (mg/kgbw)	**Route**	**Result**	**Reference**
500, 1000, 2000	Gavage in water	LD_{50}=1414mg/kgbw. No haemolysis	Shepard (1994a), Gingell (1998)
No data	Gavage in water	LD_{50}=1200mg/kgbw. No haemolysis	Smyth (1941), Carpenter (1956)

Even though these studies differ significantly as to when they were conducted, the results are consistent and support a category 4 classification and the consortium members of the REACH registration of EGBE therefore agree with the classification for this route as proposed in the submitted Annex XV dossier.

Dermal route

There are four studies available using the guinea pig, one of which is a modern guideline GLP study. The results are shown in the table below:

Doses (mg/kgbw)	Route	Result	Reference
2000 (limit dose study)	Occlusive, no vehicle	LD_{50}>2000mg/kgbw. No effects	Shepard (1994b), Gingell (1998)
No data	Occlusive, abraded and intact	LD_{50}=207mg/kgbw. Abraded LD_{50}=270mg/kgbw. Intact	Roudabush (1964)
2, 5, 6, 8, 10 mL/kgbw	Occlusive, no vehicle	LD_{50}=6650mg/kgbw	Dow (1938)
1.2, 4.8 mg/kgbw	Occlusive until all material absorbed	LD_{50}>1200mg/kgbw	Wahlberg (1979)

A weight of evidence approach should be used here. Three of the studies are consistent with each other and also with the data from the oral acute toxicity study. One study indicated a 10 fold increase in toxicity compared to the other studies. The Shepard and Roudabush studies both used the same strain of guinea pig (Hartley). The results from the Roudabush study are sufficiently out of line with all the other oral and dermal data that it is questionable whether it can be regarded as 'valid and well performed' and therefore it is given a low weighting in the overall consideration. The study used four animals at three doses and exposed the abdomens of the animals, whereas the current guideline requires the dorsal area to be used. The authors acknowledged that using the abdomen for dermal exposure produces lower results than the back. The other three studies, and particularly the most recent GLP study (Shepard) indicate that no classification is required under the CLP regulation as the weight of evidence is that in species relevant to human hazard assessment, the LD50 exceeds 2000mg/kgbw.

It should be noted that even in the rat the LD50 exceeds 2000mg/kg in all studies. It is only for the rabbit, which is exquisitely sensitive to haemolysis, that LD50 values are found in the range 450 - 850mg/kg, although even with the rabbit, under semi-occlusive conditions, the LD50 is >2000mg/kg. The clinical findings in these studies, where reported, are consistent with haemolysis occurring. For instance, Duprat (1979), who reported the lowest LD50 for rabbits, noted that the morphological changes seen in the kidney were probably as a consequence of in vivo haemolysis. Appendix 2 to this document contains the detailed clinical and pathological findings reported from acute toxicity studies by the dermal route using the rabbit to show the findings that can be attributed as secondary to haemolysis.

The guidance on application of the classification and labelling criteria states the following (section 3.1.2.3.2.):

> *In general, classification is based on the lowest ATE value available i.e. the lowest ATE in the most sensitive appropriate species tested. However, expert judgement may allow another ATE value to be used in preference, provided this can be supported by a robust justification. If there is information available to inform on species relevance, then the studies conducted in the species most relevant for humans should normally be given precedence over the studies in other species. If there is a wide range of ATE values from the same species, it may be informative to consider the studies collectively, to understand possible reasons for the different results obtained.*

We believe that this justifies selecting the most appropriate species and a weight of evidence approach that weights multiple values in the same species according to recent they are along with their compliance with standard testing methodology and GLP. We believe that such an approach should lead to a conclusion of no classification since the most recent and therefore most reliable

studies in guinea pigs (the preferred species for this substance due to its resistance to haemolysis and therefore a closer model for human toxicity) did not show 50% mortality when test animals were subjected to exposures of 2000mg/kg. Therefore, the consortium members of the REACH registration of EGBE do not agree with the category 3 classification as proposed in the submitted Annex XV dossier, but support no classification for acute toxicity by the dermal route as the most appropriate interpretation of the data with respect to the hazard to humans.

Inhalation route

There are two studies available using the guinea pig, one of which is a modern guideline GLP study for which the original study report is available. The results are shown in the table below:

Doses (mg/kgbw)	Exposure time and type	Result	Reference
3.1-3.4mg/L	1 hr, whole body	LC_0>3.1-3.4mg/L (80-87% of SVP). no adverse effects seen	Dow (1994), Gingell (1998)
2.0mg/L	7hr, whole body	LC_0>2.0mg/L no adverse effects seen	Dow (1974)
6.4mg/L	7 hr	50% mortality	Tyler (1984)

The Dow 1994 study was only performed for 1 hour as the results were intended to be used to identify the appropriate classification for transport. This study is to GLP and the original report is available. The Dow (1974) study is in the REACH registration dossier for this substance but was not included in the CLH dossier issued for public comment. The Tyler 1984 reference is a secondary source that cites a study report from 1943 that is not available.

