Committee for Risk Assessment

RAC

Opinion

Pursuant to Article 77(3)(c) of Regulation (EC) No 1907/2006 of the European Parliament and of the Council of 18 December 2006 concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals

A reassessment at the request of the European Commission of the new information on acute inhalation toxicity of 2-butoxyethanol; ethylene glycol monobutyl ether (EGBE)

EC Number: 203-905-0
CAS Number: 111-76-2

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Adopted

10 December 2020

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OPINION OF THE COMMITTEE FOR RISK ASSESSMENT

A reassessment
at the request of the European Commission of the new information on acute inhalation toxicity of 2-butoxyethanol; ethylene glycol monobutyl ether (EGBE)

Pursuant to Article 77(3)(c) of Regulation (EC) No 1907/2006 of the European Parliament and of the Council of 18 December 2006 concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals (the REACH Regulation), the Committee for Risk Assessment (RAC) has adopted an opinion on acute inhalation toxicity of EGBE.

I PROCESS FOR ADOPTION OF THE OPINION

Following a request from the European Commission on 12 May 2020, the Executive Director of ECHA in the mandate of 24 June 2020, requested RAC to prepare an opinion in relation to the acute inhalation toxicity of EGBE within 12 months following receipt of the request.

Rapporteur, appointed by RAC: **Bogusław Barański**

The draft opinion was made publicly available for targeted public consultation at https://echa.europa.eu/harmonised-classification-and-labelling-previous-targeted-consultations/-/substance-rev/25429/term on 3 August 2020. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by 7 September 2020.

The RAC opinion was adopted on **10 December 2020** by consensus of all members present and having the right to vote.

II OPINION OF RAC

Mandate

The Committee for Risk Assessment (RAC) has been requested by the European Commission, through the Executive Director of ECHA, to review the classification for acute toxicity by the inhalation route as adopted by RAC in its opinion of 14 September 2018 on 2-butoxyethanol (ethylene glycol monobutyl ether, EGBE).

Following adoption and publication of the RAC opinion, manufacturers of the substance provided information on the acute toxicity of 2-butoxyethanol via the inhalation route, additional to the information used by RAC to arrive at its

conclusion. This information comprised a study report from a new GLP-compliant acute toxicity inhalation study conducted according to OECD TG 433 (adapted), which was made available after the RAC opinion had been published; as well as the study report from another study in Guinea pigs, rabbits and dogs, which had been previously submitted in the context of the consultation on the CLH report. The latter study is briefly acknowledged in the CLH opinion (under the heading "Comments received during public consultation"), but the results were not further mentioned in the RAC opinion in the assessment and conclusions on acute toxicity.

In this re-evaluation, the Commission services requested consideration of the following aspects:

- A complete overview of all studies on acute inhalation toxicity available from the original CLH dossier, the public consultation and the submission accompanying this mandate, with an indication for each study to which extent it was taken into account in the RAC opinion.

III SCIENTIFIC GROUNDS FOR THE OPINION

Introduction

In the RAC opinion, adopted on 14 September 2018, on acute inhalation toxicity of 2-butoxyethanol (ethylene glycol monobutyl ether, EGBE), RAC took into account that the LC_{50}-values of 2-butoxyethanol in several acute inhalation toxicity studies in rats were in the range of 2.21 to 4.92 mg/L/4h, in one study in mice, 4.12 mg/L and in one study in Guinea pigs, 7.65 mg/L/4h. Thus, they were all within the classification criteria for Acute Tox. 3 of 2-10 mg/L. It is noted that due to low volatility and low vapour pressure of 2-butoxyethanol, the Guinea pigs could have been exposed, not just to pure vapour but to a mixture of vapour and mist, since the saturated vapour concentration at 20°C is 3.9 mg/L. Hence, the data on Guinea pigs alone were considered borderline between classification and no classification for acute inhalation toxicity. RAC therefore took all available studies in rats, mice and Guinea pigs into account, and concluded in their opinion that 2-butoxyethanol warrants classification as Acute Tox. 3; H331 (Toxic if inhaled), with an ATE of 3.0 mg/L (Table 3.1.2 of Regulation (EC) No 1272/2008).

RAC evaluation of acute inhalation toxicity

In the CLH report, results of 8 acute inhalation toxicity studies were included: 5 studies in rats, 1 study in mice and 2 studies in Guinea pigs. During the consultation of the original CLH report, 1 study in Guinea pigs, dogs and rabbits and 2 studies in rats were submitted, and during the consultation of the Article 77(3)(c) request, 1 additional study in Guinea pigs was submitted. The studies are summarised below.

Summary table of animal studies on acute inhalation toxicity

Studies included in the original CLH report		
Material and methods	**Results**	**Reference**
Similar to OECD TG 403 **Rat,** strain not given, 6 females/group, and groups of older rats: 13 males/group, 23 females/group 2-butoxyethanol (CAS: 111-76-2), vapour (passing air at 2.5 L per minute through a fritted glass disc immersed in 50 mL of the liquid held at room temperature) No further details available.	Mortality: 800 ppm, 8h: 3/6 young females 800 ppm, 4h: 0/6 young females 500 ppm, 8h: 0/6 young females 500 ppm, 4h: 1/6 females 375 ppm, 7h: 11/13 older males 375 ppm, 7h: 23/23 older females LC_{50}: Young female rats: 800 ppm, 8h (corresponding to **4.92 mg/L/4h**) Older male and female rats: 375 ppm, 7h (corresponding to **2.21 mg/L/4h**)	Carpenter *et al*., 1956 and Mellon Institute of Industrial Research, 1952
Similar to OECD TG 403. **Rat**, F344, 6/sex/dose 2-butoxyethanol, purity 99.4%, vapour 867, 523 or 202 ppm, 4h, whole body exposure, 14-d post exposure observation period	Mortality: 867 ppm, m+f: 6/6 on Day 2 523 ppm, m: 2/6, f: 3/6 during 14-d post exposure period 202 ppm, m+f: 0/6 Necropsy, died animals: enlarged and discoloured kidneys, urinary bladder filled with red stained urine LC_{50}: 523 ppm (corresponding to **2.56 mg/L/4h**) (Calculated: 486 ppm = 2.37 mg/L (males); 450 ppm = 2.2 mg/L (females))	Bushy Run Research Center, 1980a
OECD TG 403 (validation study, ring study) **Rat**, Wistar, 3/sex/group 2-butoxyethanol (CAS: 111-76-2), purity 99%, saturated vapour 617 ppm (3 mg/L) for 7h, 3h or 1h, whole body exposure, measurements in the exposure chamber: 750-910 ppm	Mortality: 7h, m: 1/3, f: 3/3 3h, m: 0/3, f: 1/3 1h, m/f: 0/3 Lethargy, necrosis of the tail and haemolysis was seen. LC_{50}: 617 ppm, 7h (corresponding to **3.63 mg/L/4h**)	Shell Chemicals, 1982
OECD TG 403, 1981 (interlaboratory trial, ring test data) **Rat,** SD, 5/sex/group 2-butoxyethanol (CAS: 111-76-2), purity 99%, saturated vapour Nominal concentration: 3.1 to 4.1 mg/L (mean: 3.3-3.7 mg/L); estimated concentration 4.9 mg/L; head/nose exposure (1 lab), whole body exposure (5 labs: animals sat in cages in chamber or in tubes)	The 0-lethality time (LT_0, for which at least one death was found) was 3h for 5 laboratories and 1h for 1 laboratory.	Klimisch *et al*., 1988

