CONSIDERATIONS OF ALTERNATIVE METHODS ON TESTING PROPOSALS IN YOUR REGISTRATION

Please complete this form and provide information for each of the points below.

If you have more than one testing proposal, please copy and paste the three bullet points within the same document and complete the details as appropriate for each testing proposal.

This document will be published on ECHA website along with the third party consultation on the testing proposal(s).

Public substance name: BORON ORTHOPHOSPHATE
EC Number (omit if confidential): 236-337-7
CAS Number (omit if confidential): 13308-51-5

Date of considerations: 27 June 2016

- **Hazard endpoint for which vertebrate testing was proposed:**

  Reproductive toxicity (pre-natal developmental toxicity) with the registered substance

- **Considerations that the general adaptation possibilities of Annex XI of the REACH Regulation were not adequate to generate the necessary information** (instruction: please address all points below):

  - available GLP studies
    There are no GLP-compliant studies available on pre-natal developmental toxicity with boron orthophosphate.

  - available non-GLP studies
    There are no non-GLP studies available on pre-natal developmental toxicity with boron orthophosphate.

  - historical human data
    There are no appropriate historical human data available addressing the endpoint pre-natal developmental toxicity.

  - (Q)SAR
    (Q)SAR tools sufficiently addressing the endpoint pre-natal developmental toxicity are currently not available.

  - *in vitro* methods
    No validated or regulatory accepted alternative methods are available for replacing animal testing with respect to pre-natal developmental toxicity.

  - weight of evidence
    There are no studies available on pre-natal developmental toxicity with boron orthophosphate which could be used in a weight of evidence approach.

  - grouping and read-across
    No appropriate read-across substance was identified in order to fulfil endpoint pre-natal developmental toxicity.

- substance-tailored exposure driven testing [if applicable]
  Not applicable.

- [approaches in addition to above [if applicable]
  Not applicable.

- other reasons [if applicable]
  Not applicable.

- **Considerations that the specific adaptation possibilities of Annexes VI to X (and column 2 thereof) were not applicable** (instruction: free text):

There is no data available on developmental toxicity with boron orthophosphate (CAS 13308-51-5). Furthermore, no appropriate read-across substance was identified and no validated or regulatory accepted alternative methods are available for replacing animal testing with respect to developmental toxicity.
In order to fulfil the standard information requirements, a GLP-compliant prenatal developmental toxicity study in the rat via the oral route following OECD 414 was proposed, according to Annex IX, Column I, 8.7.2.

Taking into account all available information on boron orthophosphate and structurally similar substances (boric acid and its salts), it was considered that read-across is not an adequate method for data gap filling relating to developmental toxicity for boron orthophosphate due to differences in absorption.
Boron orthophosphate is an inorganic substance with a molecular weight of 106 g/mol and a low solubility in water (0.143 g/L). In general, molecular weights below 500 g/mol are favorable for absorption via the gastrointestinal (GI) tract, but absorption may be limited by low water solubility (ECHA, 2014). Based on the molecular weight and water solubility of boron orthophosphate, absorption via the GI tract is likely to be low.
This assumption is supported by the experimental animal data on boron orthophosphate. The available data on acute oral toxicity showed that a single dose of 2000 mg/kg bw caused deaths in 3 of 5 female rats (Bradshaw, 2013). Clinical signs observed were hunched posture, lethargy, ataxia, pilo-erection, emaciation, labored respiration, decreased respiratory rate and dehydration. At necropsy, haemorrhagic, ulcerated and epithelial sloughing of the gastric mucosa was noted. At a dose level of 300 mg/kg bw, no mortality and no signs of systemic toxicity were observed. Based on the available data, boron orthophosphate caused rather local effects at the site of contact at a dose level of 2000 mg/kg bw. Furthermore, it can be assumed that the observed clinical signs such as hunched posture could be due to discomfort caused by irritation. Therefore, systemic toxicity is considered to be limited.
An acute inhalation toxicity study has shown that no deaths occurred in male and female rats at 5.31 mg/L boron orthophosphate in air (Griffiths, 2013). Clinical signs observed were: hunched posture, pilo-erection and red/brown staining around the eyes or snout, wet fur. However, these clinical signs were considered to be associated with the restraint procedure and were not indicative of toxicity. In addition to the clinical signs described above, increased respiratory rate was noted in all animals during exposure, on removal from the chamber and one hour post-exposure. Animals recovered to appear normal from Days 7 to 10 post-exposure. Two males and two female animals exhibited bodyweight losses on the first day post-exposure. No macroscopic abnormalities were found at necropsy.

