EUROPEAN CHEMICALS AGENCY

Helsinki, 08 December 2020

**Addressees**
Registrants of strontium sulfide ec 215-249-2 listed in the last Appendix of this decision

**Date of submission for the jointly submitted dossier subject of this decision**
12/05/2016

**Registered substance subject to this decision, hereafter 'the Substance'**
Substance name: Strontium sulphide
EC number: 215-249-2
CAS number: 1314-96-1

**Decision number:** [Please refer to the REACH-IT message which delivered this communication (in format CCH-D-XXXXXXXXXX-XX-XX/F)]

## DECISION ON A COMPLIANCE CHECK

Based on Article 41 of Regulation (EC) No 1907/2006 (REACH), ECHA requests that you submit the information listed below by the deadline of ***15 June 2023***.

Requested information must be generated using the Substance unless otherwise specified

### A. Requirements applicable to all the Registrants subject to Annex VIII of REACH

1. In vitro cytogenicity study in mammalian cells (Annex VIII, Section 8.4.2., test method OECD TG 473) or in vitro micronucleus study (Annex VIII, Section 8.4.2., test method OECD TG 487);

### B. Requirements applicable to all the Registrants subject to Annex IX of REACH

1. Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.; test method OECD TG 414) in a first species (rat or rabbit), oral route;

2. Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5.; test method EU C.20./OECD TG 211);

### C. Requirements applicable to all the Registrants subject to Annex X of REACH

1. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.; test method OECD TG 414) in a second species (rat or rabbit), oral route.

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

   - Ten weeks premating exposure duration for the parental (P0) generation;
   - Dose level setting shall aim to induce systemic toxicity at the highest dose level;
   - Cohort 1A (Reproductive toxicity);
   - Cohort 1B (Reproductive toxicity) without extension to mate the Cohort 1B animals to produce the F2 generation

- Cohorts 2A and 2B (Developmental neurotoxicity)

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

Reasons for the request(s) are explained in the following appendices:
- Appendix entitled "Reasons common to several requests";
- Appendices entitled "Reasons to request information required under Annexes VII to X of REACH", respectively.

**Information required depends on your tonnage band**

You must provide the information listed above for all REACH Annexes applicable to you, and in accordance with Articles 10(a) and 12(1) of REACH:
- the information specified in Annexes VII to X to REACH, for registration at more than 1000 tpa.

**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 testing and reporting requirements provided under the Appendix entitled "Requirements to fulfil when conducting and reporting new tests for REACH purposes". In addition, you should follow the general recommendations provided under the Appendix entitled "General recommendations when conducting and reporting new tests for REACH purposes". For references used in this decision, please consult the Appendix entitled "List of references".

**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.

Approved[1] under the authority of Christel Schilliger-Musset, Director of Hazard Assessment

[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 on Reasons common to several requests**

## (i) Assessment of your read-across approach, under Annex XI, Section 1.5.

You seek to adapt the following standard information requirements by applying read-across approaches in accordance with Annex XI, Section 1.5:

- In vitro cytogenicity study in mammalian cells or in vitro micronucleus study (Annex VIII, Section 8.4.2.)
- Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.)
- Pre-natal developmental toxicity study in a second species (Annex X, Section 8.7.2.)
- Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.)

ECHA has considered the scientific and regulatory validity of your read-across approach(es) in general before assessing the specific standard information requirements in the following appendices.

### Grouping of substances and read-across approach

Annex XI, Section 1.5. specifies two conditions which must be fulfilled whenever a read-across approach is used. Firstly, there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category. Secondly, it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (addressed under 'Assessment of prediction(s)').

Additional information on what is necessary when justifying a read-across approach can be found in the ECHA Guidance[2] and related documents[3, 4].

#### Predictions for toxicological properties

You have provided a read-across justification document in the CSR.

You read-across between the substances

- Sulfide substances [1]+[2]
  - **[1]** Dihydrogensulfide, $H_2S$, EC No. 231-977-3 (CAS No. 7783-06-4)
  - **[2]** Disodium sulfide, $Na_2S$, EC No. 215-211-5 (CAS No. 1313-82-2)
- Strontium substances [3]+[4]
  - **[3]** Strontium dinitrate, $Sr(NO_3)_2$, EC No. 233-131-9 (CAS No. 10042-76-9)
  - **[4]** [redacted].(CAS No. [redacted])

as source substances and the Substance as target substance.

