Committee for Risk Assessment

RAC

Opinion

proposing harmonised classification and labelling
at EU level of

methyl-1*H*-benzotriazole

EC Number: 249-596-6
CAS Number: 29385-43-1

CLH-O-0000007149-69-01/F

Adopted

15 September 2022

15 September 2022

CLH-O-0000007149-69-01/F

OPINION OF THE COMMITTEE FOR RISK ASSESSMENT ON A DOSSIER PROPOSING HARMONISED CLASSIFICATION AND LABELLING AT EU LEVEL

In accordance with Article 37 (4) of Regulation (EC) No 1272/2008, the Classification, Labelling and Packaging (CLP) Regulation, the Committee for Risk Assessment (RAC) has adopted an opinion on the proposal for harmonised classification and labelling (CLH) of:

Chemical name: **methyl-1*H*-benzotriazole**

EC Number: **249-596-6**

CAS Number: **29385-43-1**

The proposal was submitted by **Germany** and received by RAC on **20 August 2021.**

In this opinion, all classification and labelling elements are given in accordance with the CLP Regulation.

PROCESS FOR ADOPTION OF THE OPINION

Germany has submitted a CLH dossier containing a proposal together with the justification and background information documented in a CLH report. The CLH report was made publicly available in accordance with the requirements of the CLP Regulation at ***http://echa.europa.eu/harmonised-classification-and-labelling-consultation/*** on **8 November 2021**. Concerned parties and Member State Competent Authorities (MSCA) were invited to submit comments and contributions by **21 January 2022**.

ADOPTION OF THE OPINION OF RAC

Rapporteur, appointed by RAC: **Riitta Leinonen**

The opinion takes into account the comments provided by MSCAs and concerned parties in accordance with Article 37(4) of the CLP Regulation and the comments received are compiled in Annex 2.

The RAC opinion on the proposed harmonised classification and labelling was adopted on **15 September 2022** by **consensus**.

Classification and labelling in accordance with the CLP Regulation (Regulation (EC) 1272/2008)

	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	No current Annex VI entry										
Dossier submitters proposal	TBD	methyl-1*H*-benzotriazole	249-596-6	29385-43-1	Aquatic Chronic 2	H411	GHS09 Wng	H411			
RAC opinion	TBD	methyl-1*H*-benzotriazole	249-596-6	29385-43-1	Aquatic Chronic 2	H411	GHS09 Wng	H411			
Resulting Annex VI entry if agreed by COM	TBD	methyl-1*H*-benzotriazole	249-596-6	29385-43-1	Aquatic Chronic 2	H411	GHS09 Wng	H411			

GROUNDS FOR ADOPTION OF THE OPINION

ENVIRONMENTAL HAZARD EVALUATION

RAC evaluation of aquatic hazards (acute and chronic)

Summary of the Dossier Submitter's proposal

Methyl-1*H*-benzotriazole (CAS 29385-43-1) is not listed in Annex VI of the CLP Regulation. It is is a multiconstituent substance containing two isomers, 5-methyl-1*H*-benzotriazole (CAS 136-85-6) and 4-methyl-1*H*-benzotriazole (CAS 29878-31-7)). The Dossier Submitter (DS) proposed to classify the substance with Aquatic Chronic 2, H411 based on a 21-day EC_{10} of 0.40 mg/L for *Daphnia galeata* derived with the isomer 5-methyl-1*H*-benzotriazole and the application of the scheme outlined in Figure 4.1.1 of the CLP Regulation.

Degradation

In a ready biodegradability test (OECD TG 301 F, GLP), methyl-1*H*-benzotriazole (>99.9%) only 4 % degraded in 28 days, even though the inoculum was adapted. The DS concluded that the substance is not readily biodegradable.

Methyl-1*H*-benzotriazole (>99%) was hydrolytically stable in an OECD TG 111 (GLP) hydrolysis test at pH4, pH7 and pH9. Concentration of the test substance after 5 days was approximately 100%.

The biodegradability of 5-methyl-1*H*-benzotriazole and a technical mixture of 5-methyl-1*H*-benzotriazole and 4-methyl-1*H*-benzotriazole were investigated according to ISO 7827. The individual test substances were used as sole substrate with initial concentrations of 47.8 mg/L (DOC = 30 mg/L). The inoculum, fresh activated sludge, was taken from a lab-scale membrane bioreactor, which was fed with the effluent of the primary treatment of a wastewater treatment plant (mainly domestic wastewater, 30 % industrial input). Adaption of the inoculum was not specified. Complete removal of 5-methyl-1*H*-benzotriazole was observed within 17 days (based on concentrations of test substance). The test with the technical mixture showed similar results for 5-methyl-1*H*-benzotriazole.

