Annex to the BPC Opinion on a request pursuant to Article 15(2) of Regulation (EU) No 528/2012 on the
review of approval of the active substances iodine and polyvinylpyrrolidone iodine

Applicant's assessment of endocrine-disrupting properties of iodine and the rapporteur's comments on it

Date: June 2022

Rapporteur's comments

The rapporteur (Sweden) has made no changes to this assessment submitted by the applicant (Iodine Registration Group) in May 2020 within the scope of the review of approval of the active substance. The rapporteur's comments (June 2022) on the applicant's assessment are included within red boxes like this one.

Assessment of estrogen, androgen, thyroid and steroidogenic (EATS) mediated endocrine disrupting (ED) properties of iodine (CAS no. 7553-56-2)

Sponsor: Iodine Registration Group (IRG)

Reporting: [redacted]

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Date: 18 May 2020

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Rapporteur's comments

The rapporteur has updated the page numbers in the above Table of Contents.

Abbreviations

AMA	Amphibian Metamorphoses Assay
CAR	Competent Authority Report
CAS-No	Chemical Abstracts Service Number
EADB	Estrogenic Activity Database
EAS	estrogen, androgen and steroidogenic
EATS	estrogen, androgen, thyroid and steroidogenic
ECHA	European Chemicals Agency
ED	endocrine disruption
EDC	endocrine disrupting chemical
ED GD	Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009
EDKB	Endocrine Disruptor Knowledge Base
EFSA	European Food Safety Authority
FAO	Food and Agriculture Organization of the United Nations
FDA	U.S. Food and Drug Administration
GRAS	Generally Recognized As Safe
HPT	hypothalamic-pituitary-thyroid
ICCIDD	International Council for Control of Iodine Deficiency Disorders
IRG	Iodine Registration Group
NIS	sodium/iodide symporter
NOAEL	no observed adverse effect level
NRI	reference nutrient intake
OECD	Organisation for Economic Co-operation and Development
PEC_{sw}	predicted environmental concentration (surface water)
PVP	Polyvinylpyrrolidone
SCF	Scientific Committee on Food
SMILES	simplified molecular-input line-entry system
T	Thyroid
T3	triiodothyronine
T4	thyroxine
ToxCast	Toxicity Forecaster
TRH	thyrotropin releasing hormone
TSH	thyroid stimulating hormone
UL	upper limit
UNICEF	United Nations Childrens' Emergency Fund
US EPA	United States Environmental Protection Agency
WHO	World Health Organisation

Introduction

With the date of 18 February 2020 the European Commission informed members of the Iodine Registration Group (IRG) that by having indications on endocrine disruptor (ED) properties of the active substances the early review has been started for iodine and PVP iodine. In accordance with Article 15(1) of Regulation (EU) No 528/2012 the applicants got the opportunity to provide comments in this early review process.

Within this context, the IRG decided to prepare a full assessment of estrogen, androgen, thyroid and steroidogenic (EATS) mediated endocrine disrupting properties of iodine and PVP-iodine. The assessment for iodine is presented in this document.

Rapporteur's comments

In PVP-iodine, PVP is a carrier of iodine and there is no chemical bond between these. PVP is a water-soluble polymer that is biologically inert and non-toxic with good tolerance (Kurakula and Rao, 2020). Therefore, the assessment of ED properties of iodine is applicable also to PVP-iodine.

Kurakula, M. and Rao, G.S.N.K. 2020. Pharmaceutical assessment of polyvinylpyrrolidone (PVP): As excipient from conventional to controlled delivery systems with a spotlight on COVID-19 inhibition. Journal of Drug Delivery Science and Technology 60; 102046.

In Part A of the document a scientific statement on the assessment of T-mediated ED properties of iodine is provided, whereas Part B of the document focuses on the potential EAS-mediated endocrine disrupting properties of iodine.

Executive summary

Iodine is an essential dietary trace element (micronutrient), required as a structural and functional element of the thyroid hormones thyroxine (T4) and triiodothyronine (T3), which play critical roles in the carbohydrate, lipid, protein and mitochondrial energy metabolism and are particularly essential during embryogenesis and growth (WHO, 1989; EFSA, 2014). To ensure a sufficient intake and to prevent iodine deficiency disorders, iodine supplementation is required and recommended (WHO/FAO, 2004; EFSA, 2014).