The studies did not show any mortality when test animals were subjected to exposure at the maximum vapour pressure under the experimental conditions. The test results should be put into the context that the saturated vapour concentration at 20°C is 4.4mg/L and that the maximum that can normally be sustained under dynamic test conditions in the laboratory is 50-75% of this. Under real life conditions, even these levels cannot realistically be approached. These results can therefore be regarded as no toxicity up to the maximum achievable concentration. For this reason, care is also needed in extrapolating from longer exposure durations using the Haber rule as LC50 values so derived that exceed the saturated vapour concentration are meaningless.

Data is available in four other species. Of the five studies in rats, three did not reach the LC50 at the maximum tested dose (maximum achievable concentration), one just about reached the LC50 (but only in females) at around the saturated vapour concentration (SVC) and only one showed an LC50 below the SVC (and this was at around 50% of the SVC). A study that looked at the difference between older and younger rats found that the LC50 in older animals was lower than in younger animals and that the LC50 was not reached at the SVC in the latter. This finding is consistent with the sensitivity to haemolysis and the hypothesis that haemolysis is the cause of death in a species sensitive to this effect. A single study in dogs showed no adverse effects and the data in mice suggested that the 4 hour LC50 would be around the SVC (based on an extrapolation using the Haber rules from a 7 hour exposure.). Only in a single study in rabbits, a species very sensitive to haemolysis, was an LC50 clearly below the SVC obtained. Not all of this data was reported in the CLH dossier, although it is available in the disseminated registration dossier for the substance. Appendix 3 to this document contains a summary table of the additional data and appendix 4 contains a table with additional information on the pathology of those studies that were cited in the CLH dossier.

A weight of evidence approach that takes into account the fact that the guinea pig is the best species to model the acute toxic effect of EGBE in humans and rabbits the worst (due to its sensitivity to haemolysis – see discussion under oral and dermal routes), and the relatively low volatility of EGBE (vapour pressure of ~80Pa at 20°C[6]) leads to the conclusion from the scientific evidence that, outside of laboratory conditions, there is no acute toxicity human hazard from inhalation exposure to EGBE and that classification is not required for this route. Therefore, the consortium members of the REACH registration of EGBE do not agree with the proposed category 3 classification cited in the submitted Annex XV dossier, but support no classification for acute toxicity by the inhalation route for vapour exposure.

Human data

According to the guidance on the application of the CLP criteria, human data can be considered for use in classification decisions. A weakness of such data can be the lack of exposure or dose information. Useful data for EGBE from multiple sources has been identified that both reports adverse effects and also provides dose information. All of this data is for the oral route. This information can therefore be considered to inform an appropriate classification decision and is discussed in more detail below.

There is a significant amount of acute oral toxicity data available in humans primarily through case reports of suicide attempts made using cleaners and other products containing EGBE. It is notable that none of these attempts resulted in death and all patients made full recoveries. The available published data is shown in the table below. The table only includes data on adults. Data is also available on children (who also all survived) but the dose information is too imprecise to be usable for classification and labelling purposes, but it is notable again that no cases were fatal with doses up to ~2g/kg (Dean, 1992).

Sex	**Age (years)**	**Estimated human dose (g/kg) received**	**Reference**
Male	53	0.65	Bauer (1992)
Male	47	0.57	Butera (1996)
Female	51	0.4 – 1.2	McKinney (2000)
Male	19	2.0 - 4.2	Burkhart (1998)
Male	18	1.1 – 1.5	Gualtieri (1995)
Female	23	1.0	Gijsenber (1989)
Female	50	0.5 – 1.0	Rambourg-Schepens (1988)
Male	53	1.8 - 3.0	Hung (2010)**Error! Bookmark not defined.**

All of these studies are reported in detail in the IUCLID dossier.

All of this information was reviewed in the EU risk assessment of EGBE published in 2006 with the exception of the Hung data, which postdates it. The following is an extract from the risk assessment conclusions of a review of this data:

> *Acute human toxicity data comes from children accidental ingestion or adult suicide attempts made with mixtures containing EGBE and from one study on human volunteers by inhalation. For oral*

[6] Value cited in CLH dossier equivalent to a saturated vapour concentration of ~3.9mg/L..

route case reports, ingested doses are difficult to evaluate because of the lack of data concerning the body weight of all patients and the exact ingested dose, but a semi-quantitative estimation of the ingested doses was made for each case (see table above – estimate for Hung data also included). The range of doses which lead to clinical symptoms varies between 0.5 and 4.5 g/kg bw. In all cases, patients exhibited SNC depression and metabolic acidosis. Signs of haemolysis were seen in some cases but this finding was not systematic (this showed that human is much more resistant to haemolysis than rodents). After a first acute ingestion, a second administrator some days later did not exhibit the same symptoms, this finding was also seen with animals in some studies. In these cases, EGBE was ingested together with other substances (ethanol and/or unknown substances) that could have some influence on the symptoms seen. Between 0.5 and 1.5 g/kg bw the patients totally recovered after treatment. According to this data, a LOAEL of 400 mg/kg bw can be taken into account for acute toxicity by oral route in humans in the risk characterisation section. If a risk characterisation by dermal and inhalation route is needed for acute effects, kinetic data is sufficient to extrapolate this oral LOAEL to inhalation and dermal LOAEL. Human data is preferred for risk characterisation, especially for EGBE because of its haematotoxicity more marked in animals than in humans. It should be noted that this is a worst-case estimation derived from the McKinney paper in which the possible range of exposure was between 0.4 and 1.2 g/kg bw.