Similar to OECD TG 403. **Rat**, 4/sex/group 2-butoxyethanol (CAS: 111-76-2), purity commercial grade, aerosol 2400 ppm (13 mg/L) for 5h, whole body exposure	Clinical signs: comatose state, haematuria Blood: Hb concentration 35 to 50 % of the normal Mortality: m/f: 4/4 on Day 2 LC_{100}: 2400 ppm, 5h (corresponding to **12.62 mg/L/4h**)	Gage, 1970
No guideline followed. Acute study that pre-dates guidelines. Principles of current guideline methods followed, with more doses examined, increasing statistical precision of result. Some information on study protocol missing from publication. **Mouse**, strain and number of animals used not given 2-butoxyethanol (CAS: 111-76-2), purity commercial grade, vapour	Clinical signs: Dyspnoea, haemoglobinuria and death were noted in the 4th week after exposure Necropsy: Findings in spleen, liver, lungs, and kidneys LC_{50}: 700 ppm, 7h (corresponding to **4.12 mg/L/4h**)	Werner *et al.*, 1943 cited in Carpenter *et al.*, 1956
LC_{50}-test, no guideline followed **Guinea pig,** strain unspecified, adult 2-butoxyethanol (CAS: 111-76-2), "Substantially saturated vapour" 1300 ppm for 7h, whole body exposure, 14-d post exposure period	LC_{50}: 1300 ppm, 7h (corresponding to **7.65 mg/L/4h**)	Mellon Institute of Industrial Research, 1943 cited in Tyler, 1984
Similar to OECD TG 403; deviation in exposure time, only 1h was used **Guinea pig,** Dunkin Hartley albino (5 weeks of age; 400-500 g), 5 animals/sex 2-butoxyethanol (CAS: 111-76-2), purity 99.87%, vapour 633±14.2 ppm (males) and 691±37.6 ppm (females) for 1h, whole body exposure, 14-d post exposure period	No mortalities. LC_{50}: **> 633 ppm** (males) **> 691 ppm** (females) [NB: LC_0 indicated as >3.1-3.4 mg/L in Glycol Ethers Consortium comments during the consultation of the Article 77(3)(c) request.]	Dow Chemicals Company, 1994 also referred to as Gingell *et al.*, 1998
Studies submitted during the consultation of the original CLH report		
Material and methods	**Results**	**Reference**
IHT according to Smyth (1962) **Rat**, SD, 6-12 males/females	Mortality: 3h, 2.25 mg/L: 0/12 7h, 4.26 mg/L: 2/6. Clinical findings: Eyelid closure, slight salivation, accelerated respiration, haemorrhagic urine, apathy, crouch position, unstable gait, scrubby, contaminated fur, anaemic ears. Pathology findings in animals that died during the test	BASF, 1979

<table>
<tr><td></td><td>period:

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.

LC_{50}:

> 4.6 mg/L/4h [NB: This was indicated to be 119% of the saturated vapour concentration; SVC]</td><td></td></tr>
<tr><td>IHT according to Smyth (1962)

Rat, not specified, 3 males/females</td><td>Mortality:

3h, 1.44 mg/L: 0/12

8h, 4.25 mg/L: 6/6

LC_{50}:

1.1-5.3 mg/L/4h</td><td>BASF, 1968</td></tr>
<tr><td>Acute inhalation toxicity study/LC_{50}-test. No guideline, non-GLP

Three groups of animals; each consisting of 2 male Beagle dogs, 4 male albino rabbits and 8 male Guinea pigs, were exposed to vapour of 3 different purified EGBE samples (Dowanol EB; BO-USA, BO-Europe). A fourth group served as controls

1 m^3 chamber under dynamic conditions, 27-27.5°C

<u>Study protocol:</u>

1st experiment: Nominal vapour concentration of 400 ppm, exposure duration 7h, observed for 7 days post-exposure. Thereafter, 4 out of 8 Guinea pigs and all rabbits from each group were sacrificed for gross pathological examination, while each dog was given a thorough physical examination.

2nd experiment: One week after the first exposure; the 2 dogs and the 4 remaining Guinea pigs from the 1st experiment were re-used, and 4 not previously used Guinea pigs and rabbits were added. Exposed to the same samples as in the 1st experiment, under identical conditions. Thereafter, exposed to stress in the form of loud noise. Observed for 7 days post exposure.

The rabbits and Guinea pigs from 2 of the groups were sacrificed for gross pathological examinations, while the dogs were used for other studies.

A 3rd experiment with repeat exposure was also performed, 400</td><td><u>Dogs</u>: 1st and 2nd experiment: Salivation during exposure. No adverse effects during post-exposure observations. No mortality. No urinalysis taken in conjunction with the study.

LC_{50}:

> 2.36 mg/L/4h [see note further down]

<u>Rabbits</u>: 1st experiment: No adverse effects during or directly after exposure.

Within 8-16h after exposure, 3/4 rabbits in one of the exposure groups (Dowanol EB) were found in moribund condition (poor coordination and loss of equilibrium); followed by death within 1-3 days.

1/4 rabbits died in the BO-USA sample group and 0/4 rabbits died in the BO-Europe sample group.

2nd experiment (with stress), similar signs of toxicity were seen. 3/4 rabbits in the Dowanol EB and BO-Europe groups died within 1-3 days, while 0/4 of the rabbits died in the BO-USA group.