The fact that (i) iodine is an essential dietary trace element with a defined key role in the biosynthesis of the thyroid hormones T4 and T3 (T modality of the hormone system) and the point that (ii) dietary intake and even food supplementation are recommended to ensure a sufficient iodine intake in the population show, that an entirely hazard based endocrine disruption (ED) assessment as outlined in the "Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009" (ED GD; ECHA/EFSA, 2018) especially with focus on T modality is not meaningful for iodine, and possibly beyond the scope of the Commission Delegated Regulation (EU) 2017/2100.

If the ED properties of iodine and other essential micronutrients would be assessed from a scientific perspective, the assessment should consider not only the hazard, but also take into account the potency and exposure. However, risk assessments for the approval of iodine as biocidal active substance and for the authorisation of iodine-based biocidal products already consider the upper limit (UL) of iodine intake which takes into account the endocrine effects of iodine and represents a conservative basis for the risk assessment. Therefore, regulating iodine as endocrine disrupting chemical (EDC) would not provide any additional safety.

Rapporteur's comments

(Details of the rapporteur's assessment of the T-modality are available in the BPC Opinion on the Art. 15 request on (PVP-)iodine.)

The Commission Delegated Regulation (EU) 2017/2100, setting out scientific criteria for identification of biocidal active substances with endocrine-disrupting properties, has made no exceptions to substances that are essential nutrients; and the criteria are only hazard based. Therefore, the rapporteur does not agree with the applicant's comments on the scope of the Regulation and that the potency and exposure should also be taken into account.

Iodine in excess of physiological needs meets the scientific criteria set out in the Commission Delegated Regulation (EU) 2017/2100 – it shows adverse effects (thyroid disorders) in humans; it has an endocrine mode of action (disruption of thyroid hormones metabolism and hypothalamic-pituitary-thyroid axis); and the adverse effects are a consequence of the endocrine mode of action. Therefore, the biocidal active substances, iodine and PVP-iodine, are identified as having endocrine-disrupting properties with respect to humans.

Since iodine and PVP-iodine meet the criteria for humans, they also meet the criteria for being endocrine disruptor in non-target organisms. This is supported by scientifically established understanding that external administration of iodine via water or diet to amphibia interferes

with endocrine mechanisms related to the thyroid and subsequently in excess of physiological needs lead to adverse effects in intact organisms. The most obvious adverse effect is accelerated metamorphosis.

Nevertheless, an assessment of potential EAS-mediated ED properties of iodine was performed according to the ED GD to make this information available for an independent assessment.

The present assessment was based on a weight of evidence approach and revealed that no EAS-related activity is attributable to iodine. Additionally no toxicologically significant EAS–mediated adversity was observed in the available data set. Therefore "*ED criteria regarding EAS modalities are not met*" for iodine.

Rapporteur's comments

Relevant data is lacking to conclude on EAS-modalities for (PVP-)iodine. The studies required for EAS-related activity (OECD 441 and OECD 456; OECD 229/230 and OECD 231) and EAS-mediated adversity (OECD 416, ver. 2001 or OECD 443; OECD 240/OPPTS 850.1500 and OECD 241/231) to be considered as sufficiently investigated according to the ECHA/EFSA ED Guidance are lacking.

Part A: Scientific statement on the assessment of thyroid (T) mediated endocrine disrupting (ED) properties of iodine

Summary

Iodine is an essential dietary trace element (micronutrient), required as a structural and functional element of the thyroid hormones thyroxine (T4) and triiodothyronine (T3), which play critical roles in the carbohydrate, lipid, protein and mitochondrial energy metabolism and are particularly essential during embryogenesis and growth (WHO, 1989; EFSA, 2014).

As for other essential trace elements and vitamins, there is an acceptable range of daily intake for iodine. Both iodine deficiency (reference nutrient intake (NRI) not reached) and high iodine intake (no observed adverse effect level (NOAEL) exceeded) of a certain degree cannot be fully compensated by homeostatic mechanisms and consequently lead to thyroid disorders (u-shaped dose-response curve) (EFSA, 2014, Renwick, 2006).