The conclusion of a LOAEL of 400mg/kg for humans seems reasonable if rather conservative based on the available data.

The guidance on the application of the CLP criteria states that human data should be used to derive that ATE (single or dose range expected to cause mortality) without any adjustment for comparison with the classification criteria. For the oral route, the ATE based on the LOAEL above would therefore be 400mg/kg. This falls in the range for classification as category 4 under the EU CLP regulation 1272/2008 (CLP). This is clearly conservative as it is based on a LOAEL and not mortality or an LC50, which would be significantly greater than this.

This human data can be converted by route to route extrapolation to assess appropriate classifications for other routes of exposure.

For the inhalation route, it is informative to compare the dose that would derive from exposure to the saturated vapour concentration of 3.9mg/l[7]. Based on table R8.2 from the guidance on information requirements and chemical safety assessment, human respiratory volume can be considered to be 0.2L/min/kg. For a 4 hr exposure (the normal duration for an acute inhalation study), this becomes 48L/4hr/kg. At a maximum achievable concentration of 3.9mg/l, this would result in a maximum theoretical dose of 187mg/kg (or 112mg/kg assuming 60% absorption (Kumagai, 1999)) which is clearly well below the LOAEL derived for humans. On this basis, it becomes apparent that it is not possible to reach harmful levels of exposure (in terms of classification requirements) to EGBE by the inhalation route, which supports the conclusion from the animal data that no classification is required for this route.

A similar approach is also possible for the dermal route. Assuming 100% absorption by the oral route and 30% by the dermal route (figures derived in the EU risk assessment) would mean that the equivalent LOAEL by the dermal route would be 1333mg/kg. A comparison with the classification criteria would lead to the conclusion that category 4 under the EU CLP regulation 1272/2008 (CLP) is appropriate based on the face value of this data. However, this is clearly conservative as it is based on a LOAEL and not mortality or an LC50, which would be significantly greater than this. It would be

[7] Based on the figure of 80Pa for the SVP as cited in the CLH dossier

reasonable to assume that the lethal dose would be at least a factor of 2x higher than the LOAEL, which would take the ATE above 2000mg/kgbw and imply no classification is required. The data from humans is therefore supportive of the conclusions drawn from data from animals which are comparably resistant to haemolysis, i.e. no classification for the dermal route is warranted.

Summary

When classifying a substance for toxicity, it is appropriate to consider relative species sensitivity to the critical toxic effects seen. According to the CLP regulation (paragraph 3.1.2.1. of annex I) '*When experimental data for acute toxicity are available in several animal species, scientific judgement shall be used in selecting the most appropriate LD50 value'*. In this case there is compelling evidence to suggest that the rat, mouse and rabbit are not good surrogates and that the guinea pig is the most appropriate species to use. For EGBE, the critical toxic effect is haemolysis, which can be sufficiently severe to be fatal in sensitive species. In the event of humans being more sensitive to an effect than test animals, it is appropriate to take this into account when making a decision on classification and there are examples of where this has been done. When the situation is reversed, then the same considerations should apply as there is no value to be gained from 'over classifying' a substance. It is certainly not appropriate to selectively take the data from species that are particularly sensitive to an effect that is irrelevant for humans (i.e. the rabbit) to justify a classification decision. For EGBE, the use of acute toxicity data from humans and from animal species appropriate to predict toxicity in humans (i.e. eliminating from consideration species which show toxic effects not relevant to humans) leads to a consistent conclusion that EGBE should be classified as follows:

- Oral route: category 4. No specific ATE required (default sufficient)
- Dermal route: no classification warranted
- Inhalation route: no classification warranted

References

Bauer P, Weber M, Mur JM, Protois JC, Bollaert PE, Condi A, Larcan A, Lambert H (1992). Transient non-cardiogenic pulmonary edema following massive ingestion of ethylene glycol butyl ether. Intensive Med. Care. 18:250-1.

Boatman R, Kelsey J, Ball N (2014) Acute toxicity classification for ethylene glycol mono-n-butyl ether under the Globally Harmonized System. Regul Toxicol Pharmacol. 2014

Burkhart KK and Donovan JW (1998). Hemodialysis following butoxyethanol ingestion. Clinical Toxicol. 36(7), 723-725.

Butera R, Lapostolle F, Astier A, Baud FJ (1996). Metabolism and toxic effects of ethylene glycol butyl ether in a case of poisoning treated with 4-methylpyrazole. Toxicology Letters 88, p49.

Carpenter CP, Pozzani MS, Weil CS, Nair JH, Keck GA, Smyth HF (1956). The toxicity of butyl cellosolve solvent. Arch Ind Hlth, 14, -114-31.

CEPA (1999) Canadian Environmental Protection Act Priority Substances List Assessment Report, 2-butoxyethanol, Health Canada, ISBN 0-662-33597-X.