<table><tr><th>Sample</th><th>1st experiment</th><th>2nd experiment</th></tr><tr><td>Dowanol EB</td><td>3/4</td><td>3/4</td></tr><tr><td>BO-USA</td><td>1/4</td><td>0/4</td></tr><tr><td>BO-Europe</td><td>0/4</td><td>3/4</td></tr><tr><td>Total:</td><td>4/12</td><td>6/12</td></tr></table>
LC_{50}:

Ca. 2.36 mg/L/4h over 3 experiments [see note further down]

<u>Guinea pigs</u>: 1st and 2nd experiment: No adverse effects after single 7h exposure to any of the three samples.

LC_{50}:

> 2.36 mg/L/4h [see note further down]</td><td>Dow Chemicals Company, 1974</td></tr>
</table>

ppm, 7h/day for 5 days. [NB: The study was performed to address contradictory results from previous studies, where CNS effects and death in dogs (100 ppm, 2x7h; or 400 ppm, 1x7h), and death in Guinea pigs (100 ppm, 2h), were seen after exposure to production grade EGBE in two reports from Shell Research Ltd, while this had not been seen in other studies. It was suggested that the deaths of the dogs and Guinea pigs in the Shell reports might have been due to external stimuli.]	Conclusion: Based on the study, it was concluded that the vapours of the 3 samples had similar toxicity, that rabbits are more susceptible than dogs and Guinea pigs and that the results are contradictory to the Shell reports (discrepancies proposed to be due to external stimuli and impurity profile). It was also concluded that 400 ppm may be harmful to humans, but that there was no support for lowering the threshold limit value of 50 ppm. [NB: The LC_{50}-values were indicated in Glycol Ethers Consortium comments during the consultation of the Article 77(3)(c) request. 2.36 mg/L/4h was indicated to be 48% of the SVC.]	
Studies submitted during the consultation of the Article 77(3)(c) request		
Material and methods	**Results**	**Reference**
Performed as an adaption of OECD TG 433, GLP **Guinea pig**, Dunkin Hartley 1 test group, 6 males and 6 females Single inhalation exposure, 2.25 mg/L, 4h, nose-only exposure 20-24°C, 40-70% humidity with some deviations throughout the study (not considered to affect study outcome) 14-d post exposure observation Clinical condition, body weight, haematology (peripheral blood), urinalysis, organ weight and macroscopic examinations done. The mean achieved exposure level was 2.25 mg/L (=75% of the targeted concentration; 3 mg/L); this was considered the maximum achievable stable vapour only concentration under the study conditions.	One male was sacrificed on day 5 due to welfare reasons (irregular and laboured breathing, dull eyes, impaired locomotion with reduced body tone, flattened posture and brown staining around the anus; body weight loss on days 4 and 5. Macro-pathology findings were seen in GI-tract, colon and liver. This was considered to be stress related. Shallow breathing observed in 2/6 males and 2/6 females on return to home cage, resolved in all animals by 2 hours after exposure. Group mean body weight loss on day 2, but comparable to day 1 by the next weighing occasion on day 4. No test-item related effects on haematology or urinalysis parameters. No test-item related macroscopic lesions by day 14. LC_{50}: **> 2.25 mg/L/4h** The targeted concentration of 3 mg/L was chosen based on available information regarding toxicity and physicochemical properties of the substance in Guinea pigs. In an OECD TG 403 study (Dow Chemicals Company, 1994; Gingell *et al*., 1998), whole body exposure, males and females were dosed for 1h at 3.4 (males) and 3.1 (females) mg/L, considered to be the maximum achievable vapour concentration. The targeted concentration was considered to be likely non-toxic in Guinea pigs.	Covance CRS Limited, 2019

Comments received during the consultation for the Article 77(3)(c) request

During the consultation, the following comments were received.

One industrial organisation disagreed with the proposed classification as Acute Tox. 3; H331: Toxic if inhaled. According to this organisation, the key toxic effect of 2-butoxyethanol (or more specifically its main metabolite, butoxyacetic acid; BAA) is that it rapidly causes haemolysis following acute exposure. They argued that this effect varies significantly between species with rats, mice and rabbits being notably

sensitive to the effect, whereas humans and Guinea pigs are very resistant. Taking into account that the Guinea pig appears to have a sensitivity to the leading toxic effect (haemolysis mediated through the metabolite butoxy acetic acid) similar to that of humans, this industrial organisation considered that only the available Guinea pig data should be considered when determining the classification.

In support of their argument, the industrial organisation submitted results of a new study (Covance CRS Limited, 2019). In this study, it was concluded that the LC_{50} for Guinea pigs is above 2.25 mg/L. In addition, the results of other acute inhalation toxicity studies in Guinea pigs and in dogs were presented (Gingell *et al*., 1998; Dow Chemical Company, 1974). These studies were included in the RAC opinion from 2018 and showed no mortality of Guinea pigs or of dogs exposed via the inhalation route for 1h or up to 7h to 2-butoxyethanol at concentrations in the range 408-633/691 (m/f) ppm (approx. 2-3.1 mg/L).

The industrial organisation also provided a calculation of the saturated vapour concentration of 2-butoxyethanol, derived from the vapour pressure at a given temperature. After taking into account the relevant physico-chemical parameters, it came to a theoretical value of 3.9 mg/L at a temperature of 20°C. The reported temperature in the animal husbandry monitored during that study (Covance CRS Limited, 2019), was in the range of 20-24°C. The mean temperature of air with 2-butoxyethanol inhaled through snout by Guinea pigs in the study was 22.4±0.4°C.

In summary, the industrial organisation concluded that classification for acute inhalation toxicity of 2-butoxyethanol is not warranted. This conclusion was based on results of two studies in Guinea pigs (Dow Chemical Company, 1974; Covance CRS Limited, 2019), demonstrating that exposure of Guinea pigs for 4h to the maximum practically attained vapour concentration (2.25-2.5 mg/L, being 60-65 % of the saturated vapour concentration at 20°C) produced no substance-related adverse effects in the exposed animals. This conclusion was, according to the organisation, further supported by results of the Dow study in which no adverse effects were observed in Guinea pigs exposed to 2-butoxyethanol by inhalation for 1h at a concentration of 633 ppm (3.1 mg/L) (males) or 691 ppm (3.4 mg/L) (females) (Gingell *et al*., 1998; Dow Chemical Company, 1974), or in dogs exposed for 7h at 1.96 mg/L to 2-butoxyethanol (Dow Chemical Company, 1974).