Iodine deficiency is a known worldwide public health issue (Lazarus, 2014). Iodine deficiency disorders lead to insufficient thyroid function (hypothyroidism) (EFSA, 2014). They are seen at all stages of development and are particularly of concern in pregnancy and infancy (EFSA, 2014, WHO/UNICEF, 2007a). Besides iodine deficiency, excessive iodine intake may also accelerate the development of sub-clinical thyroid disorders to overt hypothyroidism or hyperthyroidism, increase the incidence of autoimmune thyroiditis and increase the risk of thyroid cancer (EFSA, 2014; Laurberg *et al*., 1998; Teng *et al*., 2006). However, if iodine intake is carefully monitored for both iodine deficiency and excess, the benefits from a decrease of the substantial risk of iodine deficiencies will clearly outweigh the relatively small risk associated with iodine excess (Laurberg, 2010; WHO/FAO, 2004, Zimmermann, 2009).

To ensure a sufficient intake, iodine supplementation is required and recommended by organisations such as the World Health Organisation (WHO), International Council for Control of Iodine Deficiency Disorders (ICCIDD), European Food Safety Authority (EFSA) and U.S. Food and Drug Administration (FDA).

Within this context, iodine is defined as “food” according to Regulation (EC) No 178/2002 and is furthermore approved as food supplement according to Directive 2002/46/EG. Likewise the FDA considered iodine compounds as Generally Recognized As Safe (GRAS) and allows its use in table salt and as dough strengthener.

The fact that (i) iodine is an essential dietary trace element with a defined key role in the biosynthesis of the thyroid hormones T4 and T3 (T modality of the hormone system) and the point that (ii) dietary intake and even food supplementation are recommended to ensure a sufficient iodine intake in the population show, that an entirely hazard based endocrine disruption (ED) assessment as outlined in the “Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009” (ED GD) especially with

focus on T modality is not meaningful for iodine, and possibly beyond the scope of the Commission Delegated Regulation (EU) 2017/2100. If the ED properties of iodine and other essential micronutrients would be assessed from a scientific perspective, the assessment should consider not only the hazard, but also take into account the potency and exposure.

However, risk assessments for the approval of iodine as biocidal active substance and for the authorisation of iodine-based biocidal products already consider the upper limit (UL) of iodine intake which takes into account the endocrine effects of iodine and represents a conservative basis for the risk assessment. Therefore, regulating iodine as endocrine disrupting chemical (EDC) would not provide any additional safety.

1. ED assessment for essential dietary trace elements (micronutrients)

The "Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009" (ED GD; ECHA/EFSA, 2018) provides guidance on the hazard identification for endocrine-disrupting properties. However, it does not provide guidance on how to further characterise the hazard potential of a substance or the risk to humans or non-target organisms (ECHA/EFSA, 2018). Therefore, the approach as indicated in the ED GD (ECHA/EFSA, 2018) is meaningful for xenobiotics with potential endocrine disruption (ED) effects. However, it does not seem meaningful for essential dietary components like vitamins, minerals or trace elements.

Essential dietary components such as iodine are required for the normal physiological function of the human body and underlie a homeostasis within the acceptable range of daily intake. They cannot be synthesized in the body, either at all or in sufficient quantities, and therefore need to be supplied usually via dietary intake. This definition shows that an entirely hazard based ED assessment is not adequate for essential dietary components like iodine with its known and desired physiological endocrine mode of action (T modality of the hormone system) playing a key role in the biosynthesis of the thyroid hormones thyroxine (T4) and triiodothyronine (T3). This conclusion is in line with the Assessment Report of iodine (including PVP-iodine) for PT 1, 3, 4 and 22 (AR, 2013) stating that "*the concept of endocrine disruption is not meaningful for essential elements such as iodine*".

Rapporteur's comments

The statement that "the concept of endocrine disruption is not meaningful for essential elements such as iodine" was made in the Assessment Report that was prepared before the scientific criteria for endocrine-disrupting properties were available. The rapporteur (which is also the eCA that prepared the Assessment Report) notes that the criteria make no exception for essential elements that are biocidal active substances.