You have provided the following reasoning for the prediction of toxicological properties:

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[2] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals. 2008 (May) ECHA, Helsinki. 134. pp. Available online: https://echa.europa.eu/documents/10162/13632/information_requirements_r6_en.pdf/77f49f81-b76d-40ab-8513-4f3a533b6ac9

[3] Read-Across Assessment Framework (RAAF). 2017 (March) ECHA, Helsinki. 60 pp. Available online: Read-Across Assessment Framework (https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across)

[4] Read-across assessment framework (RAAF) - considerations on multi-constituent substances and UVCBs. 2017 (March) ECHA, Helsinki. 40 pp. Available online: https://doi.org/10.2823/794394

ECHA understands that you base your read-across on the dissociation of the strontium and sulphide and that information on both would be sufficient to conclude on the toxicity posed by the Substance.

"***READ-across to $H_2S$ [1] and $Na_2S$ [2]:***
*[...]"In conclusion, under physiological conditions, inorganic sulfides or hydrogensulfides as well as H2S will dissociate to the respective species relevant to the pH of the physiological medium, irrespective of the nature of the "sulfide", which is why read-across between these substances and H2S is considered to be appropriate without any restrictions for the purpose of hazard and risk assessment of strontium sulfide."*

***Read-across to $Sr(NO_3)_2$ [3] and [redacted] [4]:***
*[...]"In conclusion, read across from strontium dinitrate and [redacted] to strontium sulfide is considered as justified since the toxicity of these substances may reasonably be considered to be determined by the availability of Sr cations. It is noted that although SrS is a strong base (pH 12.6 for a 1% solution - source: Anonymous, 2009), substantial neutralisation in the gastrointestinal tract at pH-levels of approx.1.5 – 2 may nevertheless be anticipated."*

ECHA understands that you predict the properties of the Substance using a read-across hypothesis which is based on the formation of common (bio)transformation products. The properties of your Substance are predicted to be quantitatively equal to those of the source substance.

ECHA notes the following deficiencies with regard to the applied read-across for the physico-chemical and (eco)toxicological properties:

*a) Adequacy and reliability of source studies*

According to Annex XI, Section 1.5., if the grouping concept is applied then in all cases the results to be read across should:
- have adequate and reliable coverage of the key parameters addressed in the corresponding test method referred to in Article 13(3);
- cover an exposure duration comparable to or longer than the corresponding test method referred to in Article 13(3) if exposure duration is a relevant parameter.

The studies that you provided for the endpoints on *in vitro* cytogenicity in mammalian cells or *in vitro* micronucleus study for the sulphide ion, developmental toxicity in two species for the sulphide ion, and reproductive toxicity for both the sulphide and strontium ions, do not provide an adequate coverage of some key parameters expected to be investigated and do not meet the requirement for adequacy and reliability under Section 1.5, Annex XI to REACH for the reasons provided under Appendix A, section 1, Appendix B, section 1 and Appendix C, sections 1 and 2.

*b) Supporting information*

Information on the impact of non-common compounds

Annex XI, Section 1.5 of the REACH Regulation states that "*physicochemical properties, human health effects and environmental effects or environmental fate may be predicted from data for reference substance(s)*". For this purpose "*it is important to provide supporting information to strengthen the rationale for the read-across*"[5]. The set of supporting

[5] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.2.1.f

information should allow to verify the crucial aspects of the read-across hypothesis and establish that the properties of the Substance can be predicted from the data on the source substance(s).

As indicated above, your read-across hypothesis is based on the (bio)transformation of the Substance and of the source substance(s) to common compound(s). In this context, the impact of exposure to these compounds on the prediction of properties of the target needs to be assessed to ensure that a reliable prediction can be made.

