Committee for Risk Assessment

RAC

Annex 1

Background document

to the Opinion proposing harmonised classification and labelling at EU level of

di-*n*-butylamine

EC Number: 203-921-8

CAS Number: 111-92-2

CLH-O-0000007007-79-01/F

The background document is a compilation of information considered relevant by the dossier submitter or by RAC for the proposed classification. It includes the proposal of the dossier submitter and the conclusion of RAC. It is based on the official CLH report submitted to consultation. RAC has not changed the text of this CLH report but inserted text which is specifically marked as 'RAC evaluation'. Only the RAC text reflects the view of RAC.

Adopted

10 June 2021

CLH report

Proposal for Harmonised Classification and Labelling

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

International Chemical Identification:

di-*n*-butylamine

EC Number: **203-921-8**

CAS Number: **111-92-2**

Index Number: **612-049-00-0**

Contact details for dossier submitter:

Environment Agency Austria, Spittelauer Lände 5, A-1090 Vienna

on behalf of the Austrian Competent Authority (Austrian Federal Ministry for Sustainability and Tourism, Stubenring 1, 1010 Vienna, Austria)

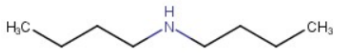
Version number: 02 **Date: December 2019**

CONTENTS

1 IDENTITY OF THE SUBSTANCE

1.1 Name and other identifiers of the substance

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

Name(s) in the IUPAC nomenclature or other international chemical name(s)	N-butylbutan-1-amine
Other names (usual name, trade name, abbreviation)	Dibutylamine Di-*n*-butylamine 1-Butanamine, N-butyl- N-Butyl-1-butanamine Dibutilamina N-butylbutan-1-amine N-Dibutylamine Di-(*n*-butyl)amine
ISO common name (if available and appropriate)	Not applicable
EC number (if available and appropriate)	203-921-8
EC name (if available and appropriate)	Di-*n*-butylamine
CAS number (if available)	111-92-2
Other identity code (if available)	RTECS Number: HR7780000 ICSC Number: 1337 UN Number: 2248 PubChem CID: 8148
Molecular formula	$C_8H_{19}N$
Structural formula	 **(source:** European Chemicals Agency, http://echa.europa.eu/**)**
SMILES notation (if available)	CCCCNCCCC
Molecular weight or molecular weight range	129.247 g/mol
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	Not applicable
Description of the manufacturing process and identity of the source (for UVCB substances only)	Not applicable
Degree of purity (%) (if relevant for the entry in Annex VI)	≥ 80 wt %

1.2 Composition of the substance

Di-*n*-butylamine is a mono-constituent substance.

Table 2: Constituents (non-confidential information)

Constituent (Name and numerical identifier)	Concentration range (% w/w minimum and maximum in multi-constituent substances)	Current CLH in Annex VI Table 3.1 (CLP)	Current self-classification and labelling (CLP)
di-*n*-butylamine (EC 203-921-8)	Not applicable	Flam. Liq. 3; H226 Acute Tox. 4*; H332 Acute Tox. 4*; H312 Acute Tox. 4*; H302	Flam. Liq. 3; H226 Acute Tox. 2; H330 Acute Tox. 3; H311 Acute Tox. 4; H302 Skin Corr. 1A; H314

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

Impurity (Name and numerical identifier)	Concentration range (% w/w minimum and maximum)	Current CLH in Annex VI Table 3.1 (CLP)	Current self-classification and labelling (CLP)	The impurity contributes to the classification and labelling
Not relevant				

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

Additive (Name and numerical identifier)	Function	Concentration range (% w/w minimum and maximum)	Current CLH in Annex VI Table 3.1 (CLP)	Current self-classification and labelling (CLP)	The additive contributes to the classification and labelling
Not relevant					

Table 5: Test substances (non-confidential information)

Identification of test substance	Purity	Impurities and additives (identity, %, classification if available)	Other information	The study(ies) in which the test substance is used
The test substance is di-*n*-butylamine in all studies where the test substance was explicitly stated. The purity is given in the study records below if available.				

2 PROPOSED HARMONISED CLASSIFICATION AND LABELLING

2.1 Proposed harmonised classification and labelling according to the CLP criteria

Table 6: Classification and labelling of di-*n*-butylamine

	Index No	Chemical name	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors and ATE	Notes
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Pictogram, Signal Word Code(s)	Hazard statement Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	612-049-00-0	di-*n*-butylamine	203-921-8	111-92-2	Flam. Liq. 3 Acute Tox. 4 * Acute Tox. 4 * Acute Tox. 4 *	H226 H332 H312 H302	GHS02 GHS07 Wng	H226 H332 H312 H302			
Dossier submitters proposal	612-049-00-0	di-*n*-butylamine	203-921-8	111-92-2	**Add** Skin Corr 1B Eye Dam 1 STOT SE 3 **Modify** Acute Tox. 2 Acute Tox. 3 Acute Tox. 3 **Retain** Flam. Liq. 3	**Add** H314 H318 H335 **Modify** H330 H311 H301 **Retain** H226	**Add** GHS05 **Modify** GHS06 Dgr **Retain** GHS02	**Add** H314 **Modify** H330 H311 H301 **Retain** H226		**Add** Oral: ATE = 220 mg/kg bw Dermal: ATE = 768 mg/kg bw Inhalation: ATE = 1.15 mg/L	
Resulting Annex VI entry if agreed by RAC and COM	612-049-00-0	di-*n*-butylamine	203-921-8	111-92-2	Flam. Liq. 3 Acute Tox. 2 Acute Tox. 3 Acute Tox. 3 Skin Corr 1B Eye Dam 1 STOT SE 3	H226 H330 H311 H301 H314 H318 H335	GHS02 GHS05 GHS06 Dgr	H226 H330 H311 H301 H314		Oral: ATE = 220 mg/kg bw Dermal: ATE = 768 mg/kg bw Inhalation: ATE = 1.15 mg/L	

Table 7: Reason for not proposing harmonised classification and status under public consultation

Hazard class	Reason for no classification	Within the scope of public consultation
Explosives	*hazard class not assessed in this dossier*	No
Flammable gases (including chemically unstable gases)	*hazard class not assessed in this dossier*	No
Oxidising gases	*hazard class not assessed in this dossier*	No
Gases under pressure	*hazard class not assessed in this dossier*	No
Flammable liquids	*hazard class not assessed in this dossier*	No
Flammable solids	*hazard class not assessed in this dossier*	No
Self-reactive substances	*hazard class not assessed in this dossier*	No
Pyrophoric liquids	*hazard class not assessed in this dossier*	No
Pyrophoric solids	*hazard class not assessed in this dossier*	No
Self-heating substances	*hazard class not assessed in this dossier*	No
Substances which in contact with water emit flammable gases	*hazard class not assessed in this dossier*	No
Oxidising liquids	*hazard class not assessed in this dossier*	No
Oxidising solids	*hazard class not assessed in this dossier*	No
Organic peroxides	*hazard class not assessed in this dossier*	No
Corrosive to metals	*hazard class not assessed in this dossier*	No
Acute toxicity via oral route	Acute Tox 3; H301	Yes
Acute toxicity via dermal route	Acute Tox 3; H311	Yes
Acute toxicity via inhalation route	Acute Tox 2; H330	Yes
Skin corrosion/irritation	Skin Corr 1B, H314	Yes
Serious eye damage/eye irritation	Eye Dam 1, H318	Yes
Respiratory sensitisation	*hazard class not assessed in this dossier*	No
Skin sensitisation	*hazard class not assessed in this dossier*	No
Germ cell mutagenicity	*hazard class not assessed in this dossier*	No
Carcinogenicity	*hazard class not assessed in this dossier*	No
Reproductive toxicity	*hazard class not assessed in this dossier*	No
Specific target organ toxicity-single exposure	STOT SE 3, H335	Yes
Specific target organ toxicity-repeated exposure	*hazard class not assessed in this dossier*	No
Aspiration hazard	*hazard class not assessed in this dossier*	No
Hazardous to the aquatic environment	*hazard class not assessed in this dossier*	No
Hazardous to the ozone layer	*hazard class not assessed in this dossier*	No

3 HISTORY OF THE PREVIOUS CLASSIFICATION AND LABELLING

Di-*n*-butylamine had a harmonized classification under the Dangerous Substances Directive (67/548/EEC). This was translated to a harmonized CLP classification in Annex VI, Regulation (EC) No 1272/2008 (CLP Regulation) and a minimum classification (according Annex VII) was applied to acute toxicity for all routes (marked as Acute Tox. 4 * for all routes).

The current harmonized classification (CLP, Annex VI Table 3.1) for di-*n*-butylamine is:

Flam. Liq. 3; H226

Acute Tox. 4*; H332

Acute Tox. 4*; H312

Acute Tox. 4*; H302

Self-classification:

The frequency of hazard classifications among all C&L notifications (occurring in at least 10% of notifications) was retrieved from ECHA dissemination site [accessed 12/2020] and is given below. In total, 731 notifiers provided information on their hazard classifications (14 aggregated notifications):

Hazard code	Hazard statement	% of notifications
H226	Flammable liquid and vapour	100
H302	Harmful if swallowed	99.7
H312	Harmful in contact with skin	71.1
H311	Toxic in contact with skin	28.7
H332	Harmful if inhaled	71.3
H330	Fatal if inhaled	18.7
H314	Causes severe skin burns and eye damage	28.6
H318	Causes serious eye damage	13

RAC general comment
Di-*n*-butylamine is an important industrial chemical registered (as dibutylamine) at 1000 to 10000 tonnes per annum. Identified uses are manufacture, formulation or re-packing, and use at industrial sites.