If the ED properties of essential dietary elements and other essential micronutrients would be assessed from a scientific perspective, the assessment should consider not only the hazard but also take into account the potency and

exposure. However, the criteria for the identification of ED substances as set out in the Commission Delegated Regulation (EU) 2017/2100 do not consider the relevant biological threshold of effect. This aspect was also criticised in a summary report recently published by the European Commission in the context of a "fitness check" of the EU legislation with regard to EDC. The stakeholders expressed that the hazard-based approach criteria for identifying EDC and for the ED assessment should be combined in the decision making with a risk-based approach (European Commission, 2020). Also the EFSA Scientific Committee suggested that uncertainties associated with a hazard-based approach for the management of EDC should be addressed using a risk assessment approach, *i.e.* considering both hazard as determined *in vivo* and exposure (EFSA, 2013).

For iodine intake a conservative Upper Limit (UL) of 600 µg/day for adults and 200 µg/day for infants was derived in the Assessment Report of iodine (including PVP-iodine) for PT 1, 3, 4 and 22 (AR, 2013) based on the UL for iodine established by the Scientific Committee on Food (SCL). This UL was used for performing human health exposure and risk assessments as well as dietary risk assessments for the approval of iodine as biocidal active substance and for the authorisation of iodine-based biocidal products. This value was derived taking into account the endocrine effects of iodine and represents a conservative basis for the risk assessment. Against this background, regulating iodine as EDC would not provide any additional safety.

The authoritative definition of an endocrine disruptor of the International Programme on Chemical Safety (IPCS), which has been endorsed and accepted by the Commission and EFSA, is: "*an exogenous substance or mixture that alters function(s) of the endocrine system and consequently causes adverse health effects in an intact organism, or its progeny, or (sub)populations*". This definition was adapted and included in the Annex to Commission Delegated Regulation (EU) 2017/2100 and does not apply to iodine and other essential elements for the following reasons:

- The definition refers to an endocrine system that functions in the absence of an exogenous substance and is disrupted in its presence. This is not the situation for iodine: without sufficient supply of iodine as essential dietary trace element (micronutrient), the endocrine system of the thyroid does not function. Consequently, iodine must be considered as an EAD (endocrine-active substance) as defined by EFSA, 2013 (EFSA Journal 2013; 11(3): 3132) but should not be considered as EDC.
- The definition further refers to adverse health effects caused by an exogenous substance in an intact organism, or its progeny, or (sub)populations. Since in the absence of iodine, the organism, or its progeny, or (sub)populations is/are not intact, this part of the definition does not apply to iodine or to other essential elements.

Rapporteur's comments

The scientific criteria set out in the Commission Delegated Regulation (EU) 2017/2100 (that are based on the WHO/IPCS definition of endocrine disruptors) are applied for the identification of

endocrine-disrupting properties of biocidal active substances. Iodine and PVP-iodine, placed on the market as biocidal active substances are exogenous substances.

2. Physiological relevance of iodine

Iodine is an essential dietary trace element (micronutrient), required as an obligatory structural and functional component of the thyroid hormones thyroxine (T4) and triiodothyronine (T3). The thyroid hormones are necessary for the regulation of mitochondrial energy metabolism as well as cellular oxidation, thermoregulation, intermediate metabolism, carbohydrate, lipid and protein metabolism and nitrogen retention. They are particularly required during embryogenesis and growth, and the development of neurological and cognitive functions (WHO, 1989; EFSA, 2014). The target organs are, in particular, the developing brain, affecting the development of hearing and vision, muscles, the heart, the pituitary gland and the kidney, but also the reproductive system and the bones (EFSA, 2014).

3. Thyroid gland's regulatory circuit

Thyroid function is regulated at multiple points, whereby the primary point of regulation is the synthesis of thyroid hormones by the thyroid gland. For thyroid hormone biosynthesis, the thyroid gland is stimulated by the hypothalamic-pituitary-thyroid (HPT) axis to trap iodine and to produce and release T4 and T3 (Figure A-1).