Regarding the biodegradability of 4-methyl-1*H*-benzotriazole isomer, the same test (ISO 7827) derived 25 % of degradation (based on concentrations of test substance) within 28 days. No degradation of the test substance was observed in the sterile abiotic control confirming that all detected effects were due to microbial activity. The DS considered that the reliability of the ISO 7827 study was restricted due to missing information on purity of the test substances.

Two inherent biodegradability tests (OECD TG 302B) were available. Methyl-1*H*-benzotriazole degraded 77 % (in the dark) and 59 % (illuminated by daylight) after 28 days (7-day pass level: 70 and 59 %). Adaption of the inoculum was not stated. In the other test with 4-methyl-1*H*-benzotriazole no degradation, based on DOC, was observed during 28 days in the dark. Information on adaption of the inoculum was missing.

An atmospheric half-life of 3.89 days was calculated (AOPWIN v1.91) for the reaction of Methyl-1*H*-benzotriazole with OH radicals.

Based on all available information, the DS concluded methyl-1*H*-benzotriazole was not rapidly degradable.

Bioaccumulation

There was no bioconcentration test data available for fish. The log K_{ow} of 1.08 ± 0.002 from an OECD TG 117 test showed low potential for bioaccumulation for methyl-1*H*-benzotriazole, according to the DS.

Aquatic toxicity

Acute

***Table**: Summary of reliable information on acute toxicity*

Species	Substance	Results [mg/L]	Test method	Reliability/Reference
Fish				
Cyprinodon variegatus	methyl-1*H*-benzotriazole CAS 29385-43-1	96h-LC_{50}= 55	PARCOM 1995 Part B Protocol for a Fish Acute Toxicity Test (modified OECD TG 203 Fish Acute Toxicity Test) (GLP); no analytical monitoring; WAF-test; semi-static; saltwater;	Reliability 1 Registration dossier: (Anonymous, 2003a)
Invertebrates				
Daphnia magna	5-methyl-1*H*-benzotriazole CAS 136-85-6	48h-EC_{50}= 51.6 (nom.)	OECD TG 202; static; analytical monitoring	Reliability 1 Seeland et al., 2012 (*
Daphnia galeata	5-methyl-1*H*-benzotriazole CAS 136-85-6	48h-EC_{50}= 8.58 (nom.)	OECD TG 202; static; analytical monitoring	Reliability 1 Seeland et al. 2012 (*
Acartia tonsa	methyl-1*H*-benzotriazole CAS 29385-43-1	48h-EC_{50}= 55	ISO/CD 14669: Determination of Acute Lethal Toxicity to Marine Copepads and PARCOM Ring Test Protocol: Acute Toxicity to the Marine Copepod Acartia tonsa. (GLP); no analytical monitoring; WAF-test; static; saltwater	Reliability 1 Registration dossier: (Anonymous, 2003b)
Algae				
Skeletonema costatum	methyl-1*H*-benzotriazole CAS 29385-43-1	72h-E_rC_{50}= 53	ISO 10253 (Water quality - Marine Algal Growth Inhibition Test with Skeletonema costatum and Phaeodactylum tricornutum) (GLP); static; saltwater; no analytical monitoring; WAF-test.	Reliability 1 Registration dossier: (Anonymous, 2003c)

(*the test concentrations were analytically verified in a separate stability test. The deviation from nominal concentrations was only 6.3 to 7.0 %.

The only acute toxicity test available for fish gave a 96-hour LC_{50} of 55 mg/L for methyl-1*H*-benzotriazole.

For aquatic invertebrates, there were two studies available (*Daphnia magna, Daphnia galeata*) with 5-methyl-1*H*-benzotriazole. In addition, there was a study with *Acartia tonsa* with methyl-1*H*-benzotriazole. The lowest short-term result for aquatic invertebrates was derived from the test with *Daphnia galeata* (48h-EC_{50} of 8.58 mg/L). *Daphnia magna* was less sensitive with a 48h-EC_{50} of 51.6 mg/L.

The only acute result for algae was a 72-hour ErC_{50} of 53 mg/L for *Skeletonema costatum* with methyl-1*H*-benzotriazole.