4 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

[B.] Justification that action is needed at Community level is required.

Reason for a need for action at Community level:

- Change in existing entry due to changes in the criteria (translation from DSD to CLP)
- Disagreement by DS with current self-classification

Further detail on need of action at Community level

There is a harmonised classification entry in Annex VI to CLP containing a minimum classification (*) and it is concluded that a refinement of the classification based on available data is justified. Differences in self-classification between different notifiers in the C&L Inventory and registration dossier are discovered.

Di-*n*-butylamine is an important industrial chemical. A correct classification for acute toxicity and corrosion is essential to minimize uncertainties in classification along the supply chain and to ensure a high level of protection of workers by setting the right risk management measures.

5 IDENTIFIED USES

Di-*n*-butylamine is manufactured and/or imported in the European Economic Area in 1 000 - 10 000 tonnes per year. Identified uses are manufacture, formulation or re-packing, and use at industrial sites (see Table 8 for details) (ECHA dissemination site, July 2019).

Table 8: Registered uses of di-*n*-butylamine (according to ECHA dissemination site, July 2019)

Uses at industrial sites	Use as laboratory chemical
	Industrial use resulting in manufacture of another substance
	Use as processing aid (catalyst) in rubber production (vulcanisation)
Uses at formulation or re-packing	Use as processing aid (catalyst) in rubber production (vulcanisation)
	Formulation of preparations

6 DATA SOURCES

Systematic searches for publications and other relevant data were performed based on the following databases:

- U.S. National Library of Medicine, Pubmed.gov[1]

[1] https://www.ncbi.nlm.nih.gov/pubmed assessed at 7.2.2019

- TOXNET[2], ChemID*plus*[3], IPCS[4], eChemPortal[5], EPA Comptox Dashboard[6], EPA Chemview[7]
- Chemical Abstracts, Medline, Biosis, Embase, SciSearch, PQScitech (at host STN International Europe[8])

in addition to unspecific databases (e.g., *google scholar*).

The REACH registration dossier for di-*n*-butylamine, available from ECHA's disseminated database (accessed 2019) has been analysed for study references, which then have been considered as data sources for this CLH report.

Relevant reviews and monographs with toxicological risk assessments on di-*n*-butylamine were analysed for study references. Used reviews are AGS (2006) and TCEQ (2016).

Whenever secondary sources were encountered, it was attempted to retrieve the respective primary sources.

[2] https://toxnet.nlm.nih.gov/ assessed at 7.2.2019

[3] https://chem.nlm.nih.gov/chemidplus/ assessed at 7.2.2019

[4] http://www.inchem.org/ assessed at 7.2.2019

[5] http://www.echemportal.org/echemportal/page.action?pageID=9 assessed at 7.2.2019

[6] https://comptox.epa.gov/dashboard/

[7] https://chemview.epa.gov/chemview

[8] http://www.stn-international.de/index.php?id=123 assessed at 13.2.2019

7 PHYSICOCHEMICAL PROPERTIES

Table 9: Summary of physicochemical properties

Property	Value	Reference	Comment (e.g. measured or estimated)
Physical state at 20°C and 101.3 kPa	Liquid	ECHA Dissemination (2019)	Visual observation
Melting/freezing point	-57 – -59 °C	ECHA Dissemination (2019)	Measured, at 1013.25 hPa
Boiling point	160 °C	ECHA Dissemination (2019)	Measured, at 1013.25 hPa
Density	0.7577 g/cm³	ECHA Dissemination (2019)	Measured, at 22.9 °C
Vapour pressure	2.26 hPa	ECHA Dissemination (2019)	Measured, at 20.3 °C and up to 1013.25 hPa
Surface tension	50.6 mN/m	ECHA Dissemination (2019)	Measured, at 20 °C, concentration of 1.005 g/L
Water solubility	3.8 g/L	ECHA Dissemination (2019)	Measured, at 20 °C and approx. pH 12
Partition coefficient n-octanol/water	2.1	ECHA Dissemination (2019)	Measured, at 23 °C and pH 12
Flash point	40.5 °C	ECHA Dissemination (2019)	Measured, at 1013 hPa
Flammability	Flammable liquid	ECHA Dissemination (2019)	Measured
Explosive properties	Non explosive	ECHA Dissemination (2019)	Derived from chemical structure
Self-ignition temperature	255 °C	ECHA Dissemination (2019)	Measured, at 1013 hPa
Oxidising properties	No oxidising properties	ECHA Dissemination (2019)	Derived from chemical structure
Granulometry	Not applicable	-	-
Stability in organic solvents and identity of relevant degradation products	Not applicable	ECHA Dissemination (2019)	Justification given: expert judgement
Dissociation constant	11 (pKa)	ECHA Dissemination (2019)	Measured, at 20 °C
Viscosity	0.85 mPa*s	ECHA Dissemination (2019)	Measured, at 20 °C

8 EVALUATION OF PHYSICAL HAZARDS

Not performed for this substance.

9 TOXICOKINETICS (ABSORPTION, METABOLISM, DISTRIBUTION AND ELIMINATION)

Evaluation not performed for this substance.

10 EVALUATION OF HEALTH HAZARDS

Acute toxicity

10.1 Acute toxicity - oral route

Table 10: Summary table of animal studies on acute oral toxicity

Method, guideline, reliability	Species, strain, sex, no/group	Test substance	Dose levels, duration of exposure	Value LD_{50}	Reference
Acute oral toxicity Similar to OECD 401 GLP: no Reliability (REACH registration): 2, key study Reliability (this assessment): 3	Rat, Wistar 5 animals, sex not stated	Dibutylamine No information on source No information on purity	Dose levels not known. Single application via gavage Vehicle: 1% Tergitol	550 mg/kg bw (95% CI: 480 – 620 mg/kg bw)	Smyth et al. (1954) [key study, REACH registration]
Acute oral toxicity Similar to OECD 401 GLP: no Reliability (this assessment): 3	rat, strain not specified 10 male and 10 female per dose group	Dibutylamine Technical purity, no further information on purity Source: Former VEB Synthesewerk Schwarzheide (today BASF Schwarzheide GmbH)	Several dose levels tested (males: 5, females: 6). Doses not specified Single application via gavage Variable concentration in vehicle (peanut oil): constant volume 5 mL/kg	male: 310 mg/kg bw (95% CI: 251 - 382) female: 220 mg/kg bw (95% CI: 191 - 253)	Schmidt et al. (1974)
Acute oral toxicity Similarity to guideline unknown GLP: no Reliability (this assessment): 3	Rat, Wistar 3-5 male and 3-5 female per dose group	Dibutylamine No information on source No information on purity	No information on dose levels Application via gavage Vehicle: oil	male: 189 mg/kg bw female: 239 mg/kg bw	Ciugudeanu et al. (1985) No English translation obtainable
Acute oral toxicity	Mouse and guinea pig (unknown	Dibutylamine No information	No information given	290 mg/kg bw (mouse)	Secondary source:

Method, guideline, reliability	Species, strain, sex, no/group	Test substance	Dose levels, duration of exposure	Value LD_{50}	Reference
Similarity to guideline unknown GLP: no Reliability (this assessment): 4	strains) No information on group size and sex	on source No information on purity		230 mg/kg bw (guinea pig)	Sax and Lewis (1989) Primary source not obtainable (given as "Gigiena i Sanitariya-40(11),21,75")

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

No animal study is available that is sufficiently conform to a guideline and is conclusive on its own. Two old studies, similar to OECD Guideline 401, are available (Table 10). However, lacking information on purity of the test material is limiting their reliability. These studies determined LD_{50} values of 220 – 550 mg/kg bw in rats (Smyth, 1954 and Schmidt, 1974). Two additional references provide LD_{50} values of 230 mg/kg bw for guinea pigs and 290 mg/kg bw for mice, but the primary sources are not obtainable (Sax and Lewis, 1989) or could not be evaluated due to the lack of a translation (Ciugudeanu et al., 1985).

No human studies with relevance for comparison with the CLP criteria are available.

10.1.2 Comparison with the CLP criteria

According to Table 3.1.1 of Regulation (EC) No. 1272/2008 a substance shall be classified as

- Acute Tox 4 (oral) if the LD_{50}/ATE values are > 300 and ≤ 2000 mg/kg bw.
- Acute Tox 3 (oral) if the LD_{50}/ATE values are > 50 and ≤ 300 mg/kg bw.