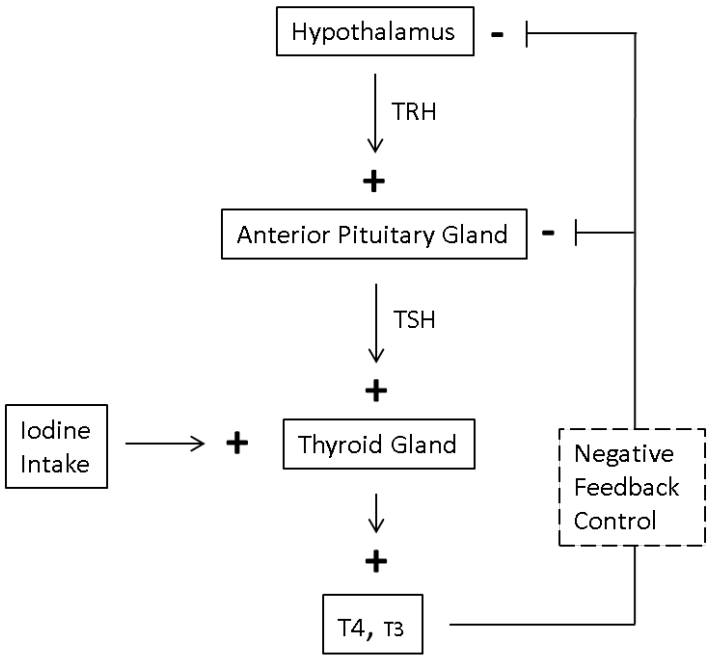

Figure A-1: Thyroid gland's regulatory circuit. The thyroid gland is stimulated by the hypothalamic-pituitary-thyroid (HPT) axis to trap iodine and to produce and release thyroxine (T4) and triiodothyronine (T3). The T3 and T4 levels in the circulation have a negative feedback on the HPT-axis. Changes in T3 and T4 levels are

regulated by an increases or / and decreases of in TSH or / and TRH (modified from Behringer, 2018).

The hypothalamus secretes thyrotropin releasing hormone (TRH) which acts on the anterior lobe of the pituitary gland. After stimulation the anterior pituitary synthesizes and secretes thyroid stimulating hormone (TSH). This acts on the thyroid gland (Behringer, 2018). TSH stimulates iodide transport via the sodium/iodide symporter (NIS) from the blood into thyroid cells, oxidation of iodide to iodine, and iodine binding to tyrosine (EFSA, 2014). The prohormone T4 and partly also the active thyroid hormone T3 are produced in the thyroid gland. They are released into the blood and transported to the target tissues where further prohormone T4 is converted into the active hormone T3 (Behringer, 2018).

Adequate T3 and T4 levels in the circulation have a negative feedback on the HPT-axis. Changes in T3 and T4 levels are regulated by increases and/or decreases in TSH and/or TRH (Behringer, 2018).

The thyroid gland has the capacity and holds the machinery to handle iodine efficiently when the availability of iodine becomes scarce, and when iodine is available in excessive quantities. The latter situation is handled by the thyroid by acutely inhibiting the organification of iodine, the so-called Wolff-Chaikoff effect (Markou *et al.*, 2001).

4. Thyroid disorders as consequences of iodine deficiency or excess

Despite the existence of the thyroid gland's regulatory circuit, clearly insufficient (reference nutrient intake (NRI) not reached) or excessive iodine intakes (no observed adverse effect level (NOAEL) exceeded) can lead to thyroid disorders (u-shaped dose-response curve) (EFSA, 2014, Renwick, 2006).

Iodine deficiency

Iodine deficiency is a known worldwide public health issue (Lazarus, 2014). Iodine deficiency disorders, which are clinical effects of iodine deficiency as a result of insufficient intakes, lead to insufficient thyroid function (hypothyroidism) (EFSA, 2014). They are seen at all stages of development and are particularly of concern in pregnancy and infancy (Table A-1) (EFSA, 2014, WHO/UNICEF, 2007a).

Chronic iodine deficiency may result in compensatory thyroid hypertrophy/ hyperplasia. The enlargement of the thyroid gland is called endemic goiter. A goiter is initially diffuse but later may become nodular with the appearance of autonomous nodules. This may subsequently cause hyperthyroidism (EFSA, 2014, WHO/UNICEF/ICCIDD, 2007).

Iodine deficiency during pregnancy, infancy, or early childhood may cause derangement in the development of the brain and central nervous system. The most serious consequence of iodine deficiency during foetal life, infancy and childhood represents the endemic cretinism, which is associated with mental and

physical retardation, deaf-mutism, and various neurological abnormalities (EFSA, 2014, WHO, 1989).