In a 90-day repeated dose toxicity study according to OECD guideline 408, no treatment-related effects were observed with 100, 300 and 1000 mg boron orthophosphate/ kg bw/day in mice (Entzian, 2016).
With respect to the doses administered and as no treatment-related effects were observed, it can be suggested that boron orthophosphate possesses either a low toxic potency or a low absorption in combination with a low systemic toxicity.
However, if absorbed, boron orthophosphate can be distributed in various tissues based on the low molecular weight. Furthermore, it can be assumed that boron orthophosphate is hydrolysed to the more soluble and polar products, borate and phosphate ions. Due to the physico-chemical properties of the hydrolysis products, urinary excretion is the most probable route of elimination and bioaccumulation is unlikely.

In contrast to the physico-chemical properties of boron orthophosphate, boric acid is highly water soluble (63.5 g/L at 20 °C) and slightly soluble in ethanol (Haynes, 2010). In humans and animals, boric acid and its salts (borate) are readily absorbed from the GI tract and from the respiratory tracts (IPCS, 1998). Greater than 90% of the administered doses of these compounds are absorbed, as evidenced by increased levels of boron in the blood, tissues, or urine or by systemic toxic effects of exposed animals (IPCS, 1998). Results from animal studies have shown that boric acid and borate caused reproductive and developmental toxicity in the exposed animals (IPCS, 1998).
Feeding studies in different animal species (rats, mice and dogs) have consistently demonstrated that the male reproductive system is the principal target in animals, although effects on the female reproductive system have also been reported. Testicular damage ranging from mildly inhibited spermiation to complete atrophy has been demonstrated following oral administration of boric acid. In a 2-year study in rats, effects on male fertility were observed at lower dose levels compared to dose levels where signs of general toxicity appeared. For this 2-year study, 17.5 mg boron/kg bw/day was considered a NOAEL for male and female fertility (EFSA, 2004). Developmental toxicity of boric acid was investigated in the rat, the rabbit and the mouse. In the rat, developmental toxicity (decreased fetal weight at 13.7 mg boron/kg bw/day) occurred in the absence of marked maternal toxicity. For developmental toxicity in rats, a NOAEL of 9.6 mg boron/kg bw/day (equivalent to 55 mg boric acid/kg bw/day) has been derived by Price et al. (1996). The adverse effects of boric on developmental toxicity and on fertility observed across species (including mice) were very similar, both in nature and effective doses (EFSA, 2013).

When comparing the data reported for boric acid and boron orthophosphate, it is obvious that the absorption behaviour is different. It is likely to be low as indicated by the low systemic toxicity in the available animal studies on boron orthophosphate, whereas boric acid is rapidly absorbed following oral exposure and has a high toxic potency relating to reproductive and developmental toxicity. Thus, read-across from boric acid to boron orthophosphate is considered to be inappropriate and not applicable in the present case due to the unsimilar toxicokinetics of both compounds. Therefore, the available data on boric acid cannot be used within a read-across approach in order to fulfil the standard information requirements of boron orthophosphate.

In conclusion, taking into account all available data, a GLP-compliant prenatal developmental toxicity study in the rat via the oral route following OECD 414 is proposed according to Annex IX, Column I, 8.7.2.

References

Bradshaw J (2013). Boron orthophosphate: Acute oral toxicity in the rat - fixed dose method. Project number: 41204125. Harlan Laboratories Ltd, Derbyshire.
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EFSA (European Food Safety Authority), 2004. Scientific Opinion of the Panel on Dietetic Products, Nutrition and Allergies on a request from the Commission related to the Tolerable Upper Intake Level of Boron (Sodium Borate and Boric Acid). The EFSA Journal 2004, 80, 1–22.
EFSA (European Food Safety Authority), 2013. Scientific Opinion of the re-evaluation of boric acid (E 284) and sodium tetraborate (borax) (E 285) as food additives. The EFSA Journal 2013, 11(10), 1-50
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Haynes WM (Ed.), 2010. Handbook of chemistry and physics, 91st edn (2010–2011). CRC Press, Boca Raton, FL, USA.
IPCS (International Programme on Chemical Safety), 1998. Environmental Health Criteria for boron. Available online: http://www.inchem.org/documents/ehc/ehc/ehc204.htm.
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