In the opinion of the industrial organisation, the results of the acute inhalation toxicity studies in rats, mice and rabbits should not be used for the classification for acute inhalation toxicity of 2-butoxyethanol, since rodents and rabbits are particularly susceptible to haemolysis caused by this substance whilst humans and Guinea pigs are remarkably resistant. This difference in the susceptibility between rats and humans for the haemolytic effect of BAA, the metabolite of 2-butoxyethanol, is internationally recognised. In support of the resistance of humans to haemolysis caused by 2-butoxyethanol, the industrial organisation referred to several cases of acute human intoxication with 2-butoxyethanol where there was no evidence of haemolytic activity (Bauer, 1992; Butera, 1996; Burkhart, 1998; Gijsenbergh, 1989; Gualtieri, 1995, 2003; Hung, 2010; McKinney, 2000; Rambourg-Schepens, 1988).

NOTE: RAC's response to the industry comments is included in the section 'Assessment and comparison with the classification criteria'.

Two Member state competent authorities (MSCAs) disagreed with the above opinion by the industrial organisation and supported RAC's original opinion adopted in 2018, to classify 2-butoxyethanol as Acute Tox. 3; H331: Toxic if inhaled, with an ATE of 3.0 mg/L/4h.

One of the MSCAs noted that they had already commented on the newly presented study (Covance CRS Limited, 2019) in December 2019, as a follow-up to the 32nd CARACAL meeting, and had concluded based on this study, that no new assessment by RAC would be necessary. They further noted that in the study by Dow (Dow Chemical Company, 1974) it was shown again that Guinea pigs and dogs are significantly less sensitive to single inhalation exposure to 2-butoxyethanol compared to rabbits (see below). This study was included in the original opinion (see section "Comments received during public consultation" in the RAC opinion of 2018). The MSCA argued that the results of the study do not contradict classification of 2-butoxyethanol as Acute Tox. 3, H331, since, when taking all available studies into account, classification as Acute Tox. 3 via the inhalation route is justified.

The study by Dow (Dow Chemical Company, 1994) was also available to RAC in 2018 and was included in the assessment (in the RAC opinion and in the CLH dossier, where it was referred to as Gingell *et al*., 1998). The MSCA also argued that taken together, the three studies referred to above do not contradict the conclusion in the RAC opinion of 2018; therefore, the RAC opinion to classify 2-butoxyethanol as Acute Tox. 3 (H331) is still supported. The MSCA further noted that in its opinion of 2018, RAC was well aware that the LC_{50} for Guinea pigs in the existing key study might not have been based on exposure to pure vapour, as the highest tested concentration was well above the saturated vapour concentration of butoxyethanol at 20°C of 4.4 mg/L (test concentration: 7.65 mg/L/4 h). RAC was also aware that the older data on Guinea pigs alone were thus borderline between classification and no classification for acute inhalation toxicity. Although the saturated vapour concentration of 2-butoxyethanol is in fact higher, it is claimed in the newly presented Covance (2019) study that the tested concentration of 2.25 mg/L/4h was the highest achievable concentration for pure vapour and that no aerosol droplets were observed during the exposure. However, the MSCA noted that in one of the older acute inhalation toxicity studies with 2-butoxyethanol in Guinea pigs (Gingell *et al*., 1998), the test atmosphere was also checked to ensure the absence of aerosol particles and concentrations of up to 691 ppm (3.4 mg/L) were tested. This indicates that the testing of higher pure vapour concentrations than 2.25 mg/L might very well be feasible.

As the new study results do not contradict the conclusion made by RAC and do not enable the setting of a different ATE-value than that established by RAC, this MSCA considered that "RAC would not be in a position to reclassify the substance" based on the new data provided by the industry. Instead, conducting an additional acute inhalation toxicity study was by this MSCA considered inappropriate in light of animal welfare and the 3R strategy, since a large data set on this endpoint was already available.

A second MSCA noted that even if the recent study provided is of good quality, they were of the opinion that the RAC opinion of 2018 was still appropriate for acute

inhalation toxicity (Acute Tox. 3; H331: Toxic if inhaled), based on the lowest ATE value available, *i.e.* the lowest ATE in the most sensitive appropriate species tested, as recommended in the Guidance on the application of the CLP criteria. In their view, the justification for using only Guinea pig data for classification, based on the similarity in sensitivity between humans and Guinea pigs for haemolytic effects, is not substantiated. Moreover, they noted that the high interindividual variation in toxicokinetics of the substance in humans must be taken into account.

Assessment and comparison with the classification criteria

In reviewing the classification for acute toxicity by the inhalation route, as requested by the European Commission, RAC has taken into account the results of all studies submitted by the DS in the CLH dossier, relevant data submitted during the consultation of the original CLH proposal, as well as the results of a study provided by Industry, accompanying this request of the European Commission. These data are summarised below for each tested species.

Humans

All data evaluated on acute inhalation toxicity in humans were provided in the CLH dossier.

No consistent effects on the lungs or the heart were observed in 4 male volunteers exposed for 2h, at a concentration of 0.24 mg/L (Johanson, 1986), and no overt signs of toxicity were observed in 7 male volunteers exposed to 0.24 mg/L for 2h (Johanson and Bowman, 1991).

Exposure of 2 men to 113 ppm (0.55 mg/L) for 4h, and 1 year later exposure of the same 2 men and 1 female to 195 ppm (0.95 mg/L) for two 4h periods separated by a 30-min interval, induced irritation to the eyes, nose and throat, a disturbance of taste, a slight increase in nasal mucous discharge and headache. One male and the female excreted considerable amounts of BAA following the two 4h periods after exposure, but the other male excreted only trace amount of BAA during the same period. The female subject, who excreted the largest amount of BAA, reacted most adversely to the exposure, and acquired a headache which lasted about 24h. Unlike rats, no effects were seen in the erythrocyte fragility test in these subjects. No evidence of changes from pre-exposure values in erythrocyte fragility, blood pressure, pulse rate or urinary levels of glucose or albumin were seen. No adverse effects were seen in haematology at either exposure concentration (Dow Chemical Company, 1955; Carpenter *et al*., 1956; European Union Risk Assessment Report (EU RAR) for 2-butoxyethanol, 2006).

RAC notes that inhalation exposure to 2-butoxyethanol at a concentration of 0.95 mg/L for 2 x 4h, with a break of 30 minutes in between, induced clear local irritative effects in the eyes and upper part of the respiratory tract, some effect on taste receptors, and in one person an effect on the central nervous system leading to long lasting headache. It is noted that concentrations higher than 0.95 mg/L, still well

below the saturated vapour concentrations (being in the range 3.9-5.6 mg/L at temperatures of 20-25°C), could lead to toxic effects that are much more severe.