All available studies are of limited reliability. In a WoE approach, the studies by Smyth (1954) and Schmidt (1974) are given more weight, as they provide clearly more information to judge the relevance of the determined LD_{50} values for comparison with the CLP criteria. The LD_{50} by the key study in the dossier (LD_{50} rat = 550 mg/kg bw) corresponds to a classification as acute oral toxicity category 4 (300 – 2000 mg/kg bw), while the Schmidt study (LD_{50} rat, female: 220 mg/kg bw, male: 310 mg/kg bw) indicates a classification as category 3 (50 – 300 mg/kg bw). There is no apparent reason to prefer one of these two studies over the other and the CLP regulation envisages the use of the lower ATE for comparison with the CLP criteria. In addition, the less reliable studies both determined values also leading to a classification in category 3. In conclusion, the weight of evidence leads to a classification for acute oral toxicity, category 3 for di-*n*-butylamine.

10.1.3 Conclusion on classification and labelling for acute dermal toxicity

Based on the CLP-Criteria di-*n*-butylamine has to be classified in category 3 for acute oral toxicity (Acute Tox 3, H301).

Based on the lowest LD_{50} used for classification an ATE value of 220 mg/kg bw is indicated.

10.2 Acute toxicity - dermal route

Table 11: Summary table of animal studies on acute dermal toxicity

Method, guideline, reliability	Species, strain, sex, no/group	Test substance	Dose levels duration of exposure	Value LD_{50}	Reference
Acute dermal toxicity Similar to OECD 402 GLP: no Reliability (REACH registration): 2, key study Reliability (this assessment): 3	Rabbit, New Zealand White 4 males per dose group	Di-*n*-butylamine No information on source No information on purity	No information on dose levels Occlusive application 24 h exposure	768 mg/kg bw (95% CI: 620 – 1130 mg/kg bw) (reported as 1.01 mL/kg bw with a 95% CI: 0.68 – 1.49 mL/kg bw)	Primary source: (Smyth et al., 1954) [key study, REACH registration]

10.2.1 Short summary and overall relevance of the provided information on acute dermal toxicity

There is very limited data available on dermal toxicity. Only a single animal study with limited reliability (due to lacking information on purity) is available. An additional result which is found in secondary literature (Sax and Lewis, 1989) is actually a conversion mistake of the former study and is not reported in Table 11.

No human studies with relevance for comparison with the CLP criteria are available.

10.2.2 Comparison with the CLP criteria

According to Table 3.1.1 of Regulation (EC) No. 1272/2008 a substance shall be classified as

- Acute Tox 4 (dermal) if the LC_{50}/ATE values are > 1000 and ≤ 2000 mg/kg bw
- Acute Tox 3 (dermal) if the LC_{50}/ATE values are > 200 ≤ 1000 mg/kg bw

A classification is proposed based on the only available study, although the reliability is limited. A major concern is the lacking information on purity, however, using this study is expected to err on the conservative side. Therefore, use of this study is considered acceptable in a conservative approach. This study reports a LD_{50} of 768 mg/kg bw, which corresponds to category 3 of the CLP criteria for acute dermal toxicity (200 – 1000 mg/kg bw).

10.2.3 Conclusion on classification and labelling for acute dermal toxicity

According to the CLP-criteria di-*n*-butylamine has to be classified in category 3 for acute dermal toxicity (Acute Tox 3, H311).

Based on the available LD_{50} value an ATE = 768 mg/kg bw is indicated.

10.3 Acute toxicity - inhalation route

Table 12: Summary table of animal studies on acute inhalation toxicity

Method, guideline, reliability	**Species, strain, sex, no/group**	**Test substance, form**	**Dose levels, duration of exposure**	**Value** LC_{50}	**Reference**
Acute inhalation toxicity Equivalent to OECD 403 GLP: yes Reliability (REACH registration): 2, key study Reliability (this assessment): 1	Rat, Sprague-Dawley 5 males and 5 females per dose group	Dibutylamine, as vapour Purity > 99.5 % No information on source	0, 0.76, 1.08, 1.18, 1.39, 3.91 mg/L 4 h exposure 14 days post exposure observation	1.15 mg/L mortalities C: m 0/5, f 0/5 0.76 mg/L: m 2/5, f 0/5 1.08 mg/L: m 0/5, f 2/5 1.18 mg/L: m 3/5, f 1/5 1.39 mg/L: m 5/5, f 5/5 3.91 mg/L: m 5/5, f 5/5	Primary source: unnamed study report, 1987 [Study 001, REACH registration]
Acute inhalation toxicity Similarity to guideline unknown GLP: no Reliability (REACH registration): 2 Reliability (this assessment): 3	Rat, Wistar 6 male rats per dose	Dibutylamine, as vapour No information on source No information on purity	No information on concentrations 4 h whole body exposure 14 days post exposure observation	> 1.34 mg/L & < 2.68 mg/L Mortalities 1.34 mg/L: 0/6 2.68 mg/L: 6/6	Primary source: Smyth et al. (1954) [Study 002, REACH registration]
Acute inhalation toxicity Similarity to guideline unknown GLP: no Reliability (this assessment): 4	Rat, strain not specified No information on group size and sex	Dibutylamine No information on source No information on purity	No information on concentrations 4 h exposure No further information on exposure	2.68 mg/L	Secondary Source: (Greim et al., 1998) Primary source not sufficiently specified (data provided by industry)

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

A guideline- and GLP-conform study on rats is available (proprietary study, reported as key study in the registration dossier). This study is of high quality and is on its own conclusive for comparison with the CLP

criteria. After 4 h exposure, this study determined a LC50 of 1.15 mg/L. During exposure the rats showed signs of sensory irritation like partial closing of the eyes, reduced respiratory rate, abnormal respiratory movements and adoption of an anormal body posture. Less frequently gasping, exsessive salivation. Lacrimation and convulsion were observed. When removed from the test chamber previously exposed rats showed abnormal breathing, lethargy, ataxia, prone posture and intermittent convulsions. Abnormal breathing, rales and sneezing were evident till day 2 with normal appearance on day 3.

One additional study in rats of insufficient reliability (study 002 in ECHA Dissemination, 2019), as well as a LC_{50} value for rats reported in a secondary source, without sufficient documentation on the primary source (Greim et al., 1998), could be found. These studies provide a range of >1.34 mg/L to 2.68 mg/L as LC_{50} values.

No human studies with relevance for comparison with the CLP criteria are available.

10.3.2 Comparison with the CLP criteria

According to Table 3.1.1 of Regulation (EC) No. 1272/2008 a substance shall be classified as

- Acute Tox 4 (inhal) if the LC_{50} values are > 10.0 mg/L and ≤ 20.0 mg/L (4h exposure)
- Acute Tox 3 (inhal) if the LC_{50} values are > 2.0 mg/L and ≤ 10.0 mg/L (4h exposure)
- Acute Tox 2 (inhal) if the LC_{50} values are > 0.5 and ≤ 2 mg/L (4h exposure)

The key study results in a LC_{50} value (1.15 mg/L, 4 h exposure), which corresponds to a classification as category 2 (0.5 – 2 mg/L). The other available study results with insufficient reliability support this classification, yet the upper bound of the determined LC_{50} range by these studies slightly exceeds the boundaries of category 2. However, given the significantly higher relevance of the key study, this has no impact on the assessment.

10.3.3 Conclusion on classification and labelling for acute inhalation toxicity

Based on the CLP criteria di-*n*-butylamine has to be classified in category 2 for acute inhalation toxicity (Acute Tox 2, H 330).

The key study gives an LC_{50}/ATE value of 1.15 mg/L.

RAC evaluation of acute toxicity
Summary of the Dossier Submitter's proposal ***Oral*** Two older acute toxicity studies via the oral route in rats (similar to OECD TG 401) revealed LD_{50} values of 220 and 550 mg/kg bw (Smyth, 1954 and Schmidt, 1974). LD_{50}

values of 230 mg/kg bw for guinea pigs and 290 mg/kg bw for mice were also reported (Sax and Lewis, 1989), however the primary sources were not available to the DS. In a weight of evidence assessment, the DS proposed classification for acute oral toxicity in category 3, H301 with an ATE of 220 mg/kg bw.

Species	**LD_{50} (95% CI) mg/kg bw**	**Reliability (DS)**	**Study**	**Remark**
Rat, Wistar (n=5)	550 (480–620)	3	Smyth *et al*., 1954	Similar to OECD TG 401, non-GLP
Rat, no strain 10/sex/group	male: 310 (251-382) female: 220 (191-253)	3	Schmidt *et al*., 1974	Similar to OECD TG 401, non-GLP
Rat, Wistar 3-5/sex/group	male: 189 female: 239	3	Ciugudeanu *et al*., 1985	No guideline, non-GLP
Mouse, guinea pig	mouse: 290 guinea pig: 230	4	Sax and Lewis, 1989	No guideline, unknown group size and strains

Dermal

Based on LD_{50} of 768 mg/kg bw from a single acute toxicity study with limited reliability, the DS proposed classification as Acute Tox 3, H311 for the dermal route of exposure.

Species	**LD50 (95% CI) mg/kg bw**	**Reliability (DS)**	**Study**	**Remarks**
Rabbit, NZ White 4 males/dose	768 (620–1130)	3	Smyth *et al*., 1954	Similar to OECD TG 402; occlusive application for 24h; LD50 reported as 1.01 mL/kg bw (95% CI: 0.68–1.49)

Inhalation

An acute inhalation toxicity study in rats performed according to GLP and OECD TG 403 revealed a LC_{50} of 1.15 mg/L (vapour) after 4 h exposure. Further data from secondary sources provide a range of LC_{50} values from >1.34 mg/L to <2.68 mg/L. Considering the outcome of the most reliable study, the DS proposed classification in category 2, H330, and ATE value of 1.15 mg/L.