In your comments to the draft decision you state that "*exposure to the sulphide ion is only critical for acute effects and that the strontium ion will drive the (reproductive) toxicity of your Substance. This is based on an assessment on the bioavailability and modes of actions of sulphide in the human body, concluding that hydrogen sulphide is a widely available endogenous compound providing various routes of enzymatic formation as well as various modes to regulate the background levels through oxidation to sulphate. The assessment is supported by recent scientific publications on this subject matter. Therefore the sulphide anion does not contribute to the systemic toxicity effects of strontium sulphide.*"

ECHA has assessed the information given in your comments and notes the following:

*Comments related to systemic toxicity and reprotoxicity*

Hydrogen sulfide has been subject to an evaluation by the U.S. Environmental Protection Agency (U.S. EPA, 2003). In their Integrated Risk Information System (IRIS) report EPA summarised findings relevant for reference concentration setting as follows[6]:

"Both nasal tract lesions and neurologic effects occur in animals exposed to the same concentration range of H2S. Skrajny et al. (1992)[7] reported altered ($p<0.01$) levels of several neurotransmitters on postpartum days 7-21 in the brains of rat pups exposed to 105 mg/m3 (75 ppm) H2S. Norepinephrine levels of the frontal cortex were decreased compared to controls on days 14 and 21 at 28 mg/m3 (20 ppm). Serotonin levels in the frontal cortex were significantly increased on day 21 postpartum in rat pups exposed to 28 mg/m3 (20 ppm) H2S. Hannah and Roth (1991)[8] reported changes that could be an indicator of a neurotoxiceffect in the brains of rat pups that had been exposed in utero during development until PND 21 at 20 or 50 ppm (28 or 42 mg/m3). The observed effect - alteration in dendritic arborization of developing cerebellar Purkinje cells - was judged by the authors to be present at both concentrations such that the 20 ppm (28 mg/m3) level may be considered a low-effect rather than a no-effect level."

Based on the summary by U.S. EPA referred to above, systemic or reproductive/developmental effects following exposure to the sulphide ion cannot be excluded as neurologic effects have been reported in rat pups exposed in utero. Therefore your statements that "*exposure to the sulphide ion is only critical for acute effects*", "*the sulphide anion does not contribute to the systemic toxicity effects of strontium sulphide*" and "*further reproductive toxicity studies with sulfide will not provide any useful information for the risk assessment of strontium sulfide*" are not supported.

ECHA further notes that the developing brain is particularly vulnerable to oxidants such as H2S as it lacks antioxidant defences comparable to that of adult animals. Increased metabolic activity required for growth makes the developing brain sensitive to oxidative stress (Gupta

[6] P. 7, IRIS Summary https://cfpub.epa.gov/ncea/iris2/chemicalLanding.cfm?substance_nmbr=61
[7] Skrajny et al., 1992, Can J Physiol Pharmacol, 70: 1515-1518, included in Section 7.9.1. in IUCLID
[8] Hannah & Roth (1991) Neurosci. Lett. 122: 225-228

2011)[9].

*Conclusion*

You have not established that a reliable prediction of the property under consideration of the Substance can be derived on the basis of your read-across hypothesis as further elaborated in your comments, i.e. that systemic exposure to the sulphide ion would not be relevant, and that the toxicity of your Substance may be determined by the strontium ion. Therefore you have not provided supporting information to strengthen the rationale for the read-across.

**Conclusions on the grouping of substances and read-across approach**

As explained above, you have not established – neither in your dossier nor in your comments - that relevant properties of the Substance can be predicted from data on the analogue substance. Therefore, your adaptation does not comply with the general rules of adaptation as set out in Annex XI, Section 1.5. and your grouping and read-across approach is rejected.

[9] Gupta RC (Ed) (2011) Reproductive and Developmental Toxicology. Elsevier Inc Academic Press, Amsterdam, The Netherlands.

## Appendix A: Reasons to request information required under Annex VIII of REACH

### 1. In vitro cytogenicity study in mammalian cells or in vitro micronucleus study (Annex VIII, Section 8.4.2.)

An *in vitro* cytogenicity study in mammalian cells or an *in vitro* micronucleus study is a standard information requirement in Annex VIII to REACH.