Rats and mice

According to data provided in CLH dossier, the LC_{50} values in rats were in the range of 2.2-4.92 mg/L/4h (Carpenter *et al*., 1956; Mellon Institute of Industrial Research, 1952; Bushy Run Research Center, 1980a; Shell Chemicals, 1982). The LC_{50}-value in mice was 4.12 mg/L/4h (Werner *et al*., 1943, cited by Carpenter *et al*., 1956).

In one study (BASF, 1979) exposure to 2-butoxyethanol at a concentration of 4.26 mg/L of rats for 7h resulted in death in 2 out of 6 animals, and exposure to 2-butoxyethanol at a concentration of 2.25 mg/L for 3h did not cause mortality among 12 exposed rats. The LC_{50} for rats extrapolated to 4h exposure was above 4.6 mg/L/4h. In the second study (BASF, 1968), no mortality was observed among 12 rats exposed for 3h at a concentration of 1.44 mg/L/3h, while 6 out of 6 rats died due to exposure to 2-butoxyethanol at a concentration of 4.25 mg/L for 8h. The LC_{50} for rats extrapolated to 4h exposure was calculated to be between 1.1 and 5.3 mg/L/4h.

It is noted that in most studies, the LC_{50} for rats was below the indicated saturated vapour concentrations: 2.2 mg/L for older male and female rats in the study of Carpenter *et al*. (1956) and Mellon Institute of Industrial Research (1952); 2.37 mg/L for male rats and 2.2 mg/L for female rats in the study of Bushy Run Research Center (1980a), and 3.63 mg/L/4h in the study of Shell Chemicals (1982).

On the other hand, it is also noted that these studies were conducted well before the introduction of OECD test guidelines and GLP. However, the fact that they were carried out independently, that the results are comparable, and that all studies resulted in LC_{50}-values below the estimated saturated vapour pressure, meant that in the view of RAC they should be taken into account and used for classification.

Dogs

No acute inhalation toxicity study in dogs was submitted in the CLH dossier. During the consultation on the original CLH dossier, results of one study in dogs were submitted (Dow Chemical Company, 1974). In this study, 2 male dogs were exposed for 7h to 2-butoxyethanol at a concentration of 1.96 mg/L with 7h exposure (LC_{50} >2,36 mg/L, extrapolated to 4h exposure). No mortality or clinical effects other than increased salivation were reported. Since the number of animals and concentration used in this study were low, well below the saturated vapour concentration, the results of the study are considered inconclusive for evaluation of acute inhalation toxicity of 2-butoxyethanol.

Rabbits

No acute inhalation toxicity study in rabbits was included in the CLH dossier. During the original consultation, the results of one study in rabbits were submitted (Dow Chemical Company, 1974). In this study, in which 4 male rabbits were exposed for 7h to 2-butoxyethanol at a concentration of approximately 2 mg/L, a 50% mortality

was seen and the LC_{50} of 2-butoxyethanol for rabbits extrapolated to 4h exposure was calculated based on Haber's rule to be approximately 2.36 mg/L/4h. Poor co-ordination and loss of equilibrium was observed in the exposed rabbits. Pathological examination of 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. The study provides sufficient evidence that 2-butoxyethanol is toxic to and results in mortality in rabbits by inhalation at a concentration below the saturated vapour concentration, and the results suggest that for this species the criteria for classification to Acute Tox. 3: 2.0 mg/L < ATE ≤ 10.0 mg/L are fulfilled.

Guinea pigs

As reported by the DS in CLH dossier, the LC_{50} in Guinea pigs was 7.65 mg/L/4h in one study (Mellon Institute of Industrial Research, 1943 cited by Tyler, 1984). Since the concentration of 2-butoxyethanol in this study was above the saturated vapour pressure, the actual exposure was at least in part due to an aerosol of 2-butoxyethanol. Hence, the results cannot be used for classification for acute inhalation toxicity of vapours.

No mortality was observed in a second study using a lower concentration of 633-691 ppm (approx. 3.1-3.4 mg/L) for 1h whole body exposure (Dow Chemicals Company, 1994; Gingell *et al*., 1998). It is noted that the exposure time was 1h, and not 4h as required for setting an LC_{50}. The study is thus considered inconclusive for assessment of acute inhalation toxicity of 2-butoxyethanol.

During the consultation on the original CLH dossier, results of one study in Guinea pigs were submitted (Dow Chemical Company, 1974). In this study, 8 male Guinea pigs were exposed for 7h to 2-butoxyethanol at a concentration of approximately 2 mg/L. No mortality nor any clinical effects were observed. No notable pathology was reported. This study was not aimed at determining an LC_{50} for Guinea pigs but performed to address contradictory results from previous studies. Since higher concentrations were not tested, the study is considered inconclusive for evaluation of acute inhalation toxicity of 2-butoxyethanol for Guinea pigs.

During the consultation of the Article 77(3)(c) request, manufacturers of the substance provided additional information, not available earlier to RAC, on the acute toxicity of 2-butoxyethanol via the inhalation route (Covance CRS Limited, 2019). This new acute inhalation toxicity study in Guinea pigs was carried out according to OECD TG 433 (2018) and is GLP-compliant. During the study, 6 male and 6 female Dunkin Hartley Guinea pigs were administered 2-butoxyethanol for 4h by inhalation (by snout only) at a concentration of 2.25 mg/L, which was 75% of the planned concentration of 3 mg/L. This was considered by the authors of the study to be the highest technically achievable vapour-only concentration. Based on results of this study it was concluded that the LC_{50} of 2-butoxyethanol for Guinea pigs is above 2.25 mg/L/4h.

In the study report of the Covance CRS Limited (2019) study, it is not further described why it was not technically possible to expose the animals to higher concentrations of 2-butoxyethanol vapour, although the authors were aware of the vapour dosing system of another acceptable study from 1994 (Dow Chemicals Company, 1994, also referred as a Gingell *et al*., 1998). In the Dow Chemicals Company (1994) study, higher concentrations were achieved, up to 3.9 mg/L, but the Guinea pigs were exposed only for 1h. Since the concentration of 2.25 mg/L used in the Covance CRS Limited study (2019) was considerably lower than the saturated vapour concentration at 22°C (approximately 4.75 mg/L), as well as the concentration of 3.9 mg/L achieved in the Dow Chemicals Company (1994) study, there is high uncertainty regarding whether this study may be considered as sufficient evidence that the saturated vapour concentration of 2-butoxyethanol is not lethal for Guinea pigs after 4h exposure.