Corley RA (1996) Assessing the risk of haemolysis in humans exposed to 2-butoxyethanol using a PBPK model, Occup Hyg, 2, 45-55.

Cunningham ML (2002). A mouse is not a rat is not a human: species differences exist. Toxicol Sci. 2002 Dec;70(2):157-8.

Dean BS and Krenzelok EP (1992). Clinical evaluation of pediatric ethylene glycol monobutyl ether poisonings. Clinical toxicol. 30(4), 557-563.

Dow Chemicals (1938). The toxicity of glycols and their derivatives 47. Single doses of methyl cellosolve, cellosolve and butyl cellosolve by skin absorption. Testing laboratory: Mellon Institute of Industrial Research. Report no.: 274-2. Owner

company: Report of the Union Carbide Corporation, a wholly owned subsidiary of The Dow Chemical Company. Study number: K-002573-024. Report date: 1939-07-11.

Dow Chemical Company (1994). Ethylene glycol butyl ether: acute vapor inhalation toxicity study in guinea pigs. Testing laboratory: Bush Run Research Centre. Report no.: 94N1392.

Dow Chemical Company (1974). Inhalation toxicity studies on three samples of ethylene glycol monobutyl ether (Dowanol EB), n-butyl Oxitol-Shell USA, n-butyl Oxitol-Shell Europe. Testing laboratory: The Dow Chemical Company, Midland, USA. Report no.: T-5-13-8-13. Owner company: The Dow Chemical Company. Report date: 1974-10-07.

Duprat P, Gradski, P (1978) Percutaneous toxicity of butyl cellosolve. IRCS Medical Sci, 7, 26

European Union (2006), Risk Assessment Report: 2-BUTOXYETHANOL , Vol 68, https://echa.europa.eu/documents/10162/e74a38e1-b9e1-4568-92c5-615c4b56f92d

Ghanayem BI (1989). Metabolic and cellular basis of 2-butoxyethanol-induced hemolitic anemia in rats and assessment of human risk in vitro. Biochem. Pharmacol., 38(10), 1679-1684.

Ghanayem BI, Burka LT, Sanders JM, Matthews HB (1987). Metabolism and disposition of Ethylene Glycol Monobutyl Ether (2-butoxyethanol) in rats. Drug Metabolism and Disposition, 15 (4), 478-484.

Ghanayem BI and Sullivan CA (1993). Assessment of the haemolitic activity of 2-butoxyethanol and its major metabolite, butoxyacetic acid, in various mammals including humans. Human & Exp. Toxicol., 12, 305-311.

Ghanayem BI, Sanders JM, Clark AM, Bailer J and Matthews HB. (1990). Effects of dose, age, inhibition of metabolism and elimination on the toxicokinetics of 2BE and its metabolites. J. Pharm. Exp. Therap., 253, 136-143.

Ghanayem BI, Sanchez IM and Matthews HB (1992). Development of tolerance to 2-butoxyethanol-induced hemolytic anemia and studies to elucidate the underlying mechanisms Bibliographic source. Toxicol. Appl. Pharmacol., 112, 198-206.

Gijsenbergh FP, Jenco M, Veulemans H, Groeseneken D, Verberckmoes R and Delooz HH, (1989). Acute butylglycol intoxication: a case report. Human Toxicol., 8, 243-245.

Gingell R (1998). Acute toxicity of ethylene glycol mono-n-butyl ether in the guinea pig. Food Chem Toxicol 36 (9-10) 825-9.

Gualtieri J, Harris C, Roy R, Corley R and Manderfield C (1995). Multiple 2-butoxyethanol intoxications in the same patient: clinical findings, pharmacokinetics and therapy. J. Toxicol. & Clin. Toxicol., 33(5), 550-551.

Gualtieri JF, DeBoer L, Harris CR, Corley R (2003). Repeat ingestion of 2-butoxyethanol: a case report and literature review`. J Toxicol Clin Toxicol, 41(1) p57-62.

Hung T, Dewitt CR, Martz W, Schreiber W, Holmes DT (2010). Fomepizole fails to prevent progression of acidosis in 2-butoxyethanol and ethanol co-ingestion. Clinical Toxicology, 48, 569-71.

IARC (2006) Formaldehyde, 2-Butoxyethanol and 1-tert-Butoxypropan-2-ol, IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, International Agency for Research on Cancer, Volume 88, www.iarc.fr.

Kumagai S, Oda H, Matsunaga I, Kosaka H, Akasaka S (1999) Uptake of 10 polar solvents during short term respiration. Toxicol Sci, 48, 255-63.

McKinney PE, Palmer RB, Blackwell W and Benson BE. (2000). Butoxyethanol ingestion with prolonged hyperchloremic metabolic acidosis treated with ethanol therapy. Clin. Toxicol., 38(7), 787-793.

Meek B, Renwick A, Sonich-Mullin C (2003). Practical application of kinetic data in risk assessment – an IPCS initiative, Toxicol Lett, 138, 151-160.