You have adapted this information requirement by using the following key studies in your Grouping of substances and read-across approach under Annex XI, Section 1.5:

a) *in vitro* micronucleus study equivalent to OECD TG 487 (2010) with the analogue substance strontium dinitrate (EC No. 233-13-9) [3]
b) *in vivo* micronucleus study similar to OECD TG 474 (1981) with the analogue substance disodium sulfide (EC No. 215-211-5) [2]

We have assessed this information and identified the following issues with the information provided for the sulphide ion:

For the reasons explained in the Appendix on Reasons common to several requests, the *in vivo* cytogenicity study used for adaptation must adequately and reliably cover the key parameters foreseen to be investigated in the OECD GD 474. These key parameters include the following:

- Each group must have a minimum of 5 analysable animals of one sex, or of each sex if both are used.
- The highest dose studied must be the maximum tolerated dose (MTD), i.e. the highest dose that is tolerated without evidence of toxicity (e.g. body weight depression or hematopoietic system cytotoxicity, but not death or evidence of pain, suffering or distress necessitating humane euthanasia). The highest dose can also be a dose that produces toxicity in the bone marrow (e.g. a reduction in the proportion of immature erythrocytes among total erythrocytes in the bone marrow or peripheral blood).
- the proportion of immature erythrocytes among total erythrocytes and the mean number of micronucleated immature erythrocytes must be reported for each group of animals.

The reported data for the *in vivo* study you submitted did not include:

- The analysis of the adequate number of animals: only 2 animals per sex were used in each group.
- A maximum studied dose that is an MTD or induces toxicity in the bone marrow.
- The reporting of the proportion of immature erythrocytes among total erythrocytes and the mean number of micronucleated immature erythrocytes for each group of animals.

Therefore, the *in vivo* test provided for the sulphide ion does not cover the key parameters of OECD TG 474. Your adaptation is rejected and a study is requested.

**Appendix B: Reasons to request information required under Annex IX of REACH**

1. **Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.) in a first species**

A Pre-natal developmental toxicity (PNDT) study (OECD TG 414) in one species is a standard information requirement under Annex IX to REACH.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. by providing the following studies:

a) screening for reproductive toxicity study performed with $H_2S$ **[1],** according to OECD TG 421 (2000), inhalation route;
b) Growth and development in the rat during subchronic exposure to low levels of hydrogen sulphide, $H_2S$ **[1]**, **(**1990) inhalation route;
c) 1990, developmental toxicity study on glucose levels in maternal rats GD 6 to day 21 PP) with hydrogen sulphide, $H_2S$ **[1]**, (1990) inhalation route;
d) developmental toxicity study on effects of two monoamine neurotransmitter under exposure to hydrogen sulphide, $H_2S$ **[1]**, (1992) inhalation route.

We have assessed this information and identified the following issue(s):

For the reasons explained in the Appendix on Reasons common to several requests, a pre-natal developmental toxicity study used for adaptation must adequately and reliably cover the key parameters of OECD TG 414. The key parameters of this test guidelines include:

- structural malformations and variations

None of the above listed studies a) to d) for the sulphide ion investigate these key parameters.

Therefore, your adaptation is rejected.

A PNDT study according to the test method OECD TG 414 must be performed in rat or rabbit as preferred species with oral[10] administration of the Substance.

ECHA notes that in your comment to the draft decision you regard testing of your Substance as challenging due to its corrosivity. For that reason every effort must be taken to ensure administration of the test material in a form that minimises corrosion. Further information about testing of corrosive substances can be found in ECHA Guidance R. 7a, Section R.7.6.2.3.2. The dose selection used for the study should be justified.

2. **Long-term toxicity testing on aquatic invertebrates (Annex IX, Section 9.1.5.)**

Long-term toxicity testing on aquatic invertebrates is a standard information requirement in Annex IX to the REACH Regulation.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5.

You have provided a read-across justification in CSR.

[10] ECHA Guidance R.7a, Section R.7.6.2.3.2.

You have provided the following reasoning for the prediction of toxicological properties: *"No ecotoxicological data are available for strontium sulfide itself. However, in the aqueous and terrestrial environment, strontium sulfide dissolves in water releasing strontium cations and sulfide anions (see physical and chemical properties)."*

ECHA understands that you predict the properties of the Substance using a read-across hypothesis which is based on the formation of common (bio)transformation products. The properties of your Substance are predicted to be quantitatively equal to those of the source substance.

Annex XI, Section 1.5. specifies two conditions which must be fulfilled whenever a read-across approach is used. Firstly, there needs to be structural similarity between substances which results in a likelihood that the substances have similar physicochemical, toxicological and ecotoxicological properties so that the substances may be considered as a group or category. Secondly, it is required that the relevant properties of a substance within the group may be predicted from data for reference substance(s) within the group (read-across approach).

