EUROPEAN CHEMICALS AGENCY

Helsinki, 23 March 2017

Addressee: [redacted]

Decision number: TPE-D-2114355512-54-01/F
Substance name: 1,3-dihydro-4(or 5)-methyl-2H-benzimidazole-2-thione
EC number: 258-904-8
CAS number: 53988-10-6
Registration number: [redacted]
Submission number: [redacted]
Submission date: 06.06.2016
Registered tonnage band: 100-1000T

DECISION ON A TESTING PROPOSAL

Based on Article 40 of Regulation (EC) No 1907/2006 (the 'REACH Regulation'), ECHA examined your testing proposal(s) and decided as follows.

While your originally proposed tests for a Pre-natal developmental toxicity study (EU B.31./OECD TG 414) and an Extended one-generation reproductive toxicity study in rats, (EU B.56./OECD TG 443) using the analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt CAS No 61617-00-3 (EC No 262-872-0) are rejected, you are requested to perform:

1. **Pre-natal developmental toxicity study (Annex IX, Section 8.7.2.; test method: EU B.31./OECD TG 414) in a first species (rat or rabbit), oral route using the registered substance;**
2. **Extended one-generation reproductive toxicity study (Annex IX, Section 8.7.3.; test method: EU B.56./OECD TG 443) in rats, oral route with the registered substance specified as follows:**
   - **Ten weeks premating exposure duration for the parental (P0) generation;**
   - **Dose level setting shall aim to induce some 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); and**
   - **Cohort 3 (Developmental immunotoxicity).**

You may adapt the testing requested above according to the specific rules outlined in Annexes VI to X and/or according to the general rules contained in Annex XI of the REACH Regulation. In order to ensure compliance with the respective information requirement, any such adaptation will need to have a scientific justification, referring and conforming to the appropriate rules in the respective Annex, and an adequate and reliable documentation.

You are required to submit the requested information in an updated registration dossier by **30 September 2019**. You shall also update the chemical safety report, where relevant. The timeline has been set to allow for sequential testing.

The reasons of this decision are set out in Appendix 1. The procedural history is described in Appendix 2. Advice and further observations are provided in Appendix 3.

Appeal

This decision can be appealed to the Board of Appeal of ECHA within three months of its notification. An appeal, together with the grounds thereof, shall be submitted to ECHA in writing. An appeal has suspensive effect and is subject to a fee. Further details are described under http://echa.europa.eu/regulations/appeals.

Authorised[1] by Ofelia Bercaru, Head of Unit, Evaluation E3

[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

The decision of ECHA is based on the examination of the testing proposal(s) submitted by you.

You have proposed to cover the human health information requirements for a pre-natal developmental toxicity study (Annex IX, 8.7.2.) and an extended-one generation reproductive toxicity study (Annex IX, 8.7.3) by applying a read-across approach whereby these studies would be conducted with the analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (ZMB2) (hereafter referred to as "source substance") and then read-across to the substance subject to this decision (hereafter referred to as "target substance"), in accordance with the principles set out in Annex XI, Section 1.5.

ECHA has considered first the scientific validity of the read-across hypothesis (preliminary considerations: Section 0, below), before assessing the testing proposed (Sections 1. and 2. below).

0. Grouping and read-across

a. Legal Background on ECHA's assessment of the grouping of substances and read-across hypothesis

The evaluation by ECHA of testing proposals submitted by registrants aims at ensuring that generation of information is tailored to real information needs. To this end, it is necessary to consider whether programmes of testing proposed by you are appropriate to fulfil the relevant information requirements and to guarantee the identification of health and environmental hazards of substances. In that respect, the REACH Regulation aims at promoting wherever possible the use of alternative means, where equivalent results to the prescribed test are provided on health and environmental hazards.

Article 13(1) of the REACH Regulation provides that information on intrinsic properties of substances may be generated whenever possible by means other than vertebrate animal tests, including information from structurally related substances (grouping of substances and read-across), "*provided that the conditions set out in Annex XI are met*".

