

Helsinki, 14 July 2022

Addressee

Registrant of JS_Perfluorotripropylamine as listed in Appendix 3 of this decision

Date of submission of the dossier subject to this decision

09/10/2020

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

Substance name: Perfluamine

EC number: 206-420-2

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)**

Based on Article 40 of Regulation (EC) No 1907/2006 (REACH), you must submit the information listed below by **21 January 2026**.

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

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

1. In vivo mammalian alkaline comet assay (test method: OECD TG 489) combined with in vivo mammalian erythrocyte micronucleus test (test method: OECD TG 474) in rats, or if justified, in mice, oral route (triggered by Annex I, Section 0.5 in conjunction with Annex IX, Section 8.4., column 2). For the comet assay the following tissues shall be analysed: liver, glandular stomach and duodenum.

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:
 - Ten 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) and
 - Cohort 1B (Reproductive toxicity) without extension to mate the Cohort 1B animals to produce the F2 generation.
 - You must report the study performed according to the above specifications. Any expansion of the study must be scientifically justified.
3. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.; test method: OECD TG 414) by oral route, in a second species (rabbit)

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 addressees of the decision and their corresponding information requirements based on registered tonnage band are listed in Appendix 3.

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

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Reasons for the decision(s) related to the information under Annex IX of REACH**1. In vivo mammalian alkaline comet assay combined with in vivo mammalian erythrocyte micronucleus test**

1 An appropriate in vivo somatic cell genotoxicity is an information requirement under Annex IX to REACH (Section 8.4., Column 2) if (1) there is a positive result in any of the in vitro genotoxicity study under Annex VII or VIII to REACH and (2) there are no results available from an in vivo study.

1.1. Information provided in the dossier

2 In relation to condition (1), your dossier does not contain any in vitro genotoxicity studies, required under REACH Annexes VII and VIII, Section 8.4. In the data waiver justification, you explain that “[i]n vitro studies cannot be conducted with this test article as the substance is not soluble in any solvent compatible with cell culture media”. In support of your statement, you provide a study report describing the results of the solubility tests conducted with the Substance in various solvents suitable for in vitro genotoxicity tests.

3 Moreover, in relation to condition (2), no data from an in vivo somatic cell genotoxicity study is available in the dossier.

4 Instead, you have submitted two separate testing proposals, for an in vivo mammalian alkaline comet assay and an in vivo mammalian erythrocyte micronucleus test, to be performed with the Substance.

5 You conclude that the in vivo studies are necessary to fulfil the genetic toxicity information requirements.

6 ECHA requested your considerations for alternative methods to fulfil the information requirement for genetic toxicity in vivo. 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.

7 ECHA considers that you have adequately demonstrated the need to perform further testing, to better address the lack of data for the endpoint of mutagenicity.

8 ECHA therefore agrees that further information is necessary for producing the chemical safety report and that this information can only be obtained by performing an in vivo mutagenicity study in accordance with Annex IX to REACH (Section 0.5, Annex I to REACH), to address gene mutation and chromosomal aberration.

1.2. Test selection

9 The proposed tests are appropriate to investigate either gene mutations or chromosomal aberrations in vivo. However, ECHA notes that the in vivo mammalian erythrocyte micronucleus test (“MN test”, OECD TG 474) and the in vivo mammalian alkaline comet assay (“comet assay”, OECD TG 489) can be combined in a single study (see OECD TG 474 para. 37c; OECD TG 489 para. 33; Guidance on IRs & CSA, Section R.7.7.6.3). While the MN test can detect both structural chromosomal aberrations (clastogenicity) and numerical chromosomal aberrations (aneuploidy), the comet assay can detect primary DNA damage that may lead to gene mutations and/or structural chromosomal aberrations. A combined study will thus address both chromosomal aberration as well as gene mutation.

10 Furthermore, the combined study can help reduce the number of tests performed and the number of animals used while addressing (structural and numerical) chromosomal aberrations as well as gene mutations.

11 Therefore, the comet assay combined with the MN test is the most appropriate study for the Substance.

1.3. Specification of the study design

12 You did not specify the species to be used for testing. According to the test method OECD TG 489, rats are the preferred species. Other rodent species can be used if scientifically justified. According to the test method OECD TG 474, the test may be performed in mice or rats. Therefore, the combined study must be performed in rats, or if justified, in mice.

13 You did not specify the route for testing. Having considered the anticipated routes of human exposure and adequate exposure of the target tissue(s) performance of the test by the oral route is appropriate.

14 In line with the test method OECD TG 489, the test must be performed by analysing tissues from liver as primary site of xenobiotic metabolism, glandular stomach and duodenum as sites of contact. There are several expected or possible variables between the glandular stomach and the duodenum (different tissue structure and function, different pH conditions, variable physico-chemical properties and fate of the Substance, and probable different local absorption rates of the Substance and its possible breakdown product(s)). In light of these expected or possible variables, it is necessary to analyse both tissues to ensure a sufficient evaluation of the potential for genotoxicity at the site of contact in the gastro-intestinal tract.

15 The combination of OECD TGs 489 and 474 should not impair the validity of and the results from each individual study. Careful consideration should be given to the dosing, and tissue sampling for the comet analysis alongside the requirements of tissue sampling for the mammalian erythrocyte micronucleus test (see OECD TG 489, e.g. Bowen et al. 2011 [1]).

1.3.1. Germ cells

16 A subsequent germ cell genotoxicity study (TGR/OECD TG 488, or CA on spermatogonia/OECD TG 483) may still be required under Annex IX of REACH, in case 1) an in vivo genotoxicity test on somatic cell is positive, and 2) no clear conclusion can be made on germ cell mutagenicity.