Species	**LC_{50} (mg/L)**	**Reliability (DS)**	**Study**	**Remarks**
Sprague-Dawley rat, 5/sex/dose	1.15 (both sexes combined)	1	Anonymous, 1987	OECD TG 403, GLP; Dose levels: 0, 0.76, 1.08, 1.18, 1.39, 3.91 mg/L (vapour)
Wistar rat, 6 males/dose	>1.34 (0/6 dead) <2.68 (6/6 dead)	3	Smyth *et al*., 1954	No guideline specified, 4 h whole body exposure to vapour
Rat, strain, sex and group size not specified	2.68	4	Greim *et al*., 1998	4h exposure, no further details

Comments received during consultation

Two MSCA supported the proposed classifications on acute toxicity. Considering the (low)

quality of the available studies, one MSCA proposed using the generic ATEs for classification of mixtures: 100 mg/kg bw for acute oral toxicity, and 300 mg/kg bw for acute dermal toxicity.

Assessment and comparison with the classification criteria

Oral

All available studies are lacking information on the specific study design and the purity of the test substance. Smyth (1954) established an LD_{50} of 550 mg/kg bw in rats, while the study by Schmidt (1974) reported an LD_{50} of 220 mg/kg bw in female and 310 mg/kg bw in male rats. An additional reference (Ciugudeanu *et al*., 1985) provided LD_{50} values of 189 mg/kg bw in male and 239 mg/kg bw in female rats, however direct access to the study is not available. A secondary source (Sax and Lewis, 1989) reported LD_{50} of 230 mg/kg bw for Guinea pigs and 290 mg/kg bw for mice.

In agreement with the DS evaluation, RAC considers the reliability of all studies as low. The studies by Smyth (1954) and Schmidt (1974) provide slightly more information on animal numbers and similarity to test guideline protocols thus allowing comparison with the CLP criteria. An LD_{50} of 550 mg/kg bw in rats indicates classification in category 4 (300–2000 mg/kg bw), while an LD_{50} of 220 mg/kg bw in female rats points to a classification in category 3 (50–300 mg/kg bw). According to DS, there were no apparent reasons to give stronger preference to one of these two studies, therefore category 3 was proposed based on the lower ATE value. Additional support is provided by the few less reliable studies with LD_{50} values within the range for category 3. RAC agrees with this weight of evidence assessment and supports the classification in category 3 for acute oral toxicity of di-n-butylamine with an ATE of 220 mg/kg bw.

During consultation, one MSCA proposed using the generic ATE of 100 mg/kg bw for classification of mixtures due to the overall low quality of the studies. RAC notes that the range of all LD_{50} values (220-550 mg/kg bw) is at least a factor of 2 to 5 above the generic ATE and supports using the lowest LD_{50} of 220 mg/kg bw established in female rats.

Dermal

The single acute dermal toxicity study in NZ White rabbits is reported as similar to OECD TG 402 (Smyth *et al*., 1954), however the lack of information on purity of the test substance and applied doses limit its reliability. The study reports an LD_{50} of 768 mg/kg bw, which corresponds to category 3 of the CLP criteria for acute dermal toxicity (200–1000 mg/kg bw). RAC agrees with DS proposal for classification of di-n-butylamine as **Acute Tox 3, H311.**

During consultation, one MSCA expressed preference for using the generic ATE of 300 mg/kg bw instead of the experimentally determined LD_{50} of 768 mg/kg bw. Considering the limitations of the study and the lack of any additional supporting data, RAC agrees on using the generic ATE of 300 mg/kg bw as a more conservative choice reflecting the uncertain database.

Inhalation

One OECD TG 403 study on rats conforming to GLP and one additional study in rats of limited reliability were available to DS for this endpoint.

In the guideline study (Anonymous 1987), Sprague-Dawley rats (5/sex/dose) were exposed for 4 hours to vapour of di-n-butylamine at concentrations of up to 3.91 mg/L. Recorded mortalities in each dose group are summarized below.

Dose level (mg/L)	**0**	**0.76**	**1.08**	**1.18**	**1.39**	**3.91**
Mortality						
males	0/5	2/5	0/5	3/5	5/5	5/5
females	0/5	0/5	2/5	1/5	5/5	5/5

The study determined an LC_{50} of 1.15 mg/L. Signs of sensory irritation such as partial closing of the eyes, reduced respiratory rate, abnormal respiratory movements and abnormal body posture were reported during exposure. Frequently gasping, excessive salivation, lacrimation and convulsion were observed to a lesser degree. After exposure, rats showed abnormal breathing, lethargy, ataxia, prone posture and intermittent convulsions. Abnormal breathing, rales and sneezing were evident during the first two days post exposure with normal appearance on day 3.

One less reliable study and a secondary source provided a range of LC_{50} values for rats between >1.34 mg/L and <2.68 mg/L. No mortalities were reported after 3 days nose-only exposure of 10 Wistar rats (5M/5F, 6h/day) to 0.450 mg/L (vapour) di-n-butylamine (Buschman *et al*., 2003; details in STOT SE section).

Based on the outcome of the most reliable study, the DS proposed classification as **Acute Tox. 2, H330 with an ATE value of 1.15 mg/L**. RAC agrees to the proposal and concludes that a rounded ATE of 1.2 mg/L is warranted.

10.4 Skin corrosion/irritation

Table 13: Summary table of animal studies on skin corrosion/irritation

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance,**	**Dose levels duration of exposure**	**Results** **-Observations and time point of onset** **-Mean scores/animal** **-Reversibility**	**Reference**
OECD 404 Non GLP Reliability (REACH registration): 2, key study	Rabbit, Vienna White (1m, 1f)	Dibutylamine >99.5%	0.5ml Occlusive (2x2cm) 3min, 1h test substance removed after exposure time Observation 8d	Erythema score, 3min exposure Mean (24, 48, 72h) = 4 Max. score = 4 Not reversible Erythema score, 1 h exposure Mean (24, 48, 72h) = 4 Max. score = 4	Anonymous (1978) [key study, REACH registration]

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance,**	**Dose levels duration of exposure**	**Results** **-Observations and time point of onset** **-Mean scores/animal** **-Reversibility**	**Reference**
				Not reversible Edema score, 3 min exposure Mean (24, 48, 72h) = 2 Max. Score = 2 Not reversible Edema score, 1 h exposure Mean (24, 48, 72h) = 2 Max. Score = 2 Not reversible Necrosis after 24h observed	
Draize-method	Rabbits, New Zealand White N=6	Dibutylamine (100%)	0.5 ml 4h Clipped intact skin One-inch square Observations: 4, 24, 48h	Erythema score, 4h exposure Mean (24, 48h) = 4 Max. score = 4 Not reversible Edema score, 1 h exposure Mean (24, 48h) = 1.58 Max. Score = 3 Necrosis at all timepoints	Virginia chemicals, 1973 OTS0515256
Skin irritation test	Albino rabbits N=3	Dibutylamine	0.5 ml 24h Clipped, intact and abraded skin	15min: skin turned brown 24h: large necrotic lesions 48h: dry, hard lesions, cracking, raw rat tissue	Pennwalt Corp (1986) OTS0513616

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

For the evaluation of skin irritation/corrosion one OECD 404 study is available. Two rabbits (one male, one female) were exposed (occlusive, 2x2 cm, clipped) to 0.5 ml of undiluted dibutylamin (>99.5%) for 3 min or 1 h. The test substance was removed at the end of exposure period with Lutrol and Lutrol/water (1:1). Animals were observed for 8 days. Untreated skin of the same animal served as control. The 3 min and 1 h exposure caused severe erythema and slight to moderate edema (see Table 14). After 24 h necrosis was observed (no earlier observations documented). At the end of the observation period of 8 days leathery necrosis was observed; this was considered as a full thickness necrosis.

Table 14: Individual animal data and mean scores after an exposure duration of 3min or 1h (Anonymous, 1978).

Exposure time	animal #	Erythema score				Edema score			
		24h	48h	72h	Mean	24h	48h	72h	Mean
3 min	#1	4	4	4	4.0	2	2	2	2.0
	#2	4	4	4	4.0	2	2	2	2.0
1 h	#1	4	4	4	4.0	2	2	2	2.0
	#2	4	4	4	4.0	2	2	2	2.0

In a Draize test 6 rabbits (clipped, intact skin) were exposed to 0.5ml of undiluted dibutylamine for 4h (Virginia chemicals, 1973). Scores and necrosis were recorded after 4, 24 and 48h of exposure and are presented below.

Table 15: Individual erythema and edema scores scores after 4h of exposure (Virginia chemicals, 1973).