The first Recital and the first Article of the REACH Regulation establish the "*promotion of alternative methods for assessment of hazards of substances*" as an objective pursued by the Regulation. In accordance with that objective, ECHA considers whether a prediction of the relevant properties of the substance subject to the present decision by using the results of the proposed tests is plausible based on the information currently available.

According to Annex XI, 1.5 there needs to be structural similarity among the substances within a group or a category and furthermore, it is required that the relevant properties of a substance within the group can be predicted from the data for reference substance(s) by interpolation, and the data should be adequate for the purpose of classification and labelling and/or risk assessment. Furthermore, Annex XI, Section 1.5 lists several additional requirements, including that adequate and reliable documentation of the applied method is to be provided.

b. Description of the proposed grouping and read-across approach and information submitted

Your read-across hypothesis is based on similarities in the chemical structures and in the physico-chemical and toxicological properties between the source substance and the target substance. You state that "*ZMB2 is the zinc salt of MB2 and both substances are assumed to dissociate in aqueous media into the corresponding anion (1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione anion) and the corresponding cations*".

In order to support this consideration you have provided information (in the CSR section 5.9 and in an attachment to the endpoint study record of a testing proposal for an extended one-generation reproductive toxicity study) outlining similarities in physico-chemical properties on molecular weight, water solubility, log Pow and dissociation constant. You derived on that basis that "*Overall, based on the chemical structure of MB2 and its zinc salt ZMB2, a read across is justified*".

You have also provided a comparison of available systemic toxicity and reproductive toxicity data for source and target substances and conclude that "*Both substances have similar acute toxicity. Liver, thyroid and cholesterol effects are reported in the sub-acute toxicity study with MB2 and the Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test with ZMB2*".

Overall, you proposed that "*based on the similar toxicity profile of both substances and the available reproductive data with ZMB2 which led to a self-classification of both substances as GHS cat 2 reproductive toxicant"* the extended one-generation reproductive toxicity study and the developmental toxicity study should be conducted with the source substance to evaluate the reproductive and developmental toxicity for both substances.

c. ECHA analysis of the grouping approach and read-across hypothesis in light of the requirements of Annex XI, 1.5.

According to the provisions of Annex XI, section 1.5 of the REACH Regulation, application of the grouping and read-across concept requires that the properties of a substance may be predicted from data on another structurally similar substance.

You have presented information establishing a structural relationship between the source substance ZMB2 and the target substance MB2. Structural similarity is a prerequisite for applying the grouping and read-across approach. ECHA acknowledges that the source substance ZMB2 and the target substance MB2 are structural analogues. However, structural similarity is not *per se* sufficient to enable the prediction of human health properties. A read-across hypothesis establishing a basis for this prediction, and developed on the basis of structural similarity, is a fundamental aspect of a read-across approach.

ECHA understands that your read-across hypothesis is based on the claim that the source substance ZMB2 and the target substance MB2 dissociate in aqueous media to the common 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione anion and the corresponding cations, i.e. the zinc cation and the hydronium ion respectively. The dissociation of both the source and the target substances is a key element in your read-across hypothesis.

ECHA notes however that your read-across hypothesis relies on the assumption that the source substance dissociates to the same anion as the one formed from the dissociation of the target substance, i.e. in the common 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione anion. No qualitative or quantitative information characterising the dissociation of the source substance ZMB2 has been provided in the technical dossier to support this assumption. You indicated in the technical dossier that investigations of the dissociation of the source substance is not technically feasible and therefore you postulated that "*it is assumed that both substances dissociate in aqueous solution to the MB2 anion and the corresponding cation*". ECHA notes, that the possibility that the source substance remains as a stable molecule in aqueous solution cannot be dismissed and is actually supported by information included in the registration dossier of the source substance: "*ZMB2 does remain as a single molecule, at least to some degree*". This aspect appears to contradict with your read-across hypothesis and has not been accounted for in your read-across approach.