OECD (1997) 2-butoxyethanol, SIDS Initial Assessment Report for 6th SIAM, Paris 9-11th June 1997). http://webnet.oecd.org/Hpv/UI/handler.axd?id=0fbae729-1207-4829-b0e8-f1f1b1f7a8d4

OECD (2004) monoethylene glycol ethers category, SIDS Initial Assessment Report For SIAM 19 Berlin, Germany, 19-22 October 2004. http://webnet.oecd.org/Hpv/UI/handler.axd?id=e50677bd-baab-4afc-92aa-76fd972a171e

OECD (2007) 2-butoxyethanol, SIDS initial assessment profile, SIAM 23, 17-20 October 2006 http://webnet.oecd.org/Hpv/UI/handler.axd?id=e703d99b-deb2-4f96-83e0-0db42ea867dc

Rambourg-Schepens MO, Buffet M, Bertault R, Jaussaud M, Journe B, Fay R and Lamiable D, (1988). Severe ethylene glycol butyl ether poisoning. Kinetic and metabolic pattern. Human Toxicol., 7, 187-189.

Roudabush RL, Terhaar CJ, Fassett DW, Dzubia SP (1964). Comparative acute effects of some chemicals on the skin of rabbits and guinea pigs. Tox Appl Pharmac 7, 559-65.

Shepard KP (1994a). Ethylene glycol monobutyl ether. Acute oral toxicity study in the Guinea Pig. Testing laboratory: Eastman Kodak, Rochester, NY. Report no.: 940300A0. Owner company: American Chemistry Council. Study number: EGE-58.0-GPIG-EASTMAN. Report date: 1994-06-23.

Shepard KP (1994b). Ethylene glycol monobutyl ether. Acute dermal toxicity study in the guinea pig. Testing laboratory: Eastman Kodak, Rochester NY. Report no.: HAEL no: 94-0300 KAN: 902270. Owner company: American Chemistry Council. Study number: EGE-58.0-GPIG-EASTMAN. Report date: 1994-06-23.

Sivarao DV and Mehendale HM (1995). 2 butoxyethanol autoprotection is due to resiliance of newly formed erythrocytes to hemolysis. Arch. Toxicol., 69, 526-532.

Smyth HF, Seaton J, Fischer L (1941). The single dose toxicity of some glycols and derivatives. J Ind Hyg Toxicol 23(6) p259-68.

The Dow Chemical Company (1955). Butyl cellosolve III, Repeat Inhalation. Mellon Institute of Industrial Research. Testing laboratory: Mellon Institute of Industrial Research. Report no.: 18-24.

Tyler T.R. (1984): Acute and subchronic toxicity of ethylene glycol monobutyl ether. Environmental Health Perspectives 57, 185-191.

Udden MM and Patton CS (1994a). Hemolysis and deformability of erythrocytes exposed to butoxyacetic acid, a metabolite of 2-butoxyethanol: sensitivity in rats and resistance in normal humans. J. Applied Toxicol., 14(2), 91-96.

Udden (MM (1994b) Hemolyis and deformability of erythrocytes exposed to butoxyacetic acid, a metabolite of 2-butoxyethanol: II Resitance in red blood cells from humans with potential susceptibility. J Appl Toxicol, 14(2), 97-102.

Udden MM (2000). Rat erythrocyte morphological changes after gavage dosing with 2-butoxyethanol: a comparison with the in vitro effects of butoxyacetic acid on rat and human erythrocytes. J. Appl. Toxicol., 20, 381-387.

UN (1994a) Report of the United Nations Sub-Committee of Experts on the Transport of Dangerous Goods, 9th session, Paragraphs 113-5, July 1994).

UN (1994b) Report of the United Nations Sub-Committee of Experts on the Transport of Dangerous Goods, 18th session, Paragraphs 57-9, July 1994.

US EPA (1999) Toxicological Review Of Ethylene Glycol Monobutyl Ether (EGBE) (CAS No. 111-76-2) In Support of Summary Information on the Integrated Risk Information System (IRIS) October 1999.

US EPA (2010) Toxicological Review Of Ethylene Glycol Monobutyl Ether (EGBE) (CAS No. 111-76-2) In Support of Summary Information on the Integrated Risk Information System (IRIS). EPA/635/R-08/006F. March 2010.

Wahlberg JE, Boman A (1979). Comparative percutaneous toxicity of ten industrial solvents in the guinea pig. Scan J Work Env Hlth 5, 345-51.

Appendix 1 Additional information on poisoning incidents omitted from CLH dossier

Type of data/report	Test substance,	Relevant information about the study (as applicable)	Observations	Reference
Case report	150-250mL of pure 2-butoxyethanol, corresponding to about 1.8 to 3 g/kg bw	Accidental ingestion by a 53-year man who mistook product for alcohol. Patient had already consumed alcohol	On admission to the hospital, fomepizol was given ~75 mins after ingestion in the belief that the patient had ingested ethylene glycol. The patient became comatose, suffered severe metabolic acidosis and needed treatment in intensive care using haemodialysis. . This resolved in 24 hours and the patient made a full recovery. Haemolytic anaemia was not observed.	Hung (2010)

Appendix 2. Clinical and pathological findings in acute dermal toxicity studies using the rabbit