Saturated vapour concentration of 2-butoxyethanol

The concentration of 2-butoxyethanol vapour in air in the inhalation toxicity studies depends on the temperature and the system used for generating vapour.

The saturated vapour pressure of 2-butoxyethanol at a temperature of 20°C, as provided in the CLH report, is equal to 80Pa. Using that value, the industrial organisation stated in their comments during the consultation, that the maximum saturated vapour concentration is equal to 3.9 mg/L. This is the theoretical maximum concentration of vapour that can be reached at 20°C. However, the saturated vapour pressure of 2-butoxyethanol increases quickly with temperature. At a temperature of 25°C, the saturated vapour pressure of 2-butoxyethanol is equal to 117Pa (0.88 mmHg) (Final Report on the Safety Assessment of Butoxyethanol, 1996); thus, at a temperature of 25°C the theoretical maximum concentration of 2-butoxyethanol vapour would be ca. 5.6 mg/L.

According to the OECD Guidance Document on Inhalation Toxicity Studies (Series on Testing and Assessment, No. 39, Second Edition, 6 July 2018), the chamber temperature should be maintained at 22±3°C. Within the temperature range 20-25°C, which is consistent with this requirement, the saturated vapour concentration of 2-butoxyethanol inhaled by the animals could be in a range of 3.9-5.6 mg/L. However, in the dynamic conditions of exposure it is not possible to achieve and maintain the saturated vapour concentration in the chamber. In line with the OECD Guidance Document on Inhalation Toxicity Studies for vapour atmospheres, the maximum attainable concentration depends on the vapour saturation concentration of a test chemical under test conditions. The maximum attainable concentration is generally defined such, that any change in the equipment and/or further increase of the nominal test chemical supply rate into the inhalation exposure system, does not increase the concentration.

In a few acute inhalation toxicity studies on 2-butoxethanol, the system for generation of vapour of 2-butoxyethanol was described, and the temperature of the air inhaled by exposed animals was provided.

In the Carpenter *et al*. study (1956) the mixture of 2-butoxyethanol vapour with air was produced by passing air at 2.5 L/minute through a fritted glass disc immersed in

50 mL of the liquid held at room temperature. No further details were provided. The highest concentration achieved with this method was 800 ppm, which is approx. 3.9 mg/L. The temperature of air in the inhalation chamber was not given. However, based on the similar experiment in the Dow Chemical Company study (1974), where the air temperature in the inhalation chamber was 27-27.5°C, and the experiment in the Dow Chemicals Company study (1994), where the temperature of air in the chamber was 24°C, an air temperature in the chamber of 25°C could be assumed. The concentration of 3.9 mg/L would then be equal to ca. 70% of the saturated vapour concentration at this temperature (3.9 x100/5.6=69.6%).

In the Dow Chemical Company study (1974), the vapour of 2-butoxyethanol was generated by metering the liquid substance at the rate of 0.76–0.81 mL/min with the precision syringe into a temperature regulated vaporisation flask maintained at 200°C. The vapours from the flask were caried into the chamber by filtered room air which was drawn through the flask at 430-480 L/min. The air temperature in the exposure chamber was 27-27.5°C. The study authors planned to achieve a nominal vapour concentration of 400 ppm (1.95 mg/L) in the air of the exposure chambers. The planned concentration was confirmed to have been achieved in all three chambers (with small deviations) by measurement using an infrared spectrophotometer.

In the Dow Chemical Company study (1994; also referred as a Gingell *et al*., 1998), filtered, compressed air was passed through two 125 mL gas washing bottles containing 84.2 g and 75.5 g of 2-butoxyethanol, placed in a water bath to maintain air at approximately 26°C. Before entering a 120 L whole body exposure chamber, a mixture of 2-butoxyethanol vapour with air was passed through an empty 2L mixing flask. The temperature of air in the chamber was 24°C and no aerosol was detected in the chamber. The concentration of 2-butoxyethanol in the chamber before exposure of animals was equal to 791 ppm (3.9 mg/L) corresponding to 69.6% of the calculated saturated vapour concentration at 25°C (1154.7 ppm; 5.6 mg/L). However, after inserting animals into a chamber, the concentration of 2-butoxyethanol was reduced to 633 ppm (3.1 mg/L) for males and 691 ppm (3.4 mg/L) for females. According to the authors of the study, the reduced concentrations were due to probable losses from adsorption of 2-butoxyethanol on the fur of the animals and solubility of the substance in urine exerted by animals.

In the study by Covance CRS Limited (2019), vapour of 2-butoxyethanol was generated in a glass retort vessel initially primed with 40 mL of the test item, and after initiation the test item was supplied to the evaporation vessel via a feed line from a syringe at a constant rate. The air with vapour of 2-butoxyethanol at the rate of 9L/min was supplied to a 30 L exposure chamber equipped with 20 ports nose-only for exposure of Guinea pigs.

The temperature of the air inhaled by Guinea pigs in the Covance CRS Limited study was 22.4±0.4°C and the measured concentration of 2-butoxyethanol was 2.25 mg/L. The target concentration of 3 mg/L was selected following review of available information on toxicity and physicochemical properties of the test item in Guinea pigs. It was indicated that in the previous OECD TG 403 study (Dow Chemicals Company study, 1994, Gingell *et al*., 1998), no adverse effects were observed in male and females Guinea pigs after 1h whole body inhalation exposure to 2-

butoxyethanol at vapour concentrations of 3.4 and 3.1 mg/L. Hence, it was concluded that the target concentration of 3 mg/L was likely to be non-toxic to Guinea pigs (Covance CRS Limited, 2019).

Prior to animal exposure, special trials were conducted to establish the inhalation exposure system operating conditions required to generate the target chamber concentration. Data from these trials were, however, not reported in the available study report (Covance CRS Limited, 2019). Thus, it is not known how various operational conditions such as e.g. the amount of 2-butoxyethanol provided into the retort vessel for evaporation in a unit of time, the temperature of a water bath warming up a retort vessel used for evaporation, or other parameters of the dosing system, could influence the final concentration of vapor in the chamber (2.25 mg/L; approximately 47% of the saturated vapour concentration at a temperature of 22°C; 3.9 mg/L at 20°C + 5.6 mg/L at 25°C/2 = 4.75 mg/L).

