

Helsinki, 12 October 2022

Addressee

Registrant of N-Butylpyrrolidone as listed in Appendix 3 of this decision

Date of submission of the dossier subject to this decision

22/03/2022

Registered substance subject to this decision ("the Substance")

Substance name: 1-butylpyrrolidin-2-one

EC number: 222-437-8

Decision number: Please refer to the REACH-IT message which delivered this communication (in format TPE-D-XXXXXXXXXX-XX-XX/F)**DECISION ON TESTING PROPOSAL(S)**

Under Article 40 of Regulation (EC) No 1907/2006 (REACH), you must submit the information listed below by **20 October 2025**.

Requested information must be generated using the Substance unless otherwise specified.

Information required from all the Registrants subject to Annex IX of REACH

1. Extended one-generation reproductive toxicity study also requested below (triggered by Annex IX, Section 8.7.3., column 1)

Information required from all the Registrants subject to Annex X of REACH

2. Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.; test method: EU B.56./OECD TG 443) by oral route, in rats, specified as follows:

- At least two weeks pre-mating exposure duration for the parental (P0) generation;
- The highest dose level in P0 animals must be determined based on clear evidence of an adverse effect on sexual function and fertility without severe suffering or deaths in P0 animals as specified further in Appendix 1, or follow the limit dose concept. The reporting of the study must provide the justification for the setting of the dose levels;
- Cohort 1A (Reproductive toxicity);
- Cohort 1B (Reproductive toxicity) with extension to mate the Cohort 1B animals to produce the F2 generation which shall be followed to weaning; and
- Cohort 3 (Developmental immunotoxicity).

You must report the study performed according to the above specifications. Any expansion of the study must be scientifically justified.

The reasons for the decision(s) are explained in Appendix 1.

Information required depends on your tonnage band

You must provide the information listed above for all REACH Annexes applicable to you in accordance with Articles 10(a) and 12(1) of REACH. The addressee of the decision and its corresponding information requirements based on registered tonnage band are listed in Appendix 3.

In the requests above, the same study has been requested under different Annexes. This is because some information requirements may be triggered at lower tonnage band(s). In such cases, only the reasons why the information requirement is triggered are provided for the lower tonnage band(s). For the highest tonnage band, the reasons why the standard information requirement is not met and the specification of the study design are provided. Only one study is to be conducted.

You are only required to share the costs of information that you must submit to fulfil your information requirements.

How to comply with your information requirements

To comply with your information requirements, you must submit the information requested by this decision in an updated registration dossier by the deadline indicated above. You must also **update the chemical safety report, where** relevant, including any changes to classification and labelling, based on the newly generated information.

You must follow the general requirements for testing and reporting new tests under REACH, see Appendix 4.

Appeal

This decision, when adopted under Article 51 of REACH, may be appealed to the Board of Appeal of ECHA within three months of its notification to you. Please refer to <http://echa.europa.eu/regulations/appeals> for further information.

Failure to comply

If you do not comply with the information required by this decision by the deadline indicated above, ECHA will notify the enforcement authorities of your Member State.

Authorised¹ under the authority of Mike Rasenberg, Director of Hazard Assessment

Appendix 1: Reasons for the decision

Appendix 2: Procedure

Appendix 3: Addressees of the decision and their individual information requirements

Appendix 4: Conducting and reporting new tests under REACH

¹ As this is an electronic document, it is not physically signed. This communication has been approved according to ECHA's internal decision-approval process.

Appendix 1: Reasons for the decision

Contents

Reasons for the decision(s) related to the information under Annex IX of REACH	4
1. Extended one-generation reproductive toxicity study	4
Reasons for the decision(s) related to the information under Annex X of REACH	5
2. Extended one-generation reproductive toxicity study	5
References	9

Reasons for the decision(s) related to the information under Annex IX of REACH**1. Extended one-generation reproductive toxicity study**

- 1 An extended one-generation reproductive toxicity study (EOGRTS; OECD TG 443) is an information requirement under Annex IX to REACH (Section 8.7.3.) if the available repeated dose toxicity studies indicate adverse effects on reproductive organs or tissues or reveal other concerns in relation with reproductive toxicity.
- 2 Your dossier contains an OECD TG 421 study (2021, conducted with the Substance), which indicates concerns in relation to reproductive toxicity. Specifically, shorter mean anogenital distances (absolute and normalized for body weight) were noted for male pups in the 325 and 500 mg/kg/day groups, when compared to the concurrent control group. The differences at 500 mg/kg/day were statistically significant and the absolute values in both groups (3.19 and 2.90 mm, respectively) were below the minimum mean value (3.4 mm) of the historical control data. The observed changes in the anogenital distance were test-item related.
- 3 Therefore, the concern for reproductive toxicity must be further investigated.
- 4 ECHA agrees that an EOGRTS is necessary to address the identified concerns in relation with reproductive toxicity.
- 5 For the assessment of the testing proposal, see request 2.