LD50	Type	Clinical and pathology findings	Reference
560mg/kg	Occlusive	Extreme kidney congestion, haemoglobinuria, pale liver, engorged spleen seen in animals that died. Bloody urine reported.	MIIR (1952, Carpenter (1956)
680mg/kg	Occlusive	No information reported	Roudabush (1965)
580mg/kg	Occlusive	At 24 hr, at 2000mg/kg all rabbits exhibited lacrimation, bloody urine, flaccid muscle tone and anorexia. No spontaneous movement were noted. No toxic signs were reported for the 250 mg/kg group; for the 500 and 1000 mg/kg group blood was found in urine and for the 1000 mg/kg group animals were flaccid. Pathology: In animals that died animals, necropsy findings were blood in urine and bladder and sometimes liver and renal injuries.	MB Research (1976)
100mg/kg	Not stated	Clinical findings: Prostration, hypothermia, haemoglobinuria. Pathology: liver congestion, necrotic foci with mesenchymatous reactions and inconstant steatosis, passive spleen congestion where sinuses filled with erythrocytes, atrophic white pulp, congestion and thickening of alveoli walls, similar to interstitial pneumonitis, enlarged kidneys with extensive haemoglobinuric nephrosis and interstitial reaction (attributed by the authors to haemolysis and the likely cause of delayed deaths), cutaneous lesions.	Duprat (1979)
569mg/kg	Occlusive	High dose animals showed erythema and necrosis at application site. Pathology: In dead animals: lungs and livers orange/red, spleens dark, kidneys very dark red, peritonea and intestines orange, blood in urine. In survivors: nothing remarkable but only one necropsied.	Bushy Run (1980)
435mg/kg	Occlusive	Clinical signs: At lower doses: anorexia, slight depression, cyanosis, ataxia, soft faeces. At higher doses: salivation, nasal discharge, iritis, significant depression, laboured breathing, prostration. Pathology: Thymus – enlarged; Liver – pale, vascular; Kidney cortex: wide, dark brown, dark red, black, discoloration (pale or dark red) dilated pelvis. Large intestines – vascular. Small intestines -vascular, yellow discoloration. Stomach – discoloration (dark brown to black). Urinary bladder - fluid, dark reddish brown, red. Thoracic cavity – contained clear fluid. Skin discoloured -treated area, red, dark red, discoloured. Eyes & body fat – tinged.	Eastman (1981)
>2000mg.kg	Semi-occlusive	Clinical signs: Lower dose group: No signs of systemic toxicity or irritation were noted during the study. Very slight to well defined erythema/slight oedema noted at dose site. Higher dose group: Lethargy, stained urine, decreased respiratory rate, hunched posture, yellow skin and eyes commonly noted. Also isolated incidents of righting reflex, hypothermia, ataxia and diarrhoea. All surviving animals except 2 recovered to normal signs within 14 days. Very slight to well defined erythema/slight oedema noted at dose site. Also noted were scattered areas of black or green necrosis, severe haemorrhage of the dermal capillaries, hardened scabs and desquamation. The yellow appearance of the skin masked evaluation of erythema in 5 animals. Pathology: Lower dose group: No abnormalities were noted at necropsy.	Safepharm (1994a)

		Higher dose group, animals that died or were sacrificed in extremis: pale kidneys, dark liver, haemorrhage of the gastric mucosa - non glandular epithelium of the stomach - small and large intestines, red fluid in the bladder. No abnormalities noted in surviving animals.	
841 mg/kg	Occlusive	Common clinical signs of toxicity in all groups were: Lethargy, ataxia, red stained urine, diuresis, decreased respiratory rate, hunched posture, yellow skin and eyes commonly noted (later particularly in two higher dose groups). Also isolated incidents of loss of righting reflex, hypothermia and diarrhoea were seen in the high dose animals. All surviving animals except 2 recovered to normal signs within 14 days. Very slight to well defined erythema and slight to severe oedema noted at dose site. Also noted were scattered areas of black or green necrosis, slight haemorrhage of the dermal capillaries, hardened scabs over dried blood and desquamation. Signs of irritation also included light brown discoloration of the epidermis or small areas of light brown discoloration and crust formation. The yellow appearance of the skin masked evaluation of erythema in some animals. Common pathology findings in animals that died or were sacrificed in extremis: haemorrhagic lungs, dark kidneys, dark or pale liver or patchy liver pallor and red fluid in the bladder. Other observed signs in the two higher dose animals that died were: haemorrhage of the gastric mucosa and the small and large intestines, red fluid in the bladder. No abnormalities noted in surviving animals. Some evidence of the latter haemorrhaging was also seen in the low dose animals. No abnormalities were noticed in animals that survived until the end of the study.	Safepharm (1994b)

References to appendix 2

Bushy Run Research Center (1980): Butyl Cellosolve: Range finding toxicity studies. Report 43-99, date: 1980

Carpenter C.P., Keck G.A., Nair J.H., III, Pozzani U.C., Smyth H.F., Jr., and Weil C.S. (1956): The toxicity of butyl cellosolve solvent. A.M.A.archives of industrial health 14 (2), 114-131

Duprat P. and Gradiski D. (1979): Percutaneous toxicity of butyl Cellosolve (Ethylene glycol monobutyl ether). The Society of Occupational Medicine / IRCS medical science 7: 26.