In the attachment to the endpoint study record for a testing proposal for an extended one-generation reproductive toxicity and in the CSR you have reported available information on the properties of the source substance ZMB2 and the target substance MB2. This information includes data obtained in a 28-day repeated-dose toxicity study performed with MB2 and results from a combined repeated dose toxicity study with the reproduction / developmental toxicity screening study conducted with the source substance ZMB2. ECHA observes similarities between the target organs identified from these studies with effects on the thyroid and the liver. ECHA notes that impaired fertility function as evidenced by a reduced mating performance, alteration of oestrous cyclicity and increased gestation length has been observed in the screening study conducted with the source substance ZMB2. No comparable information is available on reproductive toxicity for the target substance MB2. Considering the uncertainty on the dissociation of the source substance ZMB2, it is not possible to determine whether the evidence of reproductive toxicity observed in the screening study is due to exposure to the source substance ZMB2 as a single molecule or whether these findings are due to exposure to the 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione anion formed from the dissociation of the source substance ZMB2. Therefore, in the absence of information on the rate and extent of the dissociation of the source substance ZMB2, ECHA considers that you have not established that the properties of the target substance MB2 can be predicted from data on the source substance ZMB2.

In your comments to the draft decision, you refer to the CoRAP justification document produced by the evaluating member state and you assume on the basis of the information from that document that "*the evaluating member state considers a read-across between 2-mercaptobenzimidazole, MB2 and ZMB2 to be relevant for the toxicological evaluation of MB2 and ZMB2*" and that "*all information on 2-mercaptobenzimidazole, MB2 and ZMB2 are taken into account and an in depth evaluation of a potential read-across between MB2 and ZMB2 is expected to be conducted during the substance evaluation*".

ECHA stresses that in the CoRAP justification document referred to in your comments the term "group" refers to a number of structurally-related chemicals with potential endocrine disrupting properties considered together as a group for a purpose of prioritisation for further work, in this case for prioritisation for substance evaluation. This grouping does not imply that the conditions for grouping and read-across as laid down in Annex XI, section 1.5 of the REACH Regulation for the purpose of adaptation of standard information requirements are met and that the properties of substances in this group can be predicted from data on other substances within the group.

You conclude in your comments that "*no vertebrate studies should be conducted on MB2 or ZMB2 before this evaluation is conducted by the evaluating member state*". ECHA points out that the registration dossier subject to the decision issued to the registrant and the dossier update submitted on 06 June 2016 – submission number [redacted] – contain testing proposals for the information requirements of Annex IX, 8.7.2 for a pre-natal developmental toxicity study and of Annex IX, 8.7.3 for an extended one-generation reproductive toxicity study. According to Article 40 of the REACH Regulation, ECHA has the obligation to examine and to issue a decision on these testing proposals. In the context of this testing proposal examination, ECHA has assessed the read-across approach proposed by the registrant against the provisions of Annex XI, section 1.5 of the REACH Regulation and considered that for the reasons presented in the draft decision this adaptation cannot be accepted. In accordance with the provisions of Article 51 of the REACH Regulation, ECHA will notify the draft decision to the member states competent authorities (MSCAs). MSCAs may propose amendments to the draft decision to the Agency.

d. Conclusion on the read-across approach

In the absence of information characterizing the rate and extent of the dissociation of the source substance ZMB2 in the 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione anion and the zinc cation, ECHA considers that the information provided in the dossier does not verify your read-across hypothesis according to which the properties of the source and target substances are likely to be similar or to follow a regular pattern as a result of their structural similarity and behaviour in aqueous media.

Therefore ECHA concludes that you have not provided an adequate basis for predicting the properties of the target substance from the source substances as required by the provisions of Annex XI, section 1.5 of the REACH Regulation. As a consequence, the testing proposed on the read-across substance is not appropriate to fulfil the information requirement(s) of the substance subject to the present decision.

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

Pursuant to Article 40(3)(d) and (c) of the REACH Regulation, ECHA may reject a proposed test and require the Registrant to carry out other tests in cases of non-compliance of the testing proposal with Annexes IX, X or XI.