Table A-1: Effects of iodine deficiency, by life stage (WHO/FAO, 2004)

Life stage	Effects
Fetus	Abortions
	Stillbirths
	Congenital anomalies
	Increased perinatal mortality
	increased infant mortality
	Neurological cretinism: mental deficiency, deaf mutism, spastic diplegia, and squint
	Myxedematous cretinism: mental deficiency, hypothyroidism and dwarfism
	Psychomotor defects
Neonate	Neonatal goitre
	Neonatal hypothyroidism
Child and adolescent	Goitre
	Juvenile hypothyroidism
	impaired mental function
	Retarded physical development
Adult	Goitre with its complications
	Hypothyroidism
	Impaired mental function
	Iodine induced hyperthyroidism

Iodine excess

Excess iodine intake in healthy adults in iodine-replete areas is difficult to define. Many people are regularly exposed to huge amounts of iodine without apparent adverse effects. Occasionally, this may have significant thyroid effects, but generally, they are tolerated without difficulty (WHO/FAO, 2014). People without evidence of underlying thyroid disease almost always remain euthyroid in the face of large amounts of excess iodine and escape the acute inhibitory effects of excess intrathyroidal iodide on the organification of iodide and on subsequent hormone synthesis (Braverman *et al.*, 1994 cited from WHO/FAO, 2014).

However, it has to be considered that chronic excessive iodine intake may accelerate the development of sub-clinical thyroid disorders to overt hypothyroidism or hyperthyroidism, increase the incidence of autoimmune thyroiditis and increase the risk of thyroid cancer (EFSA, 2014; Laurberg *et al.*, 1998; Teng *et al.*, 2006). Also chronic excessive iodine intake can lead to goitre, as has been observed following chronic excessive iodine intakes through water in China (EFSA, 2014; Zhao *et al.*, 2000).

Conclusion

Both dietary iodine intake levels below and above the acceptable range of daily intake for iodine are associated with an increased risk of disease in the population. Optimally, iodine intake should be kept within a relatively narrow range where iodine deficiency disorders are prevented, but should not exceed the upper limit. However, if iodine intake is carefully monitored for both iodine deficiency and excess, the benefits from a decrease of the substantial health risk of iodine deficiencies will clearly outweigh the relatively small risk associated with iodine excess (Laurberg, 2010; WHO/FAO, 2004, Zimmermann, 2009).

5. Dietary iodine reference values and recommendations

To meet the iodine requirements, the World Health Organisation (WHO) recommended dietary intakes for infants, children, school children, adults and pregnant and lactating women in 1996 (WHO 1996, WHO 2004). These recommendations were updated in 2007 for children < 2 years and pregnant and lactating women (WHO, 2007). Iodine intake values as recommended by WHO are given in Table A-2.

Table A-2: Recommended intakes for iodine (WHO, 1996; WHO/FAO, 2004; WHO, 2007)

Group	Recommended intake	
	µg/d	µg/kg bw/d
infants and children (0-6 years) [1,2]	90	6-30
school children (7-12 years) [1,2]	120	4
adults (> 12 years) [1,2]	150	2
pregnant and lactating women [1,2]	200	3.5
infants and children (<2 years) [3]	90	
pregnant and lactating women [3]	250	

[1] WHO, 1996; [2] WHO/FAO, 2004; [3] WHO, 2007; [A] probably safe upper limit; [B] level of iodine intake beyond which no added health benefit can be expected

Besides recommended intakes also upper limits (UL) were defined for iodine (Table A-3). The SCF deduced an UL of 600 µg/day for adults to be safe. The UL of 600 µg/day is considered to be also acceptable for pregnant and lactating women based on evidence of lack of adverse effects at exposures significantly in excess of this level. The ULs for children were derived by adjustment of the adult UL on the basis of body weight since there is no evidence of increased susceptibility in children (SCF, 2002).

Table A-3: Upper limits (UL) for iodine (SCF, 2002)

Group	Upper limit
	µg/d
infants and children (1-3 years)	200
children (4-6 years)	250
children (7-10 years)	300
children (11-14 years)	450
children (15-17 years)	500
adults	600
pregnant and lactating women	600

6. Natural occurrence and iodine supplementation

Iodine naturally occurs in food and water mainly as iodide. The concentration in water and foods is highly variable. Iodine absorption efficiency from water is influenced by the content and nature of humic substances in water (Andersen *et al*., 2008a).