Animal No	Erythema score			Edema score			Necrosis		
	4h	24h	48h	4h	24h	48h	4h	24h	48h
1	4	4	4	3	3	1	yes	yes	yes
2	4	4	4	2	2	1	yes	yes	yes
3	4	4	4	2	2	1	yes	yes	yes
4	4	4	4	2	2	1	yes	yes	yes
5	4	4	4	2	2	1	yes	yes	yes
6	4	4	4	2	2	1	yes	yes	yes

In another test (Pennwalt Corp, 1986) 0.5 ml of dibutylamine was applied to three Albino rabbits for 24h. Each had one intact and one abraded skin site (clipped, occlusive). As a result is was recorded that the sample spread beyond the intended sites of contact and caused pain in every instance. The skin turned brown within 15 minutes. 24h after the first contact large necrotic lesions were described. These lesions became dry, hard and concave within 48h. In the following they cracked and peeled exposing raw tissue.

In a publication by Smyth (1952) corrosive effects to skin for the substance dibutylamine are reported without further details.

Registrants also mention another Draize study (Air products, 1975; Val. 2), where severe erythema, edema and necrosis persisting through 72 h were observed after 4h of exposure. Corrosive skin effects were also mentioned in another study (Elf Atochem 1976; Val. 3). However, no further details can be provided as original literature could not be located.

10.4.2 Comparison with the CLP criteria

A corrosive substance is a substance that produces destruction of skin tissue, namely, visible necrosis through the epidermis and into the dermis, in at least 1 tested animal after exposure up to a 4 hour duration. Classification is done as follows:

		Corrosive in >1 of 3 animals	
	Subcategories	Exposure	Observation
Category 1: Corrosive	1A	≤ 3 min	≤ 1h
	1B	> 3 min - ≤ 1h	≤ 14 d
	1C	> 1 h - ≤ 4 h	≤ 14 d

In the available guideline study necrosis was observed 24 h after start of exposure (3 min or 1h) in 2 of 2 animals tested. Mean erythema and edema scores were 4 and 2, respectively. At the end of the observation period (day 8) leathery necrosis was documented.

Necrosis as well as severe erythema and moderate edema were also observed in a Draize test after 4h exposure (Virginia Chemicals, 1973). Another skin irritation study with an exposure duration of 24h showed heavy necrosis after 24h (Pennwalt Corp, 1986).

10.4.3 Conclusion on classification and labelling for skin corrosion/irritation

Necrosis, severe erythema and edema were observed in animal studies. Exposure of rabbits to 0.5 ml of undiluted dibutylamine for 3 min resulted in necrosis 24h after start of exposure. Based on the CLP criteria a classification as Skin Corr. 1B is indicated.

RAC evaluation of skin corrosion/irritation

Summary of the Dossier Submitter's proposal

For the evaluation of skin irritation/corrosion endpoint one OECD TG 404 study in rabbits, one Draize test, and one further skin irritation test in rabbits are available.

Necrosis, as well as severe erythema and oedema were observed in all of these studies. In the guideline study (Anonymous, 1978), exposure of rabbits to 0.5 ml undiluted di-n-butylamine for 3 min resulted in necrosis 24h after treatment. Further, necrosis, severe erythema and moderate oedema were also observed in a Draize test (Virginia Chemicals, 1973), and in a skin irritation study with an exposure duration of 24h (Pennwalt Corp, 1986). Based on these findings, and in accordance with the CLP criteria the DS proposed classification as Skin Corr. 1B, H314.

Method	Species	Exposure/ Dose levels	Main Findings	Reference
OECD TG 404 Non GLP	Rabbit, Vienna White N=2	0.5 ml for 3 min or 1 h Occlusive application on 2x2cm	Erythema score 4 (mean 24, 48, 72h), not reversible after either 3 min or 1 h exposure Oedema score 2 (mean 24, 48, 72h), not reversible after either 3 min or 1 h exposure Necrosis after 24h	Anonymous (1978)
Draize-method	Rabbit, New Zealand White N=6	0.5 ml for 4 h Clipped intact skin of one-inch square	Erythema score 4 (mean 24, 48h), not reversible Oedema score 1.58 (mean 24, 48h), max. score 3 Necrosis at all time points	Virginia chemicals, 1973
Skin irritation test	Albino rabbit N=3	0.5 ml for 24 h Clipped, intact and abraded skin	15 min: skin turned brown 24 h: large necrotic lesions 48 h: dry, hard lesions, cracking, raw rat tissue	Pennwalt Corp (1986)

Comments received during consultation

One MSCA supported the proposed classifications.

Assessment and comparison with the classification criteria

In a skin irritation/corrosion study according to OECD TG 404, two Vienna White rabbits (one male, one female) were dermally exposed (occlusive, on 2x2 cm clipped skin) to 0.5 ml of undiluted di-n-butylamine (>99.5%) for 3 min or 1 h (Anonymous, 1978). The test substance was removed after the end of exposure, and the observation period was 8 days. Grading scores and necrosis were recorded after 24, 48, and 72h of exposure. Severe erythema (individual and mean scores of 4) and slight to moderate oedema (individual and mean scores of 2) were reported uniformly in all animals at all time points (Table 14 in CLH report). Necrosis was first reported 24h after exposure, and leathery necrosis was observed at the end of the observation period of 8 days.

In a Draize test (no specific guideline mentioned), 6 NZ White rabbits were exposed to 0.5 ml of undiluted di-n-butylamine on clipped, intact skin for 4h (Virginia chemicals, 1973). Individual erythema/oedema scores and necrosis were recorded after 4, 24 and 48h of exposure (Table below).

Animal No.	Erythema score			Oedema score			Necrosis		
	4h	24h	48h	4h	24h	48h	4h	24h	48h
1	4	4	4	3	3	1	yes	yes	yes
2	4	4	4	2	2	1	yes	yes	yes
3	4	4	4	2	2	1	yes	yes	yes
4	4	4	4	2	2	1	yes	yes	yes
5	4	4	4	2	2	1	yes	yes	yes
6	4	4	4	2	2	1	yes	yes	yes

Necrosis and severe erythema were reported in all animals at all observation time points, as well as moderate oedema (mean score of 1.58, with a maximum of 3).

Large necrotic skin lesions were described in three Albino rabbits after exposure to 0.5 ml di-n-butylamine on intact and abraded skin site (clipped, occlusive) for 24 hours (Pennwalt Corp, 1986). The test item application caused pain, and the skin turned brown within 15 minutes of application. These lesions became dry, hard and concave within 48h. Subsequently, they cracked and peeled exposing the raw tissue.

The DS mentioned several secondary sources reporting corrosive effects, severe erythema, oedema and necrosis persisting through 72 h after exposure, however there were no specific details available.

Comparison with the criteria

According to CLP Annex I: 3.2.2.1.1.1., a "*substance is corrosive to skin when it produces destruction of skin tissue, namely, visible necrosis through the epidermis and into the dermis in at least one tested animal after exposure for up to 4 hours"*.

In the guideline skin irritation/corrosion study, exposure of two rabbits to 0.5 ml undiluted di-n-butylamine for 3 minutes resulted in skin necrosis in both animals 24h after the start of exposure. Based on the CLP criteria for sub-category 1B "*Corrosive responses in at least one animal following exposure >3 min and ⩽1 h and observations ⩽ 14 days*", RAC supports the DS proposal for classification as **Skin Corr. 1B, H314**.

10.5 Serious eye damage/eye irritation

Due to a classification of di-*n*-butylamine for Skin Corrosion Category 1 serious damage to eyes is implicit as reflected in the hazard statement for skin corrosion (H314: Causes severe skin burns and eye damage) (ECHA, 2017). However, a studies on eye irritation/corrosion are available and presented in the table below.

Table 16: Summary table of animal studies on eye corrosion/irritation

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance,**	**Dose levels duration of exposure**	**Results** **-Observations and time point of onset** **-Mean scores/animal** **-Reversibility**	**Reference**
OECD 405 Non GLP Reliability (REACH registration): 2, key study	Rabbit N=4	C-902 (dibutylamine)	0.1ml (not rinsed off) Observation 7 days	Conjunctivae score Mean 24h = 2.3 (max 3) Not fully reversible within 7 days Chemosis score Mean (24h) = 2.3 (max 3) Not fully reversible within 7 days Cornea opacity score Mean 24h = 4 (max 4) Not fully reversible within 7 days	Anonymous, 1985 OTS 0515257

Method, guideline, deviations if any	**Species, strain, sex, no/group**	**Test substance,**	**Dose levels duration of exposure**	**Results** **-Observations and time point of onset** **-Mean scores/animal** **-Reversibility**	**Reference**
-	Albino rabbits	dibutylamine	0.1ml in both eyes One eye was washed after 15 sec with water, other remained unwashed		Pennwalt Corp (1986) OTS0513616

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

In the available study (Anonymous, 1985) eyes of four rabbits were exposed to 0.1 ml of dibutylamine. The test substance produced severe ocular irritation in two of the four animals. After 24h severe conjunctival irritation (redness, chemosis, discharge, necrosis), iridial changes or iritis and corneal opacity, stippling and ulceration and corneal bulging (indicative of increased intraocular pressure) are described. No information on scoring at 48h and 72h is available. By day 7, two of the four animals still exhibited conjunctival irritation, iridial changes and corneal opacity, stippling and/or ulceration, corneal bulging and pannus (neovascilarization of the corneal surface). The other two animals exhibited only slight conjunctival irritation and/or iridial changes and stippling which were reversible after 7 d. Individual animal data are presented in the table below.