Reasons for the decision(s) related to the information under Annex X of REACH**2. Extended one-generation reproductive toxicity study**

6 The basic test design of an extended one-generation reproductive toxicity study (EOGRTS) is a standard information requirement under Annex X to the REACH Regulation. Furthermore, column 2 of Section 8.7.3. defines when the study design needs to be expanded.

2.1. Information provided to fulfil the information requirement

7 You have submitted a testing proposal for an EOGRTS according to OECD TG 443 with the Substance.

8 ECHA requested your considerations for alternative methods to fulfil the information requirement for Toxicity to reproduction. You provided your considerations concluding that there were no alternative methods which could be used to adapt the information requirement(s) for which testing is proposed. ECHA has taken these considerations into account.

9 ECHA agrees that an EOGRTS is necessary.

*2.2. Specification of the study design**2.2.1. Species and route selection*

10 You proposed testing in the rat. ECHA agrees with your proposal because the rat is the species preferred by OECD TG 443.

11 You proposed testing by oral route. ECHA agrees with your proposal.

2.2.2. Pre-mating exposure duration

12 The length of the pre-mating exposure period must be ten weeks to cover the full spermatogenesis and folliculogenesis before the mating, allowing meaningful assessment of the effects on fertility.

13 You did not specify the pre-mating exposure duration.

14 A minimum of 2-week pre-mating exposure duration for P0 animals is required because the full spectrum of parameters on sexual function and fertility will be covered in the F1 animals (Guidance on IRs & CSA, Appendix R.7.6-3).

2.2.3. Dose-level setting

15 The aim of the requested test must be to demonstrate whether the classification criteria of the most severe hazard category for sexual function and fertility (Repr. 1B; H360F) and developmental toxicity (Repr. 1B; H360D) under the CLP Regulation apply for the Substance (OECD TG 443, para. 22; OECD GD 151, para. 28; Annex I Section 1.0.1. of REACH and Recital 7, Regulation 2015/282), and whether the Substance meets the criteria for a Substance of very high concern regarding endocrine disruption according to Art.57(f) of REACH as well as supporting the identification of appropriate risk management measures in the chemical safety assessment.

16 To investigate the properties of the Substance for these purposes, the highest dose level must be set on the basis of clear evidence of an adverse effect on sexual function and fertility, but no deaths (i.e., no more than 10% mortality; Section 3.7.2.4.4 of Annex I to

the CLP Regulation) or severe suffering such as persistent pain and distress (OECD GD 19, para. 18) in the P0 animals.

17 In case there are no clear evidence of an adverse effect on sexual function and fertility, the limit dose of at least 1000 mg/kg bw/day or the highest possible dose level not causing severe suffering or deaths in P0 must be used as the highest dose level. A descending sequence of dose levels should be selected to demonstrate any dose-related effect and aiming to establish the lowest dose level as a NOAEL.

18 In summary, unless limited by the physical/chemical nature of the Substance, the highest dose level in P0 animals must be as follows:

- (1) in case of clear evidence of an adverse effect on sexual function and fertility without severe suffering or deaths in P0 animals, the highest dose level in P0 animals must be determined based on such clear evidence, or
- (2) in the absence of such clear evidence, the highest dose level in P0 animals must be set to be the highest possible dose not causing severe suffering or death, or
- (3) if there is such clear evidence but the highest dose level set on that basis would cause severe suffering or death, the highest dose level in P0 animals must be set to be the highest possible dose not causing severe suffering or death, or
- (4) the highest dose level in P0 animals must follow the limit dose concept.

19 You have to provide a justification with your study results demonstrating that the dose level selection meets the conditions described above.

20 Numerical results (i.e. incidences and magnitudes) and description of the severity of effects at all dose levels from the dose range-finding study/ies must be reported to facilitate the assessment of the dose level section and interpretation of the results of the main study.

2.2.4. Cohorts 1A and 1B

21 Cohorts 1A and 1B belong to the basic study design and must be included.

Splenic lymphocyte subpopulation analysis

23 Splenic lymphocyte subpopulation analysis must be conducted in Cohort 1A (OECD TG 443, para. 66; OECD GD 151, Annex Table 1.3).

Investigations of sexual maturation

25 To improve the ability to detect rare or low-incidence effects, all F1 animals must be maintained until sexual maturation to ensure that sufficient animals (3/sex/litter/dose) are available for evaluation of balano-preputial separation or vaginal patency (OECD GD 151, para. 12 in conjunction with OECD TG 443, para. 47). For statistical analyses, data on sexual maturation from all evaluated animals/sex/dose must be combined to maximise the statistical power of the study.