Additional information on what is necessary when justifying a read-across approach can be found in the ECHA Guidance R.6 and related documents.

ECHA notes the following shortcomings with regards to the predictions of ecotoxicological properties.

Missing supporting information

Annex XI, Section 1.5 of the REACH Regulation states that "*physicochemical properties, human health effects and environmental effects or environmental fate may be predicted from data for reference substance(s)*". For this purpose "*it is important to provide supporting information to strengthen the rationale for the read-across*"[11]. The set of supporting information should allow to verify the crucial aspects of the read-across hypothesis and establish that the properties of the Substance can be predicted from the data on the source substance(s)."

Supporting information must include information to confirm the formation of the common compound.

As indicated above, your read-across hypothesis is based on the (bio)transformation of the Substance and of the source substance(s) to a common compound(s). In this context, information characterising the rate and extent of the hydrolysis of the Substance and of the source substance(s) is necessary to confirm the formation of the proposed common hydrolysis product and to assess the impact of the exposure to the parent compounds.

In the hydrolysis endpoint, you have stated: "*sulfide ($S^{2-}$), bisulfide ($HS^-$) and hydrogen sulfide ($H_2S$) coexist in aqueous solution in a dynamic pH-dependant equilibrium. In oxic systems, oxidation to - eventually - sulfate occurs".*

You have not provided any experimental data or other adequate and reliable information, neither about the rate of hydrolysis, the extent of hydrolysis or the rate and extent of hydrolysis in different environments of your Substance, nor of the source substance.

In the absence of this information, you have not provided supporting evidence establishing that the proposed common hydrolysis product is formed as assumed in your read-across

[11] Guidance on information requirements and chemical safety assessment Chapter R.6: QSARs and grouping of Chemicals, Section R.6.2.2.1.f

hypothesis for the sulphide ion.

In your comments to the draft decision, you include information on the dissociation of the Substance and outline that you will update your dossier with available information to address the long-term aquatic invertebrates for the "sulfide anion". ECHA agrees with this approach. ECHA will assess the information by the set deadline of this decision, in the updated dossier.

Therefore, you have not provided sufficient supporting information to strengthen the rationale for the read-across.

Therefore, the information requirement is not fulfilled.

**Appendix C: Reasons to request information required under Annex X of REACH**

**1. Pre-natal developmental toxicity study (Annex X, Section 8.7.2.) in a second species**

Pre-natal developmental toxicity (PNDT) studies (OECD TG 414) in two species is a standard information requirement under Annex X to REACH.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5.

We have assessed this information and identified the following issues:

For the sulphide ion you have not provided information on a second species. In order to be compliant and enable concluding if the Substance is a developmental toxicant, information provided has to meet the requirements of OECD TG 414 in two species.

Based on the above, the information you provided do not fulfil the information requirement.

*Information on study design*

A PNDT study according to the OECD TG 414 study should be performed in rabbit or rat as the preferred second species, depending on the species tested in the first PNDT study (request B.1 in this decision) with oral[12] administration of the Substance.

ECHA notes that in your comment to the draft decision you regard testing of your Substance as challenging due to its corrosivity. For that reason every effort must be taken to ensure administration of the test material in a form that minimises corrosion. Further information about testing of corrosive substances can be found in ECHA Guidance R. 7a, Section R.7.6.2.3.2. The dose selection used for the study should be justified.

**2. Extended one-generation reproductive toxicity study (Annex X, Section 8.7.3.)**

The basic test design of an Extended one-generation reproductive toxicity (EOGRT) study (OECD TG 443) is a standard information requirement under Annex X to REACH. Furthermore Column 2 of Section 8.7.3. defines when the study design needs to be expanded.

You have adapted this information requirement by using a Grouping of substances and read-across approach under Annex XI, Section 1.5. by providing the following studies:

a) Oral reproduction toxicity study in the Wistar rat, according to the ICH Harmonized Tripartite Guideline, (2001) Addendum: Toxicity to male fertility with the source substance [redacted] **[4]**
b) Fertility and development neurotoxicity effects of inhaled hydrogen sulfide in Sprague-Dawley rats, according to OECD TG 421, (2000) with the source substance dihydrogensulfide **[1].**

In your comments to the draft decision you propose to perform an OECD TG 443 study, if at all, following the basic design as an Annex X standard information requirement. You disagree on the triggers for extension of Cohort 1B to generate F2, and on the DNT Cohort.