Table 17: eye irritation testing – individual scoring (Anonymous, 1985)

Effect		Animal #1, f		Animal #2, m		Animal #3, f		Animal #4, f	
		24h	Day 7	24h	Day 7	24h	Day 7	24h	Day 7
	Redness	3	1	2	1	2	1	2	1
Conjunctivae	Chemosis	3	2	2	1	2	1	2	1
	Discharge	3	1	3	0	3	0	3	0
	Necrosis	N	N	0	0	N	0	N	0
	Ulceration	0	0	0	0	0	0	0	0
	score	18	8	14	4	14	4	14	4
Iris	Iris	1	+	+	0	1	+	1	+
	score	5	0	0	0	5	0	5	0
Cornea	Opacity	3	1	1	0	2	1	2	0

	Area	4	3	2	0	4	4	4	0
	Stippling	2	2	2	0	2	1	2	1
	Ulceration	4	0	2	0	4	1	2	0
	Fluorescein	F	F	F	F	F	F	F	F
	Score	60	15	10	0	40	20	40	0
Totol score		83	23	24	4	59	24	59	4

Note: readability of the individual scoring in the available OTS documentation was limited

In the second study (Pennwalt Corp, 1986) 0.1ml of dibutylamine was placed in the conjunctival sac of both eyes of albino rabbits (n=3). After 15 seconds one eye of each animal was washed with water, the other eye remained unwashed. Scoring was done for a periode of 7 days. The reactions in the washed and unwashed eyes were identical. The mean scores are described in the table below. Signs of recovery were evident on day 6.

Table 18: Mean scores (washed and unwashed compiled) (Pennwalt Corp, 1986).

Time	Cornea	Iris	Conjunctivae redness	Conjunctivae chemosis
10min	0	<1	2	1
1h	0	<1	2	1
2h	0	<1	2	1
4h	0	<1	2	1
24h	0	<1	3	1
48h	0	<1	3	1
72h	0	<1	3	1
4d	0	<1	3	1
5d	0	<1	2	1
6d	0	0	1	0
7d	0	0	1	0

10.5.2 Comparison with the CLP criteria

Category 1	A substance that produces:

	(a) in at least one animal effects on the cornea, iris or conjunctiva that are not expected to reverse or have not fully reversed within an observation period of normally 21 days; and/or (b) in at least 2 of 3 tested animals, a positive response of: (i) corneal opacity ≥ 3 and/or (ii) iritis > 1,5 calculated as the mean scores following grading at 24, 48 and 72 hours after installation of the test material.
Category 2	Substances that produce in at least in 2 of 3 tested animals, a positive response of: (a) corneal opacity ≥ 1 and/or (b) iritis ≥ 1, and/or (c) conjunctival redness ≥ 2 and/or (d) conjunctival oedema (chemosis) ≥ 2 calculated as the mean scores following grading at 24, 48 and 72 hours after installation of the test material, and which fully reverses within an observation period of 21 days

For comparison with the given classification criteria only limited data is available. In the first study (Anonymous, 1985) scores at 24h were 4 for corneal opacity and about 1 for iritis in at least 2 of 4 animals (no scoring at 48/72h). Observations were only done till day 7, where no reversibility was reported for 2/4 animals. In a second study immediate conjunctivae redness, increasing till day 3 was observed.

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

Di-*n*-butylamine showed severe effects in the eyes of rabbits (conjunctivae chemosis and necrosis, cornea ulceration, cornea opacity). Due to limited reporting no final conclusion can be drawn based on the documented scoring in the study by Anomymous, 1985. In the second test a mean score (24-72h) for conjunctival redness of 3 is reported indicating classification as Category 2 irritant.

However the substance showed corrosive effects in skin irritation studies and is therefore proposed to be classified as Skin Corr. 1B. According to the CLP guidance (ECHA, 2017) serious damage to eyes is implicit indicated as Eye Dam 1, which is supported by the severe effects seen in the study by Anonymous (1985).

RAC evaluation of serious eye damage/eye irritation
Summary of the Dossier Submitter's proposal Two rabbit studies were available to DS for evaluation of this endpoint. In the first study

(Anonymous, 1985), severe effects on the eyes such as conjunctivae chemosis and necrosis, corneal ulceration, and corneal opacity were observed. Mean scores of 4 for corneal opacity and about 1 for iritis were reported in at least 2 out of 4 animals at 24h reading. No information on scoring at 48h and 72h was available. In the second study (Pennwalt Corp, 1986), a mean score of 3 for conjunctival redness was reported following exposure of both eyes of Albino rabbits to 0.1 ml of the test substance.

The DS noted that due to limited reporting of the study by Anonymous (1985), no conclusion could be drawn based on the single scoring at 24 h. In the second test, a mean score of 3 for conjunctival redness from the 24, 48 and 72h readings points to a classification as Category 2 eye irritant. However, in accordance with the CLP criteria, the substance is classified as Skin Corr. 1B, and therefore serious damage to the eyes is indicated and supports classification as Eye Dam 1, H318.

Method	**Species**	**Exposure/ Dose levels**	**Main Findings**	**Reference**
OECD TG 405 Non-GLP	Rabbits, N=4	0.1 ml (not rinsed off) Observation period of 7 days	Mean scores at 24h reading: • Conjunctivae: 2.3 (max. 3) • Chemosis: 2.3 (max 3) • Cornea opacity: 4 (max. 4) All effects were not fully reversible within 7 days	Anonymous, 1985
	Albino rabbits, N=3	0.1 ml in both eyes Only one eye washed after 15 sec	Mean score conjunctivae redness for 24, 48 and 72 h reading: 3 Scoring for 7 days, with signs of recovery on day 6.	Pennwalt Corp (1986)

Comments received during consultation

Two MSCA supported the proposed classification.

Assessment and comparison with the classification criteria

In a key study reported as similar to OECD TG 405 (Anonymous, 1985), severe ocular effects were observed in the eyes of two out of four rabbits exposed to 0.1 ml di-n-butylamine. Severe conjunctival irritation (redness, chemosis, discharge, and necrosis), iridial changes or iritis and corneal opacity, stippling and ulceration and corneal bulging (indicative of increased intraocular pressure) were described 24h after exposure. Conjunctival irritation, iridial changes and corneal opacity, stippling and/or ulceration, corneal bulging and pannus (neovascularization of the corneal surface) were still present in two of the four animals by the end of the observation period of day 7. Conjunctival irritation and/or iridial changes and stippling were reversible after 7 days in the remaining two animals.

RAC notes that the CLP criteria require assessment of the mean scores for corneal, iridial and conjunctival effects following grading at 24, 48 and 72 hours after installation of the test material, and an observation period of 21 days post exposure. In this study, observations were reported only until day 7, and no information on gradings at 48h and 72h is available. Therefore, a direct comparison with the classification criteria is not

possible.

In a second study (Pennwalt Corp, 1986), 0.1 ml of di-n-butylamine was installed in the conjunctival sac of both eyes of albino rabbits (n=3). After 15 seconds, one eye of each animal was washed out with water, while the other eye remained unwashed. The gradings were performed for 7 days, and reactions in the washed and unwashed eyes were identical. A mean score of 3 from the readings at 24, 48, and 72h for conjunctival redness was reported. Signs of recovery were evident on day 6 (Table 18 in the CLH report).

Comparison with the criteria

According to Annex I: 3.3.1.1. "*Serious eye damage means the production of tissue damage in the eye, or serious physical decay of vision, following application of a test substance to the anterior surface of the eye, which is not fully reversible within 21 days of application"*.

Both available studies allow only limited possibility for comparison with the CLP classification criteria. In the key study, observation period was 7 days with gradings reported only at 24h and day 7 post-exposure. In the second study (Pennwalt Corp, 1986), the mean score of 3 for conjunctival redness indicates a classification as Category 2 eye irritant, however reversibility within 21 days cannot be assed due to the shorter observation period. Thus, no firm conclusion can be drawn based on the reported data. Nevertheless, the classification of di-n-butylamine as Skin Corr. 1B implicitly entails a classification as Eye Dam 1 as stated in Regulation (EC) No 1272/2008. In line with the argumentation of DS, RAC agrees to classify di-n-butylamine as **Eye Dam. 1**. The corresponding hazard statement (H318: Causes serious eye damage) is not indicated on the label to avoid redundancy.

10.6 Respiratory sensitisation

Evaluation not performed for this substance.

10.7 Skin sensitisation

Evaluation not performed for this substance.

10.8 Germ cell mutagenicity

Evaluation not performed for this substance.

10.9 Carcinogenicity

Evaluation not performed for this substance.

10.10 Reproductive toxicity

Evaluation not performed for this substance.

10.11 Specific target organ toxicity-single exposure

For evaluation of STOT-SE acute toxicity studies are available. The studies are presented in Chapter 10.1, 10.2 and 10.3. For most of the studies only limited descriptions are available. No effects relevant for a classification as STOT SE 1 or 2 could be identified. However one acute inhalation toxicity study is available with relevant effects for respirator tract irritation. This study is described in detail below.