2.2.5. Extension of Cohort 1B

26 If the Column 2 conditions of 8.7.3. are met, Cohort 1B must be extended by mating the Cohort 1B animals to produce the F2 generation.

27 The extension is required, among others, if the use of the Substance is leading to significant exposure of consumers and professionals (column 2, first para., point (a) of Section 8.7.3.) and if there are indications of one or more relevant modes of action related to endocrine disruption from available in vivo studies or non-animal approaches (column 2, first para., point (b), third indent of Section 8.7.3.).

28 The use of the Substance reported in the joint submission is leading to significant exposure of consumers and professionals because the Substance is used by professionals as cleaning

agents (indoor-outdoor), agrochemicals and in coatings, paints, thinners and paint removers (e.g. PROCs 10,11,13) and by consumers as in coatings, paints, thinners and paint removers and in cleaners.

29 Furthermore, there are indications of one or more modes of action related to endocrine disruption because changes in organs and parameters sensitive to endocrine activity are observed.

30 More specifically, in the OECD TG 421 study (2021) performed with the Substance, shorter mean anogenital distances (absolute and normalized for body weight) were noted for male pups in the 325 and 500 mg/kg/day groups, when compared to the concurrent control group. The differences at 500 mg/kg/day were statistically significant and the absolute values in both groups (3.19 and 2.90 mm, respectively) and were below the minimum mean value (3.4 mm) of the historical control data. The observed changes in the anogenital distance were test-item related.

31 Some effects were also reported in the adrenal glands. Higher mean adrenal gland weights are reported in the OECD TG 421 study at 325 and 500 mg/kg/day group F0 females and are correlated microscopically with minimal diffuse cortical hypertrophy. In the OECD TG 408 study (2014), vacuolation in the adrenal cortex (mainly the zona fasciculata) was present in males at an increased incidence and severity at 100 mg/kg bw/day and above.

32 You have proposed not to include an extension of Cohort 1B.

33 For the reasons stated above, i.e. significant exposure of consumers and professionals, and indications of endocrine modes of action, ECHA considers that Cohort 1B must be extended.

34 Organs and tissues of Cohort 1B animals processed to block stage, including those of identified target organs, must be subjected to histopathological investigations (according to OECD TG 443, para. 67 and 72) because there is a concern for reproductive toxicity/endocrine activity indicated by the toxicity-triggers to extend the Cohort 1B.

35 The F2 generation must be followed to weaning allowing assessment of nursing and lactation of the F1 parents and postnatal development of F2 offspring. Investigations for F2 pups must be similar to those requested for F1 pups in OECD TG 443 and described in OECD GD 151.

2.2.6. Cohort 3

36 The developmental immunotoxicity Cohort 3 needs to be conducted in case of a particular concern on (developmental) immunotoxicity.

37 Existing information on the Substance itself derived from the available OECD TG 408 study (2014) shows evidence of toxicity to the thymus. You have not provided tabular data but you reported that thymus atrophy is increased at a minimal level in males and females receiving 500 mg/kg bw/day, and also in males receiving 100 mg/kg bw/day. According to Guidance on IRs & CSA, Appendix R.7.6-2, thymus atrophy may indicate a particular concern justifying the inclusion of Cohort 3.

38 You have proposed not to include Cohort 3.

39 For the reasons stated above, the developmental immunotoxicity Cohort 3 must be conducted.

2.3. Outcome

40 Under Article 40(3)(b) your testing proposal is accepted under modified conditions, and you are requested to conduct the test with the Substance, as specified above.

2.3.1. Further expansion of the study design

- 41 No triggers for the inclusion of Cohorts 2A and 2B (developmental neurotoxicity) were identified. However, you may expand the study by including Cohorts 2A and 2B if relevant information becomes available from other studies or during conduct of this study. Inclusion is justified if the available information meets the criteria and conditions which are described in Column 2, Section 8.7.3., Annex IX/X. You may also expand the study due to other scientific reasons in order to avoid a conduct of a new study. The study design, including any added expansions, must be fully justified and documented. Further detailed guidance on study design and triggers is provided in Guidance on IRs & CSA, Section R.7.6.

References

The following documents may have been cited in the decision.

Guidance on information requirements and chemical safety assessment (Guidance on IRs & CSA)

- Chapter R.4 Evaluation of available information; ECHA (2011).
- Chapter R.6 QSARs, read-across and grouping; ECHA (2008).
Appendix to Chapter R.6 for nanoforms; ECHA (2019).
- Chapter R.7a Endpoint specific guidance, Sections R.7.1 – R.7.7; ECHA (2017).
Appendix to Chapter R.7a for nanomaterials; ECHA (2017).
- Chapter R.7b Endpoint specific guidance, Sections R.7.8 – R.7.9; ECHA (2017).
Appendix to Chapter R.7b for nanomaterials; ECHA (2017).
- Chapter R.7c Endpoint specific guidance, Sections R.7.10 – R.7.13; ECHA (2017).
Appendix to Chapter R.7a for nanomaterials; ECHA (2017).
Appendix R.7.13-2 Environmental risk assessment for metals and metal compounds; ECHA (2008).
- Chapter R.11 PBT/vPvB assessment; ECHA (2017).
- Chapter R.16 Environmental exposure assessment; ECHA (2016).