The iodine content of foods is highly variable between food categories as well as within each category. The richest food sources are marine products (such as fish, shellfish, molluscs, seaweed), eggs and milk, as well as their derivatives and iodised salt. Iodine content of milk and eggs is influenced by feeding and hygienic practices (EFSA, 2005; Flachowsky et al., 2014). Besides its use as sanitizer, iodine is used as agricultural chemical (e.g. herbicides and fungicides). Furthermore, it is used as supplement in the human and animal nutrition to ensure a sufficient iodine intake and also represents a food additive, *e.g.* for dough conditioning and maturing agents (WHO, 1989).

According to Article 2 of Regulation (EC) No 178/2002 iodine can be defined as “food”. Furthermore, iodine may be used in the manufacture of food supplements in the form of sodium iodide, sodium iodate, potassium iodide and potassium iodate according to Directive 2002/46/EG. Likewise the U.S. Food and Drug Administration (FDA) considered iodine compounds as Generally Recognized As Safe (GRAS) and allows the use of cuprous iodide and potassium iodide as a nutrient supplement in table salt and the use of potassium iodate and calcium iodate as dough strengthener. In addition, in the European Union the iodine content of infant and follow-on formulae is regulated in the Commission Directive 2006/141/EC and Directive 1999/21/EC.

Iodine supplementation programs have been developed in many countries to prevent endemic goiter and the further consequences of iodine deficiency. According to WHO/UNICEF (2007b), iodine fortification of salt has been implemented in 40 European countries, being mandatory in 13 countries, voluntary in 16 and not regulated in the remaining countries.

In 2011, it was estimated that 44 % of the Europe population had insufficient iodine intakes (UI concentration < 100 µg/L). Furthermore, it was estimated that the prevalence of insufficient iodine intakes school children in European has been

reduced by about 30 % since 2003 but that insufficient iodine intakes remain a public health problem in 14 European countries (Andersson *et al*., 2012; Zimmermann *et al*., 2011 cited from EFSA 2014).

7. Iodine in non-target animals

Iodine is an essential trace element not only for humans but also for animals. The only known role of iodine in the metabolism in animals is its incorporation into the thyroid hormones (EFSA, 2005), as already described for humans. As known for humans, also animals have to cover their iodine requirements with their food uptake.

The iodine concentration in animal tissues varies widely and is lower for terrestrial animals (mean below 10 µg/kg) and higher for marine animals (mean up to 195 µg/kg) and can be as high as 1380 µg/kg in cod (Andersen *et al.*, 2002). As far as it is known, the role of iodine in humans and animals in the metabolism is its incorporation into the thyroid hormones (T4 and T3) and the precursor iodothyrosines (EFSA, 2005). T4 contains about 65 % of the body iodine (Mc Dowell, 2003, cited from EFSA 2005). Currently, its role in the regulation of gene expression of the pathways involved in immune response and oxidative stress in ruminants is discussed (Iannaccone, 2019).

The natural sources for animals (as also for humans) are iodides in food and water. The physiological utilizable form of iodine is iodide, which is adsorbed in the total gastrointestinal tract, but mainly in the small intestine, and in ruminants also in the rumen (Ketz, 1989, Underwood, 1977, cited from EFSA 2005). The absorption rate is about 80 - 92 % (Jongbloed *et al.*, 2002, cited from EFSA 2005, SCF 2002).

Iodine deficiency affects thyroid function in animals in the same way as in humans. In all age groups, iodine deficiency decreases the production of thyroid hormones and subsequently the general metabolism and oxidation processes. Also, it is known that iodine deficiency during the critical period of foetal and early postnatal brain development can result in severe thyroid failure and irreversible brain damage (EFSA, 2005).

The iodine requirements for various food producing livestock species were investigated by different scientific bodies. Iodine requirements of animals was estimated to be between 100 and 800 µg/kg feed (1200 µg/kg feed for fish), but even higher values are reported. Summarizing the requirement data it could be concluded that slowly growing ruminants, pigs and horses need about 200 (100 - 300) µg iodine/kg feed, whereas animals with a high metabolism (lactation, fast growing, sexual activity) may double the requirement up to 500 µg iodine/kg feed. As a result of intensive breeding progress, poultry needs between 400 and 500 µg iodine/kg feed. Thus, in most cases, iodine supplementation of practical diets for food producing and companion animals is necessary (EFSA, 2005) and, therefore, also for wild animals it can be concluded that iodine is a limiting micronutrient.