We have assessed this information and identified the following issues:

[12] ECHA Guidance R.7a, Section R.7.6.2.3.2.

ECHA
EUROPEAN CHEMICALS AGENCY

## A. Information on strontium

For the reasons explained in the Appendix on Reasons common to several requests, the study used for adaptation must adequately and reliably cover the key parameters of OECD TG 443. The key parameters of this test guidelines include:

- Full histopathology of organs and tissues (P0 and F1)
- Exposure duration comparable to or longer than the corresponding test methods referred to in Article 13(3); including at least exposure of 10 weeks prior to pairing for P0 animals unless the Cohort 1B animals are mated to produce the F2 generation, which is followed to weaning.

In the provided study:

- For reproductive organs only weights were recorded and histopathology was only investigated from F0 males for epididymis and testes.
- F0 males were exposed for 28 days prior to pairing until six days after the treated females started littering. Females were exposed from day six of gestation until day twenty of lactation. The exposure in your study is considerably shorter than as required by OECD TG 443.

Regarding the study on strontium ranelate by ICH you argue in your comments that "*Since this study investigated the effects on both male and female fertility as well as embryo-fetal and pre-/postnatal development, it was used to cover the data requirements on developmental toxicity and toxicity to reproduction. For this reason the information on test design and results has been provided in the IUCLID endpoints for Developmental toxicity (Subgroup A and C) and Toxicity to Reproduction (Subgroup B and D). This might have led to the erroneous conclusion from ECHA that female fertility and pre- and postnatal development were not included in the available study.*"

ECHA has taken the complete study design into consideration. Nevertheless, as also concluded by the Expert statement on reproduction toxicity testing of strontium carbonate by [redacted], the provided ICH reproduction toxicity study does not cover the exposure duration of 10 weeks prior to pairing as required in the standard information requirement requested for this endpoint. Therefore, this study cannot be considered to provide equivalent information as requested by the OECD TG 443.

In the statement by Charles River, the experts agree with ECHA that the study according to ICH guideline ([redacted], 2001) does not fulfil the information requirements as in OECD TG 443: "*As exposure of P0 was limited compared to the exposure design of the EOGRTS and the F1 generation was not directly exposed, it is agreed with ECHA that information on reproduction toxicity is not according to current data requirements.*" Furthermore, the experts agree with ECHA that the exposure period for females is too short in order to investigate female fertility as according to the OECD TG 443. Therefore, the experts conclude that "*an EOGRTS should be performed starting 10 weeks premating – unless F2 is triggered - and with exposure of the F1 generation, which shall be followed to week 13 (Cohort 1A) and week 14 (Cohort 1B) of age and including full histopathological examination of the required organs and tissues.*"

Therefore, the provided study, which specifically investigates male fertility, does neither investigate female fertility nor post-natal developmental toxicity until adhulthood.

Therefore, your adaptation is rejected and the information requirement is not fulfilled.

**B. Sulfide**

For the reasons explained in the Appendix on Reasons common to several requests, the study used for adaptation must adequately and reliably cover the key parameters of OECD TG 443. The key parameters of this test guidelines include:

- highest dose level should aim to induce some systemic toxicity
- examination of relevant life stages
- examination of key parameters for pre/peri/postnatal developmental toxicity

You have provided a a screening for reproductive/developmental toxicity study b) (OECD TG 421) with hydrogen sulphide (EC no 231-977-3) which does not cover all relevant life stages, as the extensive postnatal investigations of the fully exposed F1 generation up to adulthood are not included. Furthermore, the highest dose level in the study did not induce any systemic toxicity and you have not shown that the aim was to induce toxicity. Therefore, the dose level selection was too low, and the study does not fulfil the criterion set in OECD TG 443 and ECHA Guidance R7a.

Therefore, the information provided for the sulphide ion is rejected. Your adaptation is rejected and the information requirement is not fulfilled. Furthermore, as explained in the Appendix on Reasons common to several requests, the additional information given in your comments is not sufficient to fulfil this information requirement.