A pre-natal developmental toxicity study for a first species is a standard information requirement as laid down in Annex IX, Section 8.7.2. of the REACH Regulation. The information on this endpoint is not available for the registered substance but needs to be present in the technical dossier to meet the information requirements. Consequently there is an information gap and it is necessary to provide information for this endpoint.

You have submitted a testing proposal for a pre-natal developmental toxicity study in rats according to EU B.31./OECD TG 414 by the oral route with the analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (EC No 262-872-0).

However, as explained above in Appendix 1, section 0 of this decision, the proposed read-across cannot be accepted. Hence there is a need to test the registered substance.

ECHA considers that the proposed study is appropriate to fulfil the information requirement of Annex IX, Section 8.7.2. of the REACH Regulation.

You proposed testing with the rat as a first species. According to the test method EU B.31./OECD TG 414, the rat is the preferred rodent species and the rabbit the preferred non-rodent species. On the basis of this default consideration, ECHA considers testing should be performed with the rat or rabbit as a first species.

You proposed testing by the oral route. ECHA agrees that the oral route is the most appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 4.1, October 2015) R.7a, chapter R.7.6.2.3.2. Since the substance to be tested is a solid, ECHA concludes that testing should be performed by the oral route.

Therefore, pursuant to Article 40(3)(c) of the REACH Regulation, you are requested to carry out the study with the registered substance subject to the present decision: Pre-natal developmental toxicity study in a first species (rats or rabbits), oral route (test method: EU B.31./OECD TG 414).

Your originally proposed test for a pre-natal developmental toxicity study (OECD 414) conducted with the analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (EC No 262-872-0) is rejected according to Article 40(3)(d) of the REACH Regulation.

Notes for your consideration

For the selection of the appropriate species you are advised to consult ECHA *Guidance on information requirements and chemical safety assessment* (version 4.1, October 2015), Chapter R.7a, section R.7.6.2.3.2.

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

Pursuant to Article 40(3)(d) and (c) of the REACH Regulation, ECHA may reject a proposed test and require the Registrant to carry out other tests in cases of non-compliance of the testing proposal with Annexes IX, X or XI.
The basic test design of an extended one-generation reproductive toxicity study (Cohorts 1A and 1B, without extension of Cohort 1B to include a F2 generation, and without Cohorts 2A, 2B and 3) is a standard information requirement as laid down in column 1 of 8.7.3., Annex IX of the REACH Regulation if the available repeated dose toxicity studies (e.g. 28-day or 90-day studies, OECD TGs 421 or 422 screening studies) indicate adverse effects on reproductive organs or tissues or reveal other concerns in relation with reproductive toxicity. If the conditions described in column 2 of Annex IX are met, the study design needs to be expanded to include the extension of Cohort 1B, Cohorts 2A/2B, and/or Cohort 3.

Further detailed guidance on study design and triggers is provided in in ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.1, October 2015).

ECHA considers that concerns in relation with reproductive toxicity are observed in a 28-day repeated dose toxicity study conducted with the registered substance in rats via the oral route. More specifically, increased thyroid weight associated with histopathological changes have been observed in the high dose group males in that study.

Therefore the conditions of Annex IX, Section 8.7.3. are fulfilled and an extended one-generation reproductive toxicity study is an information requirement for the registered substance pursuant to Annex IX, Section 8.7.3.

The information on this endpoint is not available for the registered substance but needs to be present in the technical dossier to meet the information requirements. Consequently there is an information gap and it is necessary to provide information for this endpoint.

You have submitted a testing proposal for an extended one-generation reproductive toxicity study according to EU B.56./OECD TG 443 by the oral route to be performed with the analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (EC No 262-872-0). However, as explained above in Appendix 1, section 0 of this decision, the proposed read-across cannot be accepted. Hence there is a need to test the registered substance.