Guidance on data-sharing; ECHA (2017).

Guidance for monomers and polymers; ECHA (2012).

Guidance on intermediates; ECHA (2010).

All guidance documents are available online: <https://echa.europa.eu/guidance-documents/guidance-on-reach>

Read-across assessment framework (RAAF)

- RAAF, 2017 Read-across assessment framework (RAAF); ECHA (2017)
- RAAF UVCB, 2017 Read-across assessment framework (RAAF) – considerations on multi- constituent substances and UVCBs); ECHA (2017).

The RAAF and related documents are available online:

<https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across>

OECD Guidance documents (OECD GDs)

- OECD GD 23 Guidance document on aquatic toxicity testing of difficult substances and mixtures; No. 23 in the OECD series on testing and assessment, OECD (2019).
- OECD GD 29 Guidance document on transformation/dissolution of metals and metal compounds in aqueous media; No. 29 in the OECD series on testing and assessment, OECD (2002).
- OECD GD 150 Revised guidance document 150 on standardised test guidelines for evaluating chemicals for endocrine disruption; No. 150 in the OECD series on testing and assessment, OECD (2018).
- OECD GD 151 Guidance document supporting OECD test guideline 443 on the extended one-generation reproductive toxicity test; No. 151 in the OECD series on testing and assessment, OECD (2013).

Appendix 2: Procedure

ECHA started the testing proposal evaluation in accordance with Article 40(1) on 5 November 2021.

ECHA held a third party consultation for the testing proposal(s) from 25 January 2022 until 20 March 2022. ECHA did not receive information from third parties.

ECHA followed the procedure detailed in Articles 50 and 51 of REACH.

ECHA notified you of the draft decision and invited you to provide comments.

In your comments, you requested an extension of deadline. The deadline of the draft decision was set based on standard practice for carrying out OECD TG tests. It has been exceptionally extended by 12 months from the standard deadline granted by ECHA to take into account currently longer lead times in contract research organisations.

ECHA notified the draft decision to the competent authorities of the Member States for proposals for amendment.

As no amendments were proposed, ECHA took the decision according to Article 51(3) of the REACH Regulation.

Appendix 3: Addressee of this decision and their corresponding information requirements

In accordance with Articles 10(a) and 12(1) of REACH, the information requirements for individual registrations are defined as follows:

- the information specified in Annexes VII to X to REACH, for registration at more than 1000 tpa.

Registrant Name	Registration number	Highest REACH Annex applicable to you
██████████	██████████	██████

Where applicable, the name of a third party representative (TPR) may be displayed in the list of recipients whereas ECHA will send the decision to the actual registrant.

Appendix 4: Conducting and reporting new tests for REACH purposes

1. Requirements when conducting and reporting new tests for REACH purposes

1.1. Test methods, GLP requirements and reporting

- (1) Under Article 13(3) of REACH, all new data generated as a result of this decision must be conducted according to the test methods laid down in a European Commission Regulation or to international test methods recognised by the Commission or ECHA as being appropriate.
- (2) Under Article 13(4) of REACH, ecotoxicological and toxicological tests and analyses must be carried out according to the GLP principles (Directive 2004/10/EC) or other international standards recognised by the Commission or ECHA.
- (3) Under Article 10(a)(vi) and (vii) of REACH, all new data generated as a result of this decision must be reported as study summaries, or as robust study summaries, if required under Annex I of REACH. See ECHA Practical Guide on How to report robust study summaries².

1.2. Test material

- (1) Selection of the Test material(s)
The Test Material used to generate the new data must be selected taking into account the following:
 - the impact of each constituent/ impurity on the test results for the endpoint to be assessed. For example, if a constituent/ impurity of the Substance is known to have an impact on (eco)toxicity, the selected Test Material must contain that constituent/ impurity.
- (2) Information on the Test Material needed in the updated dossier
 - You must report the composition of the Test Material selected for each study, under the "Test material information" section, for each respective endpoint study record in IUCLID.
 - The reported composition must include all constituents of each Test Material and their concentration values and other parameters relevant for the property to be tested.

This information is needed to assess whether the Test Material is relevant for the Substance.

Technical instructions on how to report the above is available in the manual on How to prepare registration and PPORD dossiers³.

2. General recommendations for conducting and reporting new tests

References to Guidance on REACH and other supporting documents can be found in Appendix 1.

² <https://echa.europa.eu/practical-guides>

³ <https://echa.europa.eu/manuals>