Therefore, it was not demonstrated that the final concentration of 2.25 mg/L can be considered as the maximum attainable concentration according to OECD Guidance Document on Inhalation Toxicity Studies (Series on Testing and Assessment No. 39, 6 July 2018). In addition, the concentration used in the Covance CRS Limited study (2.25 mg/L) was much lower than the practically achievable concentration of 2-butoxyethanol of 3.9 mg/L obtained in the exposure generation system in the Dow Chemicals Company study (1994)

Comparison with classification criteria

It has been assumed that the sensitivity to acute systemic toxicity of 2-butoxyethanol varies between different species. In the summary on haematotoxicity from the EU RAR (2006), it was concluded that some species (rats, mice, hamsters, baboons) were very sensitive to 2-butoxyethanol- (or more specifically, BAA-induced) haemolysis, whereas other species (dog, Guinea pig, pig, cat, rabbit and humans) were resistant to these effects. However, the difference in sensitivity to haemolysis caused by 2-butoxyethanol is based on mechanistic studies *in vitro* and *in vivo*. These studies show differences in the resistance of erythrocytes of different species to haemolysis, and different profiles of toxic effects and metabolism of this substance in rodents, humans and Guinea pigs. However, this is not reflected in differences in sensitivity of these species to acute toxicity of 2-butoxyethanol.

It is noted by RAC, that in species resistant to haemolysis induced by 2-butoxyethanol, other modes of action might be essential in exerting acute toxicity. As pointed out by the industrial organisation during the consultation of the request according to Article 77(3)(c), in acute oral poisonings with 2-butoxyethanol in humans, the primary toxic effect was metabolic acidosis, likely resulting from high concentrations of BAA in the blood, although no haemolysis was reported in these cases (Bauer *et al*., 1992; Butera *et al*., 1996; Burkhart & Donovan, 1998; Gijsenbergh *et al*., 1989; Gualtieri *et al*., 1995, 2003; Hung, 2010; McKinney *et al*., 2000; Rambourg-Schepens *et al*., 1988).

It is also noted that estimated doses of 2-butoxyethanol inducing severe acute poisoning in adult humans, requiring hospital treatment, were in a range of 500-1250

mg 2-butoxyethanol/kg bw; thus, relatively low, except in one case in which the estimated dose was 4.5 g 2-butoxyethanol/kg bw (Table 10 of the CLH report).

In the Bauer *et al*. case study (1992), coma, tachycardia, metabolic acidosis, hypoxemia, pulmonary oedema and acute respiratory distress syndrome (ARDS), without haemolytic anaemia and thrombopenia, were observed in 53-year old man after acute poisoning with a mixture containing 2-butoxyethanol. The estimated oral dose was 650-750 mg 2-butoxyethanol/kg bw.

In the case study of Rambourg-Schepens *et al*. (1988), coma, metabolic acidosis, hypokalemia, increase in serum creatinine level and urinary excretion of oxalate crystals were seen in a 50-year old woman, after a suicide attempt with a mixture containing 2-butoxyethanol (estimated dose of 500-1000 mg 2-butoxyethanol/kg bw).

In a 23-year old woman who made a suicide attempt with a mixture containing 2-butoxyethanol (estimated dose of ca. 1000 mg 2-butoxyethanol/kg bw), coma, hypotension, breathing difficulties, metabolic acidosis and haematuria were seen. The Hb concentration was decreased (11.9 g/dL on admission day and 8.9 g/dL on the second day), suggesting occurrence of haemolysis induced by 2-butoxyethanol (Gijsenbergh *et al*., 1989).

The dominant mode of action for toxic effects of 2-butoxyethanol after oral intake in adult humans, was induction of acidosis accompanied by CNS depression, which was observed in all cases. In these cases, 2-butoxyethanol was ingested together with other substances (ethanol and/or unknown substances) that could have some influence on the observed symptoms. All patients with acute oral intoxication with 2-butoxyethanol survived after hospital treatment. However, taking into account the severity of symptoms in patients with coma and pulmonary oedema or breathing difficulties, they could have died without any treatment, suggesting that oral doses above 1000 mg 2-butoxyethanol/kg bw could be lethal to adult humans.

The oral doses of 2-butoxyethanol inducing severe intoxication in humans is very close to median lethal oral doses in laboratory animals. Although, there was some variation in the oral LD_{50}-values in rats in the studies presented in the CLH report (from 470 mg/kg bw (Dow Chemical Company, 1959) up to 2800 mg/kg bw (Carpenter *et al*., 1956)), in the majority of the studies, the oral LD_{50}-values were within a range of 1480-2420 mg/kg bw: 1480 mg/kg bw (Smyth *et al*., 1941), 1950 mg/kg bw (Hoechst A., 1966), 1590 mg/kg bw (MB Research Laboratories, 1976), 1000-2000 mg/kg bw (Dow Chemical Company, 1981), 1746 mg/kg bw (Eastman Kodak, 1981a), and 2420 mg/kg bw (Bushy Run Research Center, 1980b). These oral LD_{50}-values in rats are within the same order of magnitude as in Guinea pigs, 1200–1414 mg/kg bw: 1200 mg/kg bw (Smyth *et al*., 1941 and Carpenter *et al*., 1956); 1414 mg/kg bw (Eastman Kodak, 1994b, cited in Gingell *et al*., 1998), and mice, 1170–2005 mg/kg bw: 1230 mg/kg bw (Carpenter *et al*., 1956), 1519 (fasted) / 2005 (fed) mg/kg bw (Eastman Kodak, 1981a), 1170 mg/kg bw (when fed as a water solution) / 1700 mg/kg bw (when fed as an oil solution) (Rowe and Wolf, 1982) and 1000-1600 mg/kg bw (Saparmamedov, 1974).

Similar level of oral LD_{50} in rats, mice and Guinea pigs, and presumably lethal oral doses of 2-butoxyethanol in humans, indicate that the differences between species in susceptibility to haemolytic action of 2-butoxyethanol has rather limited influence on potency of acute toxicity of this chemical to cause lethal acute poisoning.

The potency of 2-butoxyethanol to cause lethal effects seem to be similar in rats, mice, Guinea pigs and humans, despite presumably different mode of action as the oral LD_{50} in these animals are, in the majority of the studies, within a range of 1000–2000 mg/kg bw and that doses of 2-butoxyethanol inducing heavy acute poisoning in adult humans are within the same range.

In humans, metabolic acidosis, depression of the central nervous system, and, in some cases, acute respiratory distress syndrome (ARDS), seem to be the dominant modes of action, *i.e.*, mechanisms other than haemolysis.