You have provided the following justification and specification of the design of the proposed study:

- Extension of cohort 1B: based on effects observed on the thyroid weight and associated histopathological changes in males and females and increased cholesterol in a 28-day repeated dose toxicity conducted with the registered substance and based on indications of similar toxicity and reproductive effects observed with the zinc salt of the registered substance in a combined repeated dose toxicity study with the reproduction /developmental toxicity screening test. Thyroid effects were supported also by an *in vitro* study. You conclude that "*the proposed EOGRTS should cover potential endocrine related toxicity and cohort 1B should be extended to include the F2 generation to elucidate toxicity to thyroid*".
- Inclusion of cohort 2: no triggers identified.
- Inclusion of cohort 3: no triggers identified for the registered substance. However, you considered that the "statistically significant reductions in male absolute organ weight values compared to control values were observed for the thymus in all dose groups" observed in a combined repeated dose toxicity study with the reproduction /developmental toxicity screening test performed with the zinc salt of the registered substance do constitute triggers for inclusion of cohort 3.
- Premating period duration: you indicated that "*The above mentioned thyroid effects might be due to direct toxicity or indirect as a consequence of a negative feedback via enhanced liver metabolism since liver weight and enzymes are increased at lower MB2 doses ([redacted] 1999). Since alterations in liver metabolism might take some time to become toxicologically significant a prolonged premating exposure is proposed. Overall, based on the liver toxicity and to investigate the thyroid mechanism, a 10 week premating period is proposed.*"

Adequate information on this endpoint needs to be present in the technical dossier for the registered substance to meet this information requirement. Thus, an extended one-generation reproductive toxicity study according to columns 1 and 2 of 8.7.3., Annex IX is required. ECHA considers that the proposed study designs requires modification to fulfil the information requirement of Annex IX, Section 8.7.3. of the REACH Regulation.
The following refers to the specifications of this required study.

Premating exposure duration and dose-level setting

You proposed a prolonged premating exposure period of 10 weeks based on potential alteration of liver metabolism requiring some time to become toxicologically relevant and in order to investigate the mechanism of thyroid toxicity.

To ensure that the study design adequately addresses the fertility endpoint, the duration of the premating exposure period and the selection of the highest dose level are key aspects to be considered. According to ECHA Guidance on information requirements and chemical safety assessmemtn R.7a, chapter R.7.6 (version 4.1, october 2015), the starting point for deciding on the length of premating exposure period should 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 because there is no substance specific information in the dossier supporting shorter premating exposure duration as advised in the ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.1, October 2015).

The highest dose level shall aim to induce some toxicity to allow comparison of effect levels and effects of reproductive toxicity with those of systemic toxicity. The dose level selection should be based upon the fertility effects with the other cohorts being tested at the same dose levels.

If there is no existing relevant data to be used for dose level setting, it is recommended that results from a range-finding study (or range finding studies) are reported with the main study. This will support the justifications of the dose level selections and interpretation of the results.

Extension of Cohort 1B

If the column 2 conditions of 8.7.3., Annex IX are met, Cohort 1B must be extended, which means that the F2 generation is produced by mating the Cohort 1B animals. This extension provides information also on the sexual function and fertility of the F1 animals.

You proposed to include an extension of Cohort 1B and provided justifications which are not fulfilling all the criteria described in column 2 of Section 8.7.3 of Annex IX and detailed in ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.1, October 2015). You have indicated in your testing proposal justification document that the Cohort 1B should be extended on the basis of increased in thyroid weight and histopathological findings in the thyroid observed in a 28-day repeated dose toxicity study performed with the registered substance. You also refer in your justification to positive findings in an *in vitro* lactoperoxidase inhibition study suggesting that the registered substance might be toxic to the thyroid gland. In addition you refer to the findings from a screening study for developmental/reproductive toxicity conducted with a substance structurally analogous to the registered substance and showing signs of reproducitve toxicity and other effects including thyroid toxicity. However, your justification does not include considerations establishing that the substance has uses leading to significant exposure of consumers or professionals as required by the provisions of letter (a) of column 2 of 8.7.3., Annex IX.

ECHA notes, that the use of the registered substance is leading to significant exposure of workers (PROCs 5, 6, 7, 8a, 8b, 10, 13, 14 and 21) during the production of tyres, rubber and plastic goods. However, based on the information in the registration dossier, there are no uses leading to significant exposure of consumers or professionals to the registered substance.