Table 19: Summary table of relevant animal studies on acute toxicity

Method, guideline, reliability	**Species, strain, sex, no/group**	**Test substance, form**	**Dose levels, duration of exposure**	**Value** **LC_{50}**	**Reference**
Acute inhalation toxicity Equivalent to OECD 403 GLP: yes Reliability (REACH registration): 2, key study Reliability (this assessment): 1	Rat, Sprague-Dawley 5 males and 5 females per dose group	Dibutylamine, as vapour Purity > 99.5 % No information on source	0, 0.76, 1.08, 1.18, 1.39, 3.91 mg/L 4 h exposure 14 days post exposure observation	1.15 mg/L mortalities C: m 0/5, f 0/5 0.76 mg/L: m 2/5, f 0/5 1.08 mg/L: m 0/5, f 2/5 1.18 mg/L: m 3/5, f 1/5 1.39 mg/L: m 5/5, f 5/5 3.91 mg/L: m 5/5, f 5/5 - partial closing of the eyes - reduced respiratory rate, - abnormal respiratory movements - adoption of an anormal body posture - gasping, exsessive salivation, lacrimation	Anonymous (1987) [Study 001, acute Tox, inhalation, REACH registration]

10.11.1 Short summary and overall relevance of the provided information on specific target organ toxicity – single exposure

In this guideline- and GLP-conform study rats were exposed to dibutylamine (vapour) in 5 different concentrations. After 4 h exposure, this study determined a LC50 of 1.15 mg/L. During exposure (concentration not indicated) the rats showed signs of sensory irritation like partial closing of the eyes, reduced respiratory rate, abnormal respiratory movements and adoption of an anormal body posture. Less frequently gasping, exsessive salivation, lacrimation and convulsion were observed. When removed from the test chamber previously exposed rats showed abnormal breathing, lethargy, ataxia, prone posture and

intermittent convulsions. Abnormal breathing, rales and sneezing were evident till day 2 with normal appearance on day 3.

10.11.2 Comparison with the CLP criteria

Category 1	Substances that have produced significant toxicity in humans or that, on the basis of evidence from studies in experimental animals, can be presumed to have the potential to produce significant toxicity in humans following single exposure Substances are classified in Category 1 for specific target organ toxicity (single exposure) on the basis of: a) reliable and good quality evidence from human cases or epidemiological studies; or b) observations from appropriate studies in experimental animals in which significant and/or severe toxic effects of relevance to human health were produced at generally low exposure concentrations. Guidance dose/concentration values are provided below (see 3.8.2.1.9) to be used as part of weight-of-evidence evaluation.
Category 2	Substances that, on the basis of evidence from studies in experimental animals can be presumed to have the potential to be harmful to human health following single exposure Substances are classified in Category 2 for specific target organ toxicity (single exposure) on the basis of observations from appropriate studies in experimental animals in which significant toxic effects, of relevance to human health, were produced at generally moderate exposure concentrations. Guidance dose/concentration values are provided below (see 3.8.2.1.9) in order to help in classification. In exceptional cases, human evidence can also be used to place a substance in Category 2 (see 3.8.2.1.6).
Category 3	Transient target organ effects This category only includes narcotic effects and respiratory tract irritation. These are target organ effects for which a substance does not meet the criteria to be classified in Categories 1 or 2 indicated above. These are effects which adversely alter human function for a short duration after exposure and from which humans may recover in a reasonable period without leaving significant alteration of structure or function. Substances are classified specifically for these effects as laid down in 3.8.2.2

There are currently no validated animal tests that deal specifically with respiratory tract irritation, however, useful information may be obtained from the single and repeated inhalation toxicity tests. For example, animal studies may provide useful information in terms of clinical signs of toxicity (dyspnoea, rhinitis etc) and histopathology (e.g. hyperemia, edema, minimal inflammation, thickened mucous layer) which are reversible and may be reflective of characteristic clinical symptoms. Such animal studies can be used as part of weight of evidence evaluation.

According to the CLP guidance (ECHA, 2017) it is a reasonable assumption that corrosive substances may also cause respiratory tract irritation when inhaled at exposure concentrations below those causing frank respiratory tract corrosion. If there is evidence from animal studies or from human experience to support this then Category 3 may be appropriate. In general, a classification for corrosivity is considered to implicitly cover the potential to cause RTI and so the additional Category 3 is considered to be

superfluous, although it can be assigned at the discretion of the classifier. The Category 3 classification would occur only when more severe effects in the respiratory system are not observed.

For evaluation of transient target organ effects one acute toxicity study (inhalation) is available, showing effects like partial closing of the eyes, reduced respiratory rate, abnormal respiratory movements and adoption of an anormal body posture. Less frequently gasping, exsessive salivation, lacrimation and convulsion were observed. These effects were fully reversible. Tissue changes were not investigated.

10.12 Conclusion on classification and labelling for STOT SE

The available acute toxicity study shows sensory irritation (reduced respiratory rate, abnormal respiratory movements) after short term 4h exposure of rats to vapour of di-n-butylamine. As the substance shows corrosive properties in skin irritation studies these irritant effects may be expected, when tested as vapour. A classification for corrosivity is considered to implicitly cover the potential to cause respiratory tract irritation but STOT SE 3 can be assigned in addition. Therefore, a classification as STOT SE Category 3 for di-*n*-butylamine is proposed.

RAC evaluation of specific target organ toxicity-single exposure

Summary of the Dossier Submitter's proposal

For evaluation of STOT SE, several acute toxicity studies are available. Reporting of the most studies is only limited, and no effects relevant for a classification as STOT SE 1 or 2 were identified by DS. However, signs of sensory irritation (reduced respiratory rate, abnormal respiratory movements) were reported in rats after short-term exposure to vapour of di-n-butylamine (Anonymous 1987). Specific doses at which these effects occurred were not indicated.

Acknowledging that classification for skin/eye corrosivity implicitly covers the potential of a substance to cause respiratory tract irritation, the DS proposed an additional classification as STOT SE 3; H335 to address effects at exposure concentrations below those causing frank respiratory tract corrosion.

Species	Dose levels (mg/L)	Reliability (DS)	Study	Remarks
Sprague-Dawley rat, 5/sex/dose	0, 0.76, 1.08, 1.18, 1.39, 3.91 4-h exposure	1	Anonymous 1987 OECD TG 403, GLP	Reported effects: • partial closing of the eyes • reduced respiratory rate, • abnormal respiratory movements • abnormal body posture • gasping, excessive salivation, lacrimation

Comments received during consultation

One MSCA and one manufacturer provided comments on this endpoint. The commenting MSCA disagreed with the proposed classification as STOT SE 3; H335 pointing out that the data is not sufficient to conclude on respiratory tract irritation, and a classification for corrosivity would implicitly cover these effects. The manufacturer disagreed with a classification of the pure substance arguing that respiratory tract irritation is already covered by corrosivity but proposed to classify mixtures as STOT SE 3; H335 including specific concentration limits of 1% ≤ C ≤ 5% to indicate the hazard at concentrations that are not corrosive.

Assessment and comparison with the classification criteria

RAC notes that the acute studies in animals did not reveal evidence of toxicity to any specific target organ that are not explicitly addressed under other hazard classes, which is a requirement for classification in categories 1 and 2. As regards classification with STOT SE 3 (narcotic effects), no narcotic effects were reported in any of the toxicity studies.

Classification STOT SE 3; H335 for respiratory tract irritation is primarily based on information from human studies, however no such data was identified for di-n-butylamine. While there are currently no validated animal tests dealing specifically with this endpoint, animal studies can be used as a part of a weight of evidence evaluation.

Signs of sensory irritation such as partial closing of the eyes, reduced respiratory rate, abnormal respiratory movements and abnormal posture are reported in an acute inhalation study with di-n-butylamine performed according to OECD TG 403 (Anonymous 1987). Gasping, excessive salivation, lacrimation and convulsions were observed less frequently. When removed from the test chamber, rats showed abnormal breathing, lethargy, ataxia, prone posture and intermittent convulsions. Abnormal breathing, rales and sneezing were evident until day 2 with normal appearance on day 3. These effects are consistent with sensory irritation and were fully reversible after cessation of exposure in surviving animals. Taking into account the corrosive nature of the substance, the described effects on the respiratory system seem to be associated with its irritating properties. However, the available acute toxicity data is not sufficient to conclude on respiratory tract irritation.

According to the CLP guidance (ECHA, 2017), "*a classification for corrosivity is considered to implicitly cover the potential to cause respiratory tract irritation and so the additional Category 3 is considered to be superfluous"*. RAC concludes that the classifications for acute inhalation toxicity and corrosivity adequately cover the toxicological profile of the substance for this endpoint. Consequently, **no classification for STOT SE is warranted**.

Supplemental information - In depth analyses by RAC

Expiratory bradypnea indicative of upper airway irritation was reported after acute inhalation exposure of male Swiss OF1 mice to vapour of di-n-butylamine (Gagnaire *et al*., 1993, as cited in TCEQ (Texas Commission on Environmental Quality) 2016). In these

experiments, the reduction of breathing frequency was used as an index of upper respiratory tract irritation as outlined by Alarie (1973). The calculated concentration resulting in a 50% decrease in respiratory rate (RD_{50}) in mice was 173 ppm, while in tracheal-cannulated mice (indicative of pulmonary sensory irritation) the RD_{50} was 106 ppm. Further, an RD_{50} of 81 ppm was reported in male Ssc:CF-1 mice, with an RD_{50} of 101 ppm in tracheal-cannulated mice (Nielsen and Yamagiwa 1989, as provided in TCEQ 2016).