The lowest short-term result was a 48-hour EC_{50} of 8.58 mg/L (5-methyl-1*H*-benzotriazole) for *Daphnia galeata*.

All aquatic acute toxicity values were above the classification cut-off 1 mg/L and thus no aquatic acute classification was proposed by the DS.

Chronic

Table: Summary of reliable data on chronic toxicity

Species	substance	Results [mg/L]	Test method	Reliability/Reference
Fish – no studies				
Invertebrates				
Daphnia magna	methyl-1*H*-benzotriazole CAS 29385-43-1	21d-NOEC= 18.4 (nom.)	OECD TG 202, Part II (Draft 7/1993) (GLP); semi-static; analytical monitoring	Reliability 1 Registration dossier: Anonymous, 1995b
Daphnia magna	5-methyl-1*H*-benzotriazole CAS 136-85-6	21d-EC_{10}= 5.93 (nom.)	OECD TG 211; semi-static; analytical monitoring	Reliability 2 Seeland et al., 2012
Daphnia galeata	5-methyl-1*H*-benzotriazole CAS 136-85-6	**21d-EC_{10}= 0.40 (nom.)** 21d-NOEC=1.0 (nom.)	OECD TG 211; semi-static; analytical monitoring	Reliability 2 Seeland et al., 2012
Algae and aquatic plants				
Desmodesmus subspicatus (previous name: *Scenedesmus subspicatus*)	5-methyl-1*H*-benzotriazole CAS 136-85-6	72h-E_rC_{10} = 2.86 (nom.)	OECD TG 201; static; concentrations; analytical monitoring	Reliability 2 Seeland et al., 2012
Skeletonema costatum	methyl-1*H*-benzotriazole CAS 29385-43-1	72h-NOE_rC= 30	ISO 10253 (Water quality - Marine Algal Growth Inhibition Test with Skeletonema costatum and Phaeodactylum tricornutum) (GLP); static; saltwater; no analytical monitoring; WAF-test	Reliability 1 Registration dossier: (Anonymous, 2003c)
Lemna minor	5-methyl-1*H*-benzotriazole CAS 136-85-6	7d-EC_{10}= 2.11	OECD TG 221; static; analytical monitoring	Reliability 2 Seeland et al., 2012

*Test concentrations were analytically verified in a separate stability test. The deviation from nominal concentrations was only 6.3 to 7.0 %.

There were no chronic toxicity data on fish available for methyl-1*H*-benzotriazole.

For invertebrates there was one study available for methyl-1*H*-benzotriazole and two studies for 5-methyl-1*H*-benzotriazole. The lowest chronic toxicity value was a 21-day EC_{10} of 0.40 mg/L for *Daphnia galeata* with 5-methyl-1*H*-benzotriazole.

There was one study on algae available for methyl-1*H*-benzotriazole, whilst for 5-methyl-1*H*-benzotriazole there were studies available for algae and aquatic plant. The lowest chronic toxicity value was a 7-day EC_{10} of 2.11 mg/L for *Lemna minor* with 5-methyl-1*H*-benzotriazole.

Based on Figure 4.1.1 of the CLP Regulation, the aquatic chronic classification is based on the most stringent outcome of the two assessments according to Table 4.1.0 (b) (i) and (iii). The lowest value available was a 21-day EC_{10} of 0.40 mg/L 5-methyl-1*H*-benzotriazole with *Daphnia galeata* which warrants an **Aquatic Chronic 2**, **H411** classification for a not rapidly degradable substance (0.1 mg/L<EC_{10}≤ 1 mg/L, Table 4.1.0 (b)(i)).

Comments received during consultation

Two Member States supported the proposed classification. A National Authority (NA) asked for further information on the solubility of the test item in test media and actual tests concentrations in the acute Water Accommodated Fraction tests. The DS answered that no more data was available although the substance had a water solubility of 4049.4 mg/L in the OECD TG 105 solubility test.

In addition, the NA asked for clarifications on the appropriateness of the use of data performed on *Daphnia galeata* for aquatic hazard classification. According to the NA, OECD TG 211 states: 'Other daphnids may be used provided they meet the validity criteria as appropriate (the validity criterion relating to the reproductive output in the controls should be relevant for all species)'.