Thus, although the toxicity-criteria of indications of modes of action relating to endocrine disruption for extension of Cohort 1B is fulfilled (letter (b) of column 2 of 8.7.3., Annex IX) the exposure-criteria of significant exposure to consumers and/or professionals is not fulfilled (letter (a) of column 2 of 8.7.3., Annex IX).

Therefore, based on the information currently available ECHA does not consider that the criteria defined in column 2 of 8.7.3. of Annex IX to extend the Cohort 1B are met. Also, no further justification, according to the provision of Annex I, section 0.5. of the REACH Regulation, has been submitted which would merit exceeding the standard set of information requirement in order to address a specific risk.

The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Cohorts 2A and 2B

The developmental neurotoxicity Cohorts 2A and 2B need to be conducted in case of a particular concern on (developmental) neurotoxicity as described in column 2 of Section 8.7.3., Annex IX. When there are triggers for developmental neurotoxicity, both the Cohorts 2A and 2B are to be conducted as they provide complementary information.

You proposed not to include Cohorts 2A and 2B. You considered that there are no triggers to include the neurotoxicity endpoints on the basis of the information obtained from the 28-day repeated-dose toxicity study conducted with the registered substance.

ECHA notes that existing information on the registered substance itself and on substance structurally analogous to the registered substance, derived from available *in vivo* studies included in the technical dossier, show evidence of thyroid toxicity. Specifically, increased thyroid weight and histopathological changes have been observed in males from the high dose group in a 28-day repeated-dose toxicity study conducted with the registered substance. You hypothesised that this finding could be "*a consequence of a negative feedback via enhanced metabolism of circulating thyroid hormones*". Furthermore, thyroid gland hypertrophy has been reported in a combined repeated dose toxicity study with the reproduction /developmental toxicity screening test performed with the structurally analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (EC No 262-872-0). ECHA is of the opinion that these findings suggest that the registered substance itself and a substance structurally analogous to the registered substance may have the potential to cause relevant changes in thyroid hormones levels or signs of thyroid toxicity indicating such changes, suggesting a potential mode of action closely linked with (developmental) neurotoxic effects.

Therefore, ECHA considers that there are adequate triggers for developmental neurotoxicity investigations.

ECHA concludes that the developmental neurotoxicity cohorts 2A and 2B need to be conducted because there is a particular concern on (developmental) neurotoxicity based on the results from the above-identified *in vivo* studies on the registered substance itself and on the substance structurally analogous to the registered substance.

The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Cohort 3

The developmental immunotoxicity Cohort 3 needs to be conducted in case of a particular concern on (developmental) immunotoxicity as described in column 2 of 8.7.3., Annex IX.

You proposed to include Cohort 3 and provided justifications following the criteria described in column 2 of Section 8.7.3 of Annex IX and detailed in ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.1, October 2015).

You indicated in your justification for the testing proposal that no specific triggers for immunotoxicity have been observed in the 28-day repeated dose toxicity study conducted with the registered substance. Nevertheless, you considered that the significant reduction in thymus weight observed in all dose groups in the screening study conducted with the analogue substance 1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (EC No 262-872-0) do warrant the inclusion of the cohort 3 in the design of the proposed study.

ECHA agrees that based on the information from the structurally analogous substance to the registered substance the criteria to include Cohort 3 are met.

ECHA concludes that the developmental immunotoxicity Cohort 3 needs to be conducted because there is a particular concern on (developmental) immunotoxicity based on the results from the above-identified *in vivo* study on a substance structurally analogous to the registered substance.

The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Species and route selection

You proposed testing in rats. According to the test method EU B.56./OECD TG 443, the rat is the preferred species. On the basis of this default consideration, ECHA considers that testing should be performed in rats.