RAC notes that the Alarie (1973) gives specific information on the potential for sensory irritation in rodents, however a direct correlation to irritation thresholds in humans is not sufficiently established (Guidance on IR&CSA, Section R.7.2.). It is further acknowledged that while the generic term respiratory tract irritation covers two different effects, 'sensory irritation' and 'local cytotoxic effects', classification in STOT SE 3; H335 is generally limited to local cytotoxic effects. No cytotoxic effects in the respiratory tract that are potentially relevant for STOT SE 3 classification (i.e. inflammation, haemorrhage, and necrosis) were reported in the studies above.

In a more recent study, Wistar rats (5/sex/dose) were exposed nose-only for 6 h/day to 0, 150, or 450 mg/m^3 of di-n-butylamine (purity 99%) for 3 days (Buschmann *et al*., 2003). The study was conducted in compliance with GLP, and toxicity was also assessed after exposures to 0, 50, 150, or 450 mg/m^3 for 28 d and 91 d (6 h/day, 5 days/week). These intervals were chosen in order to evaluate effects after acute, subacute and subchronic exposures. Rats were observed once daily for clinical symptoms, and histological examinations were done on the tissues of the respiratory tract (nasal cavity, larynx, laryngo-pharynx, trachea, lungs, and lung-associated lymph nodes). Bronchoalveolar lavage (BAL) was performed on the left lung lobe, and leukocytes were determined as well as differential cell count for percent of macrophages, granulocytes, and lymphocytes.

Dose (mg/m^3)	**0**	**150**	**450**
Animals	5M/5F	5M/5F	5M/5F
Observed lesions [a] (after 3 days exposure)			
Ulceration	0/10	0/10	5/10
Epithelial erosion(s)	0/10	0/10	9/10
Mucosal inflammatory cell infiltration	0/10	0/10	10/10
Squamous metaplasia of the respiratory epithelium	0/10	0/10	10/10
Mucus (goblet) cell hyperplasia	1/10	1/10	8/10
Submucosal haemorrhage	0/10	0/10	9/10
Mucosal/submucosal oedema	0/10	0/10	1/10
Respiratory epithelial hyperplasia	0/10	0/10	0/10

[a] There were no substantial sex differences so data for males and females were combined

Gross pathology findings were not observed, and BAL fluid analysis in exposed animals was not statistically significantly different from controls. In the histopathological examination, significant irritating effects (e.g., ulceration, epithelial erosion, mucosal inflammatory cell infiltration, squamous metaplasia of the respiratory epithelium) in the nasal cavities were observed only in animals at 450 mg/m^3 after 3 days of exposure. Notably, acute ulcerations and epithelial erosions were not evident after 28 and 91 days of exposure. The authors indicated that this might be due to the increased production of mucus as an adaptive response to prolonged DBA exposure. In the lung, only slight, not

statistically significant histopathological effects were observed.

RAC notes that significant respiratory irritation was observed in Wistar rats after 3 days (6h/d) of nose-only inhalation exposure to 0.45 mg/L di-n-butylamine. Observations after a single exposure, at shorter exposure durations, and information on potential reversibility of these effects are not available. However, RAC considers that the dose level of 0.45 mg/L where these effects were observed is close to the LC_{50} of 1.15 mg/L and classified for acute inhalation toxicity. Furthermore, in the acute inhalation study (Anonymous, 1987), two males died already at the lowest exposure concentration of 0.76 mg/L. Considering the above, RAC concludes that the data does not warrant additional classification as STOT SE 3; H335 for respiratory tract irritation.

Additional labelling

According to Annex I: 3.1.2.3.3. of the CLP regulation, "*In addition to the application of the classification for acute inhalation toxicity, the substance or mixture must also be labelled as EUH071 where data are available which indicate that the mode of toxic action was corrosivity (see Note 1 to Table 3.1.3)"*.

There is no specific data on the mode of action of di-n-butylamine provided in the CLH dossier, and the DS did not propose any additional labelling. The substance is strongly alkaline with a dissociation constant (pKa) of 11, and the reported effects on rabbit skin (necrosis after exposure for 3 minutes to undiluted substance) and eye (severe conjunctival reactions such as redness, chemosis, discharge and necrosis) are consistent with its corrosive properties. A common MoA for amines with a high pKa is the release of a hydroxide ion after protonation causing local necrosis upon contact with tissues at physiologic pH. Thus, for the category of aliphatic secondary amines, the corrosive properties are expected to be a general feature in relation to acute toxicity and respiratory tract effects (OECD 2013).

Based on the above, RAC considers that there is relevant evidence indicating that the mechanism of toxicity is corrosivity and therefore proposes **additional labelling with the EUH071: Corrosive to the respiratory tract**.

10.13 Specific target organ toxicity-repeated exposure

Evaluation not performed for this substance.

10.14 Aspiration hazard

Evaluation not performed for this substance.

11 EVALUATION OF ENVIRONMENTAL HAZARDS

Evaluation not performed for this substance.

12 EVALUATION OF ADDITIONAL HAZARDS

Evaluation not performed for this substance.

13 ADDITIONAL LABELLING

Not applicable for this substance.

14 REFERENCES

AGS, Ausschuss für Gefahrstoffe (2006). Begründung zu Di-*n*-butylamin in TRGS 900. Stand: Februar 1992 online: https://www.baua.de/DE/Angebote/Rechtstexte-und-Technische-Regeln/Regelwerk/TRGS/pdf/900/900-di-n-dibutylamin.pdf?__blob=publicationFile

Anonymous (1985). REACH registration data. https://echa.europa.eu/de/registration-dossier/-/registered-dossier/13527/7/4/3. TSCA submission OTS 0515257.

Anonymous (1987). REACH registration data. https://echa.europa.eu/de/registration-dossier/-/registered-dossier/13527/7/3/3

Ciugudeanu M., Gabor S., Gocan M., Ivanescu G., Buzas A. (1985). Experimental researches regarding the acute toxicity of butanol monoamine and diamine and tributylamine . Revista de Chimie, 36, 667-670.

ECHA (2017). Guidance on the application of the CLP criteria. Version 5.0 https://echa.europa.eu/documents/10162/23036412/clp_en.pdf/58b5dc6d-ac2a-4910-9702-e9e1f5051cc5

ECHA Dissemination site (2019). Information on Chemicals - Registered Substances, European Chemicals Agency. Online: http://echa.europa.eu/web/guest/information-on-chemicals/registered-substances

Greim H., Bury D., Klimisch H.J., Oeben-Negele M., Ziegler-Skylakakis K. (1998). Toxicity of aliphatic amines: structure-activity relationship. Chemosphere, 36, 271-295.

Pennwalt CORP (1986). Eye & Skin irritancy in rabbits using dibutylamine. TSCA submission OTS0513616

Sax N.I., Lewis R.J. (1989). Dangerous Properties of Industrial Materials, 7th ed., Van Nostrand Reinhold New York.

Schmidt P., Burck D., Weigmann H.J. (1974). Zur akuten Toxizitat einiger für die Kunststoffindustrie bedeutsamer Amine [Acute toxicity of some for plastic processing industry important amines]. Zeitschrift für die gesamte Hygiene und ihre Grenzgebiete, 20, 393-398.

Smyth H.F., Carpenter C.P., Weil C.S., Pozzani U.C. (1954). Range-finding toxicity data: list V. Archives of Industrial Hygiene and Occupational Medicine, 10, 61-68.

TCEQ, Texas Commission on Environmental Quality (2016). Dibutylamine. Development Support Document. Final, July 8, 2016. Prepared by Roberta L. Grant, Jong-Song Lee. https://www.tceq.texas.gov/assets/public/implementation/tox/dsd/final/may16/dibutylamine.pdf

Virginina Chemicals (1973). Skin corrosivity of di-*n*-butylamine in rabbits. OTS0515256.

Additional references
Y. Alarie, Sensory irritation by airborne chemicals. CRC Crit. Rev. Toxicol. 2, 299-363 (1973). Buschmann, J, C Dasenbrock, R Fuhst, G Pohlmann, W Bartsch, O Creutzenberg, H Ernst, M Ketkar, B Schneider, H Ott. 2003. Effects of di-n-butylamine on the

respiratory system of Wistar (WU) rats after subchronic inhalation. Inhal Toxicol 15:701-13.

Gagnaire, F, S Azim, P Simon, B Cossec, P Bonnet, and J De Ceaurriz. 1993. Sensory and pulmonary irritation of aliphatic amines in mice: A structure-activity relationship study. J Applied Toxicology 13: 129-135

Nielsen, GD, and M Yamagiwa. 1989. Structure-activity relationships of airway irritating aliphatic amines. Receptor activation mechanisms and predicted industrial exposure limits. Chem.-Biol. Interactions 71: 223-244.

Organisation for Economic Co-operation and Development (OECD). 2013. CoCAM4, 16-18 April 2013. US/ICCA. SIDS Initial Assessment Profile, Aliphatic Secondary Amines