- The NA asked the Dossier Submitter for a confirmation of the test protocol validity criterion regarding 'the mortality of the parent animals (female *Daphnia*) does not exceed 20% at the end of the test'.
- According to NA, the second validity criterion of the test protocol regarding 'the mean number of living offspring produced per parent animal surviving at the end of the test above 60' was not met. NA however, considered that the *Daphnia galeata* brood size may be smaller than that of *Daphnia magna* given the smaller physical size of the organism. Therefore, they were unclear whether this cut off is appropriate to assess the reproductive output of this species.
- The NA noted also that the percentage or number of dead offspring are also not reported. If this information would be available, it would be useful to calculate the coefficient of variation (CoV) of living offspring as an indicator of experimental reliability.

The DS confirmed that the *Daphnia galeata* species is smaller than *Daphnia magna*. Seeland et al 2021 describes that the difference in brood size was coherent with different body length for the species. Another publication[1] compared both species, e.g. in their life span, first brood, total number of offspring.

[1] Cui, R., Kwak, J.I., & An, Y. (2016). Characteristics and Toxicity Sensitivity of Korean Dominant Species Daphnia galeata for Ecotoxicity Testing: Comparative Study with Daphnia magna. Journal of Korean Society of Environmental Engineers, 38, 193-200. https://doi.org/10.4491/KSEE.2016.38.4.193

Table 2. Life span, first brood, total number of offspring per adult, and the number of offspring per broods of *Daphnia galeata* and *Daphnia magna*.

	Daphnia galeata[a]	*Daphnia magna*[29]
Life span (days)	28±8	50,1±10,9
First brood (days)	9±2	9,0±0,0
Total number of offspring per adult	29±23	357,8±104,7
Number of offspring per brood	4±2	13,3±3,7

[a] Data from this study

The mean number of juveniles in the control was 37 for *Daphnia galeata* in Seeland et al., 2012 in comparison with 99 for *Daphnia magna.* Taking into account the findings in Cui et al., 2016, this reproductive output of *Daphnia galeata* appears to be normal and appropriate for the assessment for toxicity testing.

In their comments, the NA argued that the NOEC might be a preferable key effects endpoint for *Daphnia galeata* for hazard classification regarding that the 21-day EC_{10} of 0.4 mg/L is below the 21-day NOEC of 1 mg/L and has confidence intervals of 0.08 – 1.95 mg/L which span the CLH classification band.

The DS did not, however, consider using the NOEC value necessary, as EC_{10} values are generally preferred over NOECs.

Finally, the NA wondered if the Fish Sexual Development Test (OECD TG 234) test for 1*H*-benzotriazole (CAS: 95-14-7) can be used for the purpose of aquatic hazard classification and noted that the study is currently undergoing review as part of the ongoing REACH Substance Evaluation.

The DS agreed that the study could be considered within the overall weight of evidence. RAC decided not to take this study into account for the reasons explained later in this document.

Assessment and comparison with the classification criteria

Comparison with the criteria

Methyl-1*H*-benzotriazole (29385-43-1) is a multiconstituent substance containing two isomers, 5-methyl-1*H*-benzotriazole (CAS 136-85-6) and 4-methyl-1*H*-benzotriazole (CAS 29878-31-7).

Degradation

RAC agrees with the DS conclusion that methyl-1*H*-benzotriazole is not rapidly degradable based on:

- No ready biodegradation test available with non-adapted inoculum.
- 4 % degradation in 28 days in an OECD TG 301 F ready biodegradability test.
- No surface water simulation test available.
- Methyl-1*H*-benzotriazole being hydrolytically stable at pH 4, pH 7 and pH 9.
- 25% of 4-methyl-1*H*-benzotriazole was degraded within 28 days in a technical mixture in ISO 7827 study.
- No degradation of 4-methyl-1*H*-benzotriazole in an OECD TG 302 B test.

Therefore, RAC agrees with the DS conclusion that methyl-1*H*-benzotriazole can be considered not rapidly degradable.

Bioaccumulation

RAC agrees with the DS conclusion that methyl-1*H*-benzotriazole has low potential for bioaccumulation even in the absence of an experimental fish bioconcentration study, as the measured low Pow of 1.081 ± 0.002 was below the classification cut-off value of 4.

Aquatic toxicity

Daphnia magna vs Daphnia galeata

RAC notes that regarding the use of other daphnids than *Daphnia magna* in the OECD TG 211 study, the validity criteria of the test guideline should be met. RAC recognises however, that the fulfilment of the validity criterion in the control(s) 'the mortality of the parent animals (female Daphnia) does not exceed 20% at the end of the test' was not explicitly mentioned in the Seeland et al 2012 article available for RAC.