Eastman Kodak (1981b): Comparative toxicity of nine glycol ethers: II acute dermal LD50. Study n° TX-81-38, date: 1981-07-01. Eastman Kodak. Eastman Kodak, Study report.

MB Research (1976) MB research laboratories (1976): Report on acute dermal toxicity in rabbits. Report n° MB75-988.

MIIR (1952) 1952): Butyl "Cellosolve". Acute and subacute toxicity. Evaluation of Red Blood Cell Fragility as a measure of initial response. Mellon Institute of Industrial Research Report n° 15-37, date: 1952.

Roudabush R.L., Terhaar C.J., Fassett D.W., and Dziuba S.P. (1965): Comparative Acute Effects of Some Chemicals on Skin of Rabbits and Guinea Pigs. Toxicology and Applied Pharmacology 7 (4), 559-565.

Safepharm (1994a): Ethyleneglycol monobutyl ether: Acute dermal toxicity test in the rabbit. Safepharm laboratories Report n° 13/605

Safepharm (1994b): Ethyleneglycol monobutylether: acute eye irritation test in the rabbit. Safepharm laboratories Report n° 13/564

Appendix 3. Additional inhalation toxicity studies not cited in the CLH document

Method/ guideline	Species, strain, sex, number	Dose levels, duration of exposure. Clinical and pathology findings	LC50 or other result	Reference
IHT according to Smyth (1962)	Rat (SD), M/F, 6-12	3 h: 2.25 mg/l. Mortality 0/12 7 h: 4.26 mg/l. Mortality 2/6. Clinical findings: Eyelid closure, slight salivation, accelerated respiration, haemorrhagic urine, apathy, crouch position, unstable gait, scrubby, contaminated fur, anaemic ears. Pathology: The animals that died during the test period showed the following findings: Heart: acute dilatation on the right side, sallow left heart ventricle. Lungs: moderate acute exhalation. Liver: clay-grey tone. Stomach: bloody ulcerations in the area of the glandular stomach. Intestine: hematinic contents. Surviving animals were without findings.	>4.6mg/L (119% of SVC) extrapolated to 4hrs exposure	BASF (1979)
Exposure to single limit dose concentration. Animals used for subsequent exposure before pathology	Dog (beagle), M, 2	7hrs: 1.96mg/L. No mortality. Clinical effects: salivation, otherwise no other outward clinical signs. No notable pathology findings	> 2.36mg/L (48% of SVC) extrapolated to 4hrs exposure	Dow (1974)
Exposure to single limit dose concentration. Animals used for subsequent exposure before pathology	Rabbit (no data), M, 4	7hrs: ~2.0mg/L. ~50% mortality taken over three experiments. Clinical effects: Poor co-ordination and loss of equilibrium. Pathology: Dead rabbits showed reddish ocular and nasal discharges and yellow discoloration of the sclera of the eyes. Kidneys were severely congested and haematuria was evident. Haemorrhagic ulcers were noted in the gastric mucosa as was mottled or yellow discoloration of the liver. One rabbit also showed slight congestion of the lungs and nasal turbinates. Pathology on surviving rabbits showed darkened or congested kidneys and mottled livers.	~ 2.36mg/L (48% of SVC) extrapolated to 4hrs exposure	Dow (1974)
Exposure to single limit dose concentration. Animals used for subsequent exposure before pathology	Guinea pig (no data), M, 8	7hrs: ~2.0mg/L. No mortality, no clinical effects, no notable pathology	> 2.36mg/L (48% of SVC) extrapolated to 4hrs exposure	Dow (1974)
IHT according to Smyth (1962)	Rat (not specified), M/F, 3	3 h: 1.44 mg/l. Mortality 0/12 8 h: 4.25 mg/l. Mortality 6/6	LC50 between 1.1 – 5.3mg/L extrapolated to 4 hrs	BASF (1968)

SVC calculated based on vapour pressure of 80Pa as cited in CLH document.

References to table

BASF (1968) Butylglykol: Ergebnis der Gerwerbetoxikologischen Vorpruefung, Report XVIII/354, BASF AG, Dept Toxicology, 29-11-1968

BASF (1979) Bericht ueber die Pruefung der akuten inhalationsgefahr (akutes inhalationsrisiko) von “Aethylenglykolmonoethylaether” an Sprague-Dawley Ratten, Report 78/789, BASF, 22-11-1979

Dow (1974) Inhalation toxicology studies on three samples of ethylene glycol monobutyl ether (Dowanol EB), n-butyl Oxitol-Shell USA, n-butyl Oxitol-Shell Europe, Study T-5-13-8-13, The Dow Chemical Company, Midland, USA. 7-10-1974

Appendix 4. Clinical and pathological findings in acute inhalation toxicity studies demonstrating species differences