You proposed testing by the oral route. ECHA agrees that the oral route is the most appropriate route of administration for substances except gases to focus on the detection of hazardous properties on reproduction as indicated in ECHA *Guidance on information requirements and chemical safety assessment* (version 4.1, October 2015) R.7a, chapter R.7.6.2.3.2. Since the substance to be tested is a solid, ECHA concludes that testing should be performed by the oral route.

In your comments to the draft decision you state that ECHA used an "*inconsistent argumentation concerning a potential read-across throughout the draft decision*". ECHA understands that this refers to the rejection of the read-across approach between MB2 and ZMB2 and the reference to data on ZMB2 as triggers for inclusion of Cohort 3 in the design of the EOGRTS requested to be performed on MB2.

ECHA outlines that these two separate approaches, i.e. prediction of properties of a substance by means of read-across and consideration on particular concerns stemming from information on substances structurally analogous to the registered substance when setting the design of an EOGRTS follow different legal provisions and toxicological purpose.

The REACH Regulation Annex XI, section 1.5 sets the rules for using of grouping and read-across whereas the particular concerns from substances structurally analogous to the registered substance justify inclusion of further investigations in an EOGRTS as described in Annex IX, Section 8.7.3, column 2.

ECHA has assessed the proposed read-across against the requirements of Annex XI, Section 1.5 of the REACH Regulation and, for the reasons presented in the draft decision, concluded that these requirements were not met. ECHA did not question the structural similarity between the substance subject to the decision, MB2, and the analogue substance ZMB2, in its rejection of the read-across approach.

Regarding the design of the EOGRTS, the inclusion of Cohorts 2A and 2B and/or Cohort 3 is justified if there is "*existing information on effects caused by substances structurally analogous to the substance being studied, suggesting such effects or mechanisms/modes of action*". As shown by this provision of Annex IX, 8.7.3, Column 2, and further explained in ECHA *Guidance on information requirements and chemical safety assessment* R.7a, chapter R.7.6 (version 4.1, October), read-across assessment by ECHA is not needed to identify a particular concern and justify inclusion of Cohorts 2A and 2B and/or Cohort 3. A particular concern to include the Cohorts 2A, 2B and/or Cohort 3 can be based on information from structurally analogous substances independently of whether a read-across from such structural analogues is accepted by ECHA.

In this specific case, even though ECHA concluded on a rejection of the read-across approach for the reasons indicated in the draft decision, ECHA recognises that ZMB2 is a structurally related analogue to the substance subject to this decision, MB2. As outlined above, ECHA considers that the information obtained in a screening study conducted with ZMB2 show a concern for immunotoxic and neurotoxic effects suggesting that similar effects or similar mechanisms/modes of action are likey to apply also for the substance subject to this decision. Therefore, these effects are considered to constitute triggers for the inclusion of Cohorts 2A/2B and Cohort 3 in the design of the requested study. ECHA does not consider that this constitutes an inconsistent argumentation on the analogue substance in the context of the read-across approach.

You also indicate in your comments that "*ECHA concluded on the study-design specifications without taking into account the CoRAP justification document on MB2 and ZMB2*" where the evaluating member state considered that it is not yet demonstrated that ZMB2 and MB2 do interfere with the activity of the thyroid-peroxidase-enzyme and the activity of the deiodinase-enzyme and that potential non-standard ED-relevant tests might be required to clarify this point. You further considered that the endocrine-related adverse effects observed with the substance subject to this decision may be secondary to a non-endocrine-related toxicity, i.e. the induction of liver metabolizing enzymes.

On that basis you concluded that an assessment of reproductive toxicity and potential direct endocrine disrupter activity or indirect endocrine-related adverse effects and a decision on the number and design of EOGRTS to be conducted should occur during the Substance Evaluation process.

ECHA observes that the standard information requirements for both the pre-natal developmental toxicity study and the extended one-generation reproductive toxicity study are required to be met for compliance under REACH. These studies have been proposed by you and the testing proposals are still included in the updated dossier submitted on 06 June 2016 – submission number [redacted].