The specifications for the study design

*Premating exposure duration and dose-level setting*

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

Ten weeks premating 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 premating exposure duration.

Therefore, the requested premating exposure duration is ten weeks.

In order to be compliant and not to be rejected due to too low dose levels, the highest dose level shall aim to induce systemic toxicity, but not death or severe suffering of the animals, to allow comparison of reproductive toxicity and systemic toxicity. The dose level selection should be based upon the fertility effects. A descending sequence of dose levels should be selected in order to demonstrate any dose-related effect and to establish NOAELs.

If there is no relevant data to be used for dose level setting, it is recommended that range-finding results are reported with the main study.

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

*Cohorts 1A and 1B*

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

*Cohorts 2A and 2B*

The developmental neurotoxicity Cohorts 2A and 2B need to be conducted in case of a particular concern on (developmental) neurotoxicity.

Existing information on the analogue substance strontium chloride **[3]** (EC no 233-971-6) derived from a sub-chronic study following a protocol similar or equivalent to OECD TG 408 (Kroes et al., 1977), show evidence of thyroid toxicity in males. Relative thyroid weights were statistically significantly increased at 1200 and 1400 ppm (by 33% ($p>0.01$) and 26% ($p>0.001$), respectively). Thyroid toxicity rises a particular concern on developmental neurotoxicity (ECHA Guidance R.7a).

In your comments to the draft decision to the draft decision you argue that *"As in the repeated dose toxicity study increased thyroid weight was restricted to high dose males only, inclusion of a DNT cohort in the EOGRTS is not scientifically justified. Developmental neurotoxicity may be linked to disturbed thyroid hormone regulation in females during pregnancy, but male thyroid effects have no direct link to endocrine disrupting reproductive effects, i.e. effects on neurodevelopment in the offspring."*

In relation to historical controls you explain that *The applicant has approached the laboratorium that performed the 90-day study for historical control data. However, the laboratorium did not have historical control data for the Wistar rat treated around 1977 anymore. A literature search for historical control data did also not yield relevant information. Therefore, historical control data of the Wistar rat used by* [redacted] *were collected from the old database* [redacted] *going back to 2014 and the new database* [redacted] *(2019)."* Based on this information you conclude that the reported value of the relative thyroid weight of the control group of 0.0054 is quite low compared to all values of the treated groups (and is just outside the P95 limit of the [redacted] historical control data).

You also state that *"...developmental neurotoxicity has been examined in the available reproduction study with strontium ranelate (*[redacted] *2001)... No effects on F1 breeders was seen on any of the developmental neurotoxicity parameters determined."*

ECHA notes that according to ECHA Guidance R.7a, Appendix R.7.6–2 EOGRTS Study Design, the statistically significant higher mean relative thyroid weights in males is relevant and an acceptable trigger for a repeated dose toxicity study. Furthermore, there was a trend in increased mean relative thyroid weights also in treated females compared to controls.

You agreed that thyroid hormones are essential for normal brain development ([redacted] 2018), but *"only effects on female thyroid during pregnancy can influence developmental neurotoxicity of the pups"*. In your comments you refer to the study with strontium ranelate ([redacted] 2001) stating that the developmental neurotoxicity studies partly cover the parameters required for OECD TG 443.

ECHA notes however that the [redacted] 2001 study uses a different exposure period for P0 and F1 animals compared to the OECD TG 443 and therefore the results do not fully cover the study design for this endpoint. Furthermore, it is important to acquire information on similar exposure duration to both sexes, and especially, exposure on females during pregnancy.

ECHA acknowledges that the lack of individual data, standard deviations and historical control data is challenging, but the study does provide reliable comparison with a control group, and this is sufficient to trigger the cohort. Further, ECHA notes that the historical control data should always reflect the same strain, same laboratory, and the same study design/duration, collected from fairly recent studies (± 2 years to the study year). For these reasons, the

historical control data you have referred to [redacted] 2014; [redacted] 2019) does not provide relevant information for the study under consideration.

ECHA concludes that available data indicate that, in the absence of overt general toxicity, the study by Kroes 1977 demonstrate thyroid toxicity. A concern for reproductive (and developmental) toxicity has therefore been identified and a need for further information is triggered. Taken together, ECHA maintains its opinion that the mentioned effects are indicative of mode(s) of action related to endocrine disruption.