LC50 (4 hr)	Duration	Clinical and pathology findings	Reference
Young animals: 4.9mg/L (recalculated) (127% of SVC) 1 year old animals: Young animals: 2.2mg/L (recalculated) (51% of SVC)	7 or 8hr original (rat)	Bloody urine and poor co-ordination. Hemoglobinuria observed.	Carpenter (1956), MIIR (1952)
2.6mg/L (recalculated) (67% of SVC)	4hr (rat)	In rats exposed to 867 ppm, loss of coordination was observed concurrently with laboured breathing and red fluid discharge around the urogenital region of both sexes. Respiration was shallow and rapid. In rats exposed to 523 ppm, coordination loss was also observed and red fluid discharge around the urogenital region. By the end of the observation period, a portion of the tail (distal end) of two males and two females appeared necrotic and/or had become severed. In rats exposed to 202 ppm, apart from a slight amount of dried red material observed on the tail the day following exposure, no other sign of toxicity was observed. Pathology: Animals that died had enlarged and discoloured kidneys with urinary bladder filled with red stained urine and intestinal contents appeared similar to the consistency and colour of tar. Effects were more noticeable in the high exposure group. Surviving animals showed no remarkable pathology.	Bushy Run (1980)
3.6mg/L extrapolated using Haber rule to 4 hours) (93% of SVC)	7 hrs originally (rat)	During exposure animals exhibited signs of lethargy. 7hrs exposure: Necrosis of the end of tails, blood in urine and paleness of eyes and feet. 3 hours exposure: Except for necrosis of the tail, the same symptoms than the 7 hr group were observed. 1 hour exposure: symptoms were limited to the presence of blood in urine and paleness eyes and feet. Recovery was full 2 days after exposures for surviving animals.	Shell (1982)
LC50 cannot be derived. Time for 1 rat death out of 5 at maximum attainable concentration (SVC) 1-3 hours.		No details reported	Klimisch (1988)
LC50 cannot be derived. Single 5hr exposure of rats to 11.7mg/L (2.7x SVC) produced 100% mortality		Haematuria, 50-65% reduction in haemoglobin level.	Gage (1970)
4.4mg/L extrapolated using Haber rule to 4 hours) (114% of SVC)	7 hours originally (mouse)	Dyspnoea was the major toxic symptom described moreover when doses were near the lethal level, severe haemoglobinuria was seen. Histopathology: Moderate to marked follicular phagocytosis and congestion of the cavernous veins were frequent findings in spleens of exposed animals. Focal necrosis and lymphoid hyperplasia was also seen in 3 surviving animals. Interstitial nephritis and typical pictures of bronchopneumonia was seen in a few animals.	Werner (1943)
7.65mg/L (198% of SVC)	7 hours original (guinea pig)	No details available (secondary source)	Tyler (1984)

not reached at 3.1-3.4mg/L 80-88% of SVC)	1 hour (guinea pig)	No adverse clinical observations recorded. Pathology: Findings in majority of both sexes but not all: Hyperinflation of lungs (3 males), dark red discoloration (punctate foci) of all lung lobes (1 male), dark red discolouration of the liver (1 female), abnormal pink fluid in gall bladder (1 female). Also a traumatized liver (median lobe appeared torn) and evidence of haemorrhage in the peritoneal cavity (1 female.) All findings regarded as incidental. Hyperinflation common in guinea pigs due to holding of breath when in contact with anaesthetic gas.	Gingell (1998), Dow (1994)

SVC calculated based on vapour pressure of 80Pa as cited in CLH document.

References to table

Bushy Run Research Center (1980a): Butyl Cellosolve: Four hour LC50 inhalation study on rats. Report 43-42, date: 1980. Bushy Run Research Center.

Carpenter C.P., Keck G.A., Nair J.H., III, Pozzani U.C., Smyth H.F., Jr., and Weil C.S. (1956): The toxicity of butyl cellosolve solvent. A.M.A.archives of industrial health 14 (2), 114-131.

Dow (1994) Ethylene glycol butyl ether: acute vapor inhalation study in guinea pigs, The Dow Chemical Company, Report 94N1392, dated 19-7-1994.

Gage J.C. (1970): The subacute inhalation toxicity of 109 industrial chemicals. British journal of industrial medicine 27 (1), 1-18.

Gingell R., Boatman R.J., and Lewis S. (1998): Acute toxicity of ethylene gylcol mono-n-butyl ether in the guinea pig. Food and Chemical Toxicology 36 (9-10), 825-829.

Klimisch H.J., Pauluhn J., Hollander H.W., Doe J.E., Clark D.G., and Cambridge G.W. (1988): Inhalation Hazard Test - Interlaboratory Trial with Oecd Method-403. Archives of Toxicology 61 (4), 318-320.)

MIIR (1952) Butyl "Cellosolve". Acute and subacute toxicity. Evaluation of Red Blood Cell Fragility as a measure of initial response. Mellon Institute of Industrial Research, Report n° 15-37,

Shell (1982) Shell Chemicals (1982): Test standardisation : inhalation toxicity of eight chemicals according to the OECD inhalation hazard test. Report n° RTB 2220.

Tyler T.R. (1984): Acute and subchronic toxicity of ethylene glycol monobutyl ether. Environmental Health Perspectives 57, 185-191.

Werner H.W., Mitchell J.L., Miller J.W., and von Oettingen W.F. (1943): The acute toxicity of vapours of several monoalkyl glycol ethers of ethylene glycol. The Journal of industrial hygiene and toxicology 25 (4), 157-163.