RAC notes also that the test protocol validity criterion regarding the mean number of living offspring produced per parent animal surviving at the end of the test being above 60 was not met. RAC, however, agrees that considering the findings from Seeland et al 2021 and Cui et al 2016 articles as presented by the DS, the reproductive output of *Daphnia galeata* appears to be appropriate for the assessment for toxicity testing.

As indicated in the ECHA's CLP Guidance (v5.0, July 2017) the EC_{10} values are preferred over NOEC values for chronic toxicity studies when both are available for the same endpoint, therefore RAC agrees to use EC_{10} of 0.40 mg/L with confidence limits of 0.08 – 1.95 mg/L as a key endpoint for *Daphnia galeata* but also recognises that the NOEC of 1 mg/L would lead to a same classification.

Classification approach used by RAC

There were data available both for methyl-1*H*-benzotriazole and one of its ingredients 5-methyl-1*H*-benzotriazole. RAC agrees with the DS that data on 5-methyl-1*H*-benzotriazole is relevant for classification in the absence of data on chronic toxicity of methyl-1*H*-benzotriazole for the most sensitive species (*Daphnia galeata*).

There is no toxicity data available for 4-methyl-1*H*-benzotriazole in the CLH Report but Pillard et al 2001 concluded that all test organisms tested (*Ceriodaphnia dubia, Pimephales promelas*) were more sensitive to 5-methyl-1*H*-benzotriazole than to 4-methyl-1*H*-benzotriazole both in acute and chronic tests.

Acute

RAC agrees with the DS in concluding that **acute aquatic classification is not warranted** for methyl-1*H*-benzotriazole. All reliable LC/EC_{50} values both for methyl-1*H*-benzotriazole and 5-methyl-1*H*-benzotriazole (fish, *Daphnia magna, Daphnia galeata* and algae) were over the cut-off value of 1 mg/L for classification, the lowest value being a 48-hour EC_{50} of 8.58 mg/L for *Daphnia galeata*.

Chronic

For methyl-1*H*-benzotriazole there were reliable chronic toxicity data available for *Daphnia magna* and *Skeletonema costatum*. For 5-methyl-1*H*-benzotriazole there were data on *Daphnia magna*, *Daphnia galeata, Desmodesmus subspicatus* and *Lemna minor*. The surrogate approach for fish for which there were no chronic fish data on methyl-1*H*-benzotriazole available based on the 96-hour LC_{50} of 55 mg/L methyl-1H-benzotriazole would warrant an Aquatic Chronic 3 classification (Table 4.1.0 (b) (iii) of the CLP Regulation).

RAC is of the view that the use of the Fish Sexual Development Test (OECD TG 234) result of 1.07 mg/L for 1*H*-benzotriazole (CAS: 95-14-7) in the place of the missing information on fish

chronic toxicity is not appropriate in this specific case, where there is available reliable acute data on the substance itself to be used as a surrogate approach. Moreover, the acute data leads to a chronic classification whereas the use of data on 1*H*-benzotriazole does not and such approaches are typically not supported by the RAC for removing a classification.

The lowest value available was a 21-day EC_{10} of 0.40 mg/L 5-methyl-1*H*-benzotriazole with *Daphnia galeata* which warrants an Aquatic Chronic 2, H411 classification for a not rapidly degradable substance (0.1 mg/L<EC_{10}≤ 1 mg/L, Table 4.1.0 (b)(i)).

RAC agrees with the Dossier Submitter's proposal to **classify the substance as Aquatic Chronic 2.**

Additional references

Pillard DA et al. (2001) Toxicity of Benzotriazole and Benzotriazole derivatives to three aquatic species, Wat Res, 35, 2, 557-560.

Seeland, A.; Oetken, M.; Kiss, A.; Fries, E.; Oehlmann, J. 2012. Acute and chronic toxicity of benzotriazoles to aquatic organisms; Environ Sci Pollut Res: (19) p.1781-1790.

ANNEXES:

Annex 1 The Background Document (BD) gives the detailed scientific grounds for the opinion. The BD is based on the CLH report prepared by the Dossier Submitter; the evaluation performed by RAC is contained in 'RAC boxes'.

Annex 2 Comments received on the CLH report, response to comments provided by the Dossier Submitter and RAC (excluding confidential information).