The haemolysis of erythrocytes is not the only mode of action in lethal acute poisoning with 2-butoyxethanol, since the median lethal oral doses are similar in animals species sensitive to this effect (rats, mice) and in species which are relatively resistant to 2-butoxyethanol induced haemolysis (Guinea pigs).

The results of the cases studies assessing the toxic potency of 2-butoxyethanol and the described symptoms of acute oral poisoning in humans who ingested 2-butoxyethanol during suicide attempts, also indicate that human susceptibility to acute lethal toxicity of 2-butoxyethanol is similar to that in rats, mice and Guinea pigs.

Therefore, the data from oral exposure indicate that data from acute inhalation toxicity studies in animals other than Guinea pigs should be taken into account when classifying 2-butoxyethanol.

The dermal LD_{50}-values in rats and Guinea pigs are in most studies above 2000 mg/kg (see Table 12 of the CLH report) and were used to justify no classification of 2-butoxyethanol for acute dermal toxicity. Thus, no major difference in sensitivity of rats and Guinea pigs to acute dermal toxicity was noted.

Based on this comparison it is concluded that, despite differences in toxicokinetics and metabolism, there is no evidence that there is a major difference in sensitivity between Guinea pigs and rats to acute systemic toxicity of 2-butoxyethanol when it comes to lethal oral and dermal doses. Therefore, RAC does not consider it justified that Guinea pig data would be more relevant for assessment of acute toxicity hazard for humans compared to data in rats.

There is also no essential contradiction between the results of acute inhalation toxicity studies with 2-butoxyethanol in rats and Guinea pigs.

The LC_{50}-values in rats were in the range of 2.2-4.92 mg/L/4h (Carpenter *et al.*, 1956; Mellon Institute of Industrial Research, 1952; Bushy Run Research Center,

1980a; Shell Chemicals, 1982), except for the BASF study (1968) in which the LC_{50}-value was calculated to be in the range of 1.1–5.3 mg/L, extrapolated to 4h, but an exact LC_{50} was not possible to calculate.

No mortality was observed in Guinea pigs exposed to 2-butoxyethanol by inhalation for 4h at a concentration of 2.25 mg/L (Covance CRS Limited, 2019), although some local and systemic symptoms of toxicity were observed. No mortality was observed in Guinea pigs exposed to 2-butoxyethanol by inhalation for 1h at a concentration of 633 ppm (3.1 mg/L) (males) or 691 ppm (3.4 mg/L) females (Gingell *et al*., 1998). The results of the acute inhalation toxicity studies in Guinea pigs do not provide convincing evidence that inhalation exposure for 4h to 2-butoxyethanol at a concentration of e.g. 3.8 mg/L, equal to 80% of the saturated vapour concentration, at 22°C, would not be lethal for Guinea pigs.

There are several acute inhalation toxicity studies in which higher concentrations of vapour of 2-butoxyethanol than 2.25 mg/L were reported: 800 ppm (4.92 mg/L; Carpenter *et al*., 1956 and Mellon Institute of Industrial Research, 1952), 867 ppm (which, from information in Table 13 of the CLH report (2.85x(867/468) can be converted to 4.23 mg/L; Bushy Run Research Center, 1980a) and 3.7 mg/L (Klimisch *et al*., 1988).

The data on acute inhalation toxicity indicate that humans are sensitive to local or systemic effects of 2-butoxyethanol. Exposure for 4h at a concentration of 0.55 or 0.95 mg/L induced irritation to the eyes, nose and throat, a disturbance of taste, a slight increase in nasal mucous discharge and headache (Carpenter *et al*., 1956). The exposure at lower concentration of 0.24 mg/L for 2h did not produce noticeable toxic effects (Johanson, 1986 and Johanson and Boman, 1991).

The toxicokinetics studies in humans exposed to 2-butoxyethanol by inhalation, show similar patterns as seen in animal experiments: rapid uptake of 2-butoxyethanol with peaks in plasma concentrations after approx. 2h, followed by decay. The blood half-life of 2-butoxyethanol (40 minutes) was higher than in rats (10 minutes) and mice (5 minutes) after inhalation. The main metabolite was, as in the other mammal species tested, BAA and most of the test dose and BAA was also excreted via urine. Some studies demonstrated high interindividual variability in toxicokinetics of 2-butoxyethanol and its metabolites which may suggest interindividual variability also in susceptibility to toxicity of this substance in humans (Johanson *et al*., 1986a, Johanson and Johnsson, 1991; Jones and Cocker, 2003; Dow Chemical Company, 1955).

Despite differences in resistance of erythrocytes of different species to haemolysis induced by 2-butoxyethanol, different profiles of toxic symptoms and metabolism of this substance in rats, mice, rabbits, Guinea pigs and humans, their sensitivity to the acute systemic toxicity and lethal effects caused by 2-butoxyethanol, assessed based on LD_{50}-values in these animals, or on doses causing heavy intoxication in humans, is quite similar.

RAC notes that the study provided by industry during the consultation of the original CLH proposal and the new evidence provided in the form of a single inhalation study (Covance CRS Limited, 2019), does not override the overall weight of the other studies in several species.

Therefore, in the opinion of RAC, the LC_{50} values in three animal species: 2.2–4.92 mg/L/4h in rats (Carpenter *et al*., 1956; Mellon Institute of Industrial Research, 1952; Bushy Run Research Center, 1980a; Shell Chemicals, 1982; BASF, 1979; BASF, 1968), 4.12 mg/L/4h in mice (Werner *et al*., 1943 cited by Carpenter *et al*., 1956), and 2.36 mg/L/4h in rabbits (Dow Chemical Company, 1974) provide sufficient evidence that 2-butoxyethanol meets the criteria (LC_{50} in a range of 2-10 mg/L of air) given in the CLP Regulation, for classification of a substance present in air as a vapour, as Acute toxicity Category 3 via the inhalation route.

It is proposed to use the default value of 3 mg/L/4h from Table 3.1.2 of the CLP Regulation as the ATE.

Taking the above arguments into account, and consistent with the earlier opinion of 2018, RAC reaffirms its opinion that **2-butoxyethanol warrants classification as Acute Tox. 3; H331 (Toxic if inhaled), with an ATE of 3.0 mg/L/4h (vapour)**.

ANNEX: Records of the targeted consultation following the submission of new information on acute inhalation toxicity of 2-butoxyethanol; ethylene glycol monobutyl ether (EGBE)

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