17 Therefore, you may consider collecting the male gonadal cells from the seminiferous tubules in addition to the other aforementioned tissues in the comet assay, as it would optimise the use of animals. You can prepare the slides for male gonadal cells and store them for up to 2 months, at room temperature, in dry conditions and protected from light. Following the generation and analysis of data on somatic cells in the comet assay, in accordance with Annex IX, Section 8.4., column 2, you should consider analysing the slides prepared with gonadal cells. This type of evidence may be relevant for the overall assessment of possible germ cell mutagenicity including classification and labelling according to the CLP Regulation.

[1] Bowen D.E. et al. 2011. Evaluation of a multi-endpoint assay in rats, combining the bone-marrow micronucleus test, the comet assay and the flow-cytometric peripheral blood micronucleus test. *Mutation Research* 722 7–19.

1.4. Outcome

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

19 In the comments to the draft decision, you agree to perform the requested study.

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

20 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

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

22 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.

23 ECHA agrees that an EOGRTS is necessary.

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

24 You did not specify the species to be used for testing for testing. According to the test method OECD TG 443, the rat is the preferred species. Therefore, the study must be conducted in the rat.

25 You proposed testing by oral route in rats. ECHA agrees with your proposal.

2.2.2. Pre-mating exposure duration

26 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.

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

28 Ten weeks pre-mating exposure duration is required to obtain results adequate for classification and labelling and/or risk assessment. There is no substance specific information in the dossier supporting shorter pre-mating exposure duration (Guidance on IRs & CSA, Appendix R.7.6-3).

29 In addition, the substance is lipophilic (log Kow range of 5.3-6.1 (read across)). Therefore, ten weeks pre-mating is required to ensure that a steady state is reached in the parental animals before mating.

30 In the comments to the draft decision, you agree to having a ten-week pre-mating exposure duration.

2.2.3. Dose-level setting

31 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.

- 32 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.
- 33 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.
- 34 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) (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.
- 35 You must provide a justification with your study results demonstrating that the dose level selection meets the conditions described above.
- 36 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.
- 37 In your comments to the draft decision, you propose the use of 1000 mg/kg/day (the limit dose) as the highest dose level in the P0 animals based upon NOAELs of ≥ 1000 mg/kg bw/day in the available OECD TG 408, 414 and 421 studies conducted in rats with the Substance.
- 38 Based on the information provided, ECHA agrees that the highest dose level in P0 animals in the OECD TG 443 study should follow the limit dose concept (point 4 above).

2.2.4. Cohorts 1A and 1B

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

40 Histopathological investigations in Cohorts 1A and 1B

- 41 In addition to histopathological investigations of cohorts 1A, 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) if

- the results from Cohort 1A are equivocal,
- the test substance is a suspected reproductive toxicant or

- the test substance is a suspected endocrine toxicant.

42 Splenic lymphocyte subpopulation analysis

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

44 Investigations of sexual maturation

45 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.

46 In the comments to the draft decision, you agree to including Cohort 1A and 1B, without extension to mate the Cohort 1B.

2.3. Outcome

47 Your testing proposal is accepted under Article 40(3)(a) and you are requested to conduct the test with the Substance, as specified above.

2.4. Further expansion of the study design

48 The conditions to include the extension of Cohort 1B are currently not met. Furthermore, no triggers for the inclusion of Cohorts 2A and 2B (developmental neurotoxicity) and Cohort 3 (developmental immunotoxicity) were identified. However, you may expand the study by including the extension of Cohort 1B, Cohorts 2A and 2B and/or Cohort 3 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.

3. Pre-natal developmental toxicity study

49 A pre-natal developmental toxicity (PNDT) study (OECD TG 414) in two species is a standard information requirement under Annex X, Section 8.7.2. to REACH.

3.1. Information provided to fulfil the information requirement

50 You have submitted a testing proposal for a PNDT study in a second species according to OECD TG 414 with the Substance.

51 ECHA requested your considerations for alternative methods to fulfil the information requirement for Developmental toxicity. 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.

52 ECHA agrees that a PNDT study in a second species is necessary.

3.2. Specification of the study design

- 53 You proposed testing in the rabbit as a second species. The study in the first species was conducted in the rat. The rat or the rabbit are the preferred species under the OECD TG 414 (ECHA Guidance R.7a, Section R.7.6.2.3.2.). Therefore, the study must be conducted in the rabbit.
- 54 You proposed testing by the oral route. ECHA agrees with your proposal because this route of administration is the most appropriate to investigate reproductive toxicity (ECHA Guidance R.7a, Section R.7.6.2.3.2.).

3.3. Outcome

- 55 Your testing proposal is accepted under Article 40(3)(a) and you are requested to conduct the test with the Substance, as specified above.
- 56 In the comments to the draft decision, you agree to perform the requested study.

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).

All Guidance on REACH is 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

For this Substance the process of substance evaluation started in 2020.

ECHA started the testing proposal evaluation in accordance with Article 40(1) on 11 March 2019.

ECHA held a third-party consultation for the testing proposals from 21 December 2021 until 4 February 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.

ECHA took into account your comments and did not amend the requests.

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

As no amendments were proposed, ECHA adopted the decision under Article 51(3) of REACH.

The deadline of the decision is 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.

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
[REDACTED]	[REDACTED]	[REDACTED]

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 boundary composition(s) of the Substance,
 - 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 <https://echa.europa.eu/practical-guides>

³ <https://echa.europa.eu/manuals>