Therefore, the developmental neurotoxicity Cohorts 2A and 2B need to be conducted.

Species and route selection

The study must be performed in rats with oral[13] administration.

ECHA notes that in your comment to the draft decision you regard testing of your Substance as challenging due to its corrosivity. For that reason every effort must be taken to ensure administration of the test material in a form that minimises corrosion. Further information about testing of corrosive substances can be found in ECHA Guidance R. 7a, Section R.7.6.2.3.2. The dose selection used for the study should be justified.

*Further expansion of the study design*

The conditions to include the extension of Cohort 1B are currently not met. Furthermore, no triggers for the inclusion of Cohort 3 (developmental immunotoxicity) were identified. However, you may expand the study by including the extension of Cohort 1B and Cohort 3 if relevant information becomes available from other studies or during the 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 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 ECHA Guidance[14].

[13] ECHA Guidance R.7a, Section R.7.6.2.3.2.
[14] ECHA Guidance R.7a, Section R.7.6.

## Appendix D: Requirements to fulfil when conducting and reporting new tests for REACH purposes

### A. 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 summaries[15].

### B. 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 dossiers[16].

[15] https://echa.europa.eu/practical-guides
[16] https://echa.europa.eu/manuals

**Appendix E: Procedural history**

This decision does not prevent ECHA from initiating further compliance checks at a later stage on the registrations present.

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

The compliance check was initiated on 05 June 2019.

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.

## Appendix F: List of references - ECHA Guidance[17] and other supporting documents

Evaluation of available information
Guidance on information requirements and chemical safety assessment, Chapter R.4 (version 1.1., December 2011), referred to as ECHA Guidance R.4 where relevant.

QSARs, read-across and grouping
Guidance on information requirements and chemical safety assessment, Chapter R.6 (version 1.0, May 2008), referred to as ECHA Guidance R.6 where relevant.

Read-across assessment framework (RAAF, March 2017)[18]

RAAF - considerations on multiconstituent substances and UVCBs (RAAF UVCB, March 2017)[18]

Physical-chemical properties
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Toxicology
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

Environmental toxicology and fate
Guidance on information requirements and chemical safety assessment, Chapter R.7a (version 6.0, July 2017), referred to as ECHA Guidance R.7a in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7b (version 4.0, June 2017), referred to as ECHA Guidance R.7b in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.7c (version 3.0, June 2017), referred to as ECHA Guidance R.7c in this decision.

PBT assessment
Guidance on information requirements and chemical safety assessment, Chapter R.11 (version 3.0, June 2017), referred to as ECHA Guidance R.11 in this decision.

Guidance on information requirements and chemical safety assessment, Chapter R.16 (version 3.0, February 2016), referred to as ECHA Guidance R.16 in this decision.

Data sharing
Guidance on data-sharing (version 3.1, January 2017), referred to as ECHA Guidance on data sharing in this decision.

OECD Guidance documents[19]

[17] https://echa.europa.eu/guidance-documents/guidance-on-information-requirements-and-chemical-safety-assessment
[18] https://echa.europa.eu/support/registration/how-to-avoid-unnecessary-testing-on-animals/grouping-of-substances-and-read-across
[19] http://www.oecd.org/chemicalsafety/testing/series-testing-assessment-publications-number.htm

Guidance Document on aqueous-phase aquatic toxicity testing of difficult test chemicals – No 23, referred to as OECD GD 23.

Guidance document on transformation/dissolution of metals and metal compounds in aqueous media – No 29, referred to as OECD GD 29.

Guidance Document on Standardised Test Guidelines for Evaluating Chemicals for Endocrine Disruption – No 150, referred to as OECD GD 150.

Guidance Document supporting OECD test guideline 443 on the extended one-generation reproductive toxicity test – No 151, referred to as OECD GD 151.

**Appendix G: List of the registrants to which the decision is addressed and the corresponding information requirements applicable to them**

| Registrant Name | Registration number | (Highest) Data requirements to be fufilled |
|---|---|---|
| [redacted] | [redacted] | [redacted] |

Note: where applicable, the name of a third party representative (TPR) may be displayed in the list of recipients whereas the decision is sent to the actual registrant.