ECHA clarifies that the design of the EOGRTS requested has been established on the basis of the information provided in the registration dossier with submission number [redacted] and maintained in the updated dossier with the submission number [redacted]. ECHA considers that the information reported in the CoRAP justification document does not provide reasons to change the requested study design, as that information supports the concern that is related to thyroid effects.

Hence, the proposed studies requested in this decision are necessary to fulfil the information requirements under the REACH Regulation and will provide further information on the toxicological properties, including certain endocrine disrupting modes of action, also to be used during the substance evaluation process
Therefore, pursuant to Article 40(3)(c) of the REACH Regulation, you are requested to carry out the study with the registered substance subject to the present decision: Extended one-generation reproductive toxicity study (test method EU B.56./ OECD TG 443), in rats, oral route, according to the following study-design specifications:
Ten weeks premating exposure duration for the parental (P0) generation;

- Dose level setting shall aim to induce some 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); and
- Cohort 3 (Developmental immunotoxicity)

while your originally proposed test for Extended one-generation reproductive toxicity study (test method OECD TG 443) with the analogue substance (1,3-dihydro-4(or5)-methyl-2H-benzimidazole-2-thione, zinc salt (EC No 262-872-0) is rejected according to Article 40(3)(d) of the REACH Regulation.

Note for your considerations:

The conditions to include the extension of Cohort 1B are currently not met. However, you may expand the study by including the extension of Cohort 1B if new information becomes available after this decision is issued to justify such an inclusion. Inclusion is justified if the new information shows triggers which are described in column 2 of Section 8.7.3., Annex IX and further elaborated in ECHA *Guidance on information requirements and chemical safety assessment* R.7.a, chapter R.7.6 (version 4.1, October 2015). You may also expand the study to address a concern identified during the conduct of the extended one-generation reproduction toxicity study and also due to other scientific reasons in order to avoid a conduct of a new study. The justification for the expansion must be documented. The study design must be justified in the dossier and, thus, the existence/non-existence of the conditions/triggers must be documented.

Appendix 2: Procedural history

ECHA received your registration containing the testing proposal(s) for examination pursuant to Article 40(1) on 29 April 2013.

ECHA held a third party consultation for the testing proposal(s) from 15 April 2014 until 30 May 2014. ECHA did not receive information from third parties.

This decision does not take into account any updates after **8 August 2016**, 30 calendar days after the end of the commenting period.

The decision making followed the procedure of Articles 50 and 51 of the REACH Regulation, as described below:

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.
You updated your registration on 06 June 2016. ECHA took the information in the updated registration into account, and did not amend the draft decision. The updated information is reflected in the Reasons (Appendix 1).

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

ECHA received proposal(s) for amendment and modified the draft decision.

ECHA invited you to comment on the proposed amendment(s).

ECHA referred the draft decision to the Member State Committee.

Your comments on the proposed amendment(s) were taken into account by the Member State Committee.

In addition, you provided comments on the draft decision. These comments were not taken into account by the Member State Committee as they were considered to be outside of the scope of Article 51(5).

The Member State Committee reached a unanimous agreement on the draft decision in its MSC-52 written procedure and ECHA took the decision according to Article 51(6) of the REACH Regulation.

Appendix 3: Further information, observations and technical guidance

1. The substance subject to the present decision is provisionally listed in the Community rolling action plan (CoRAP) for start of substance evaluation in 2018.

2. This decision does not imply that the information provided in your registration dossier is in compliance with the REACH requirements. The decision does not prevent ECHA from initiating a compliance check on the registration at a later stage.

3. Failure to comply with the request(s) in this decision, or to fulfil otherwise the information requirement(s) with a valid and documented adaptation, will result in a notification to the Enforcement Authorities of the Member States.

4. In carrying out the test(s) required by the present decision it is important to ensure that the particular sample of substance tested is appropriate to assess the properties of the registered substance, taking into account any variation in the composition of the technical grade of the substance as actually manufactured or imported. If the registration of the substance covers different grades, the sample used for the new test(s) must be suitable to assess these. Furthermore, there must be adequate information on substance identity for the sample tested and the grade(s) registered to enable the relevance of the test(s) to be assessed.