This is confirmed by the Amphibian Metamorphoses Assay (AMA) (OECD 231) where it is stated that iodide (I-) is essential for a proper test performance.

Concentrations of iodide in test water should range between 0.5 and 10 µg/L, because sufficient iodide needs to be available to the larvae through a combination of aqueous and dietary sources in order for the thyroid gland to synthesize the thyroid hormones. The maximum predicted environmental concentration surface water (PEC_{sw}) values for the intended uses of iodine (PT1, 3, 4 and 22, i.e. 1.55 µg/L, 0.61 µg/L, 0.55 µg/L and 0.186 µg/L, respectively), which are summarised in the Assessment Report of iodine (including PVP-iodine) (AR, 2013), demonstrate that the calculated environmental concentrations are lower than or within the recommended range for iodide in the AMA test guideline (OECD 231) that would be needed to assess T-mediated effects. Thus, neither adverse T related activity nor T mediated adversity is attributable to iodine in non-target organisms. Furthermore it should be pointed out that the natural background level for iodine in freshwater and marine water is 0.5 – 20 µg/L and 45 - 60 µg/L, respectively (AR, 2013).

Rapporteur's comments

To support normal metamorphosis, sufficient iodide should be made available to the larvae in tests with amphibians. Currently, there are no empirically derived guidelines for minimal iodide concentrations. However, iodide availability may affect the responsiveness of the thyroid system to thyroid active agents and is known to modulate the basal activity of the thyroid gland (OECD TG 231, 2009, and OECD TG 241, 2015).

However, the argument made by the applicant that calculated environmental concentrations are lower than or within the recommended range for iodide in the AMA test guideline (OECD 231) is not valid in the context of the hazard-based assessment of endocrine disruption. It should also be borne in mind that the levels recommended in the guideline are specific for the test species and conditions of the test.

The applicant's literature search did not retrieve any relevant information on potential endocrine disrupting mechanisms or effects related to the EAS modalities with respect to non-target organisms. Neither do the REACH registration dossier or the biocidal assessment report (CAR; 2013) contain any relevant information in that respect. The rapporteur is not aware of any published information related to effects related to the EAS modalities from iodine, PVP iodine or any other iodine containing compound in non-mammalian species.

Relevant information on potential endocrine effects of iodine in other organisms than humans or mammals is scarce and appears to be limited to investigations on amphibian metamorphosis, done already 100 years ago. Then, scientists established that iodine and various iodine-containing compounds affect the metamorphosis in amphibia if administered either to the surrounding water or via diet (among others: Swingle, 1919a, 1919b; Spaul, 1924; Abderhalden and Hartmann, 1928). Thereafter, it appears that very few attempts were made to further investigate the effects of iodine compounds on non-mammalian species. Only recently, the 100-year-old findings were confirmed by Krishnapriya et al. (2014) and Olker et al. (2018). The results obtained by Krishnapriya et al. confirm that exposure to iodine via diet leads to adverse outcome. The accelerated metamorphosis observed, showing a clear dose-related response, is a

specific T-mediated endocrine effect. Advanced development is only known to occur through effects which are thyroid hormone related, as stated in the OECD Guideline for The Amphibian Metamorphosis Assay (OECD 231; 2009). The results by Olker et al. support the understanding that mechanisms in amphibians and mammals are similar, and that amphibia efficiently take up iodine from the aquatic medium.

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SPAUL EA. Accelerated Metamorphosis of Frog Tadpoles by Injections of Extract of Anterior Lobe Pituitary Gland and the Administration of Iodine. Journal of Experimental Biology 1924;1:313-21.

Swingle WW. IODINE AND THE THYROID : IV. QUANTITATIVE EXPERIMENTS ON IODINE FEEDING AND METAMORPHOSIS. The Journal of general physiology 1919;2:161-71.

Swingle W. IODINE AND THE THYROID: III. The Specific Action of Iodine in Accelerating Amphibian Metamorphosis. The Journal of General Physiology 1919;1:593.

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OECD. 2009. Test No. 231: Amphibian Metamorphosis Assay. OECD Guidelines for the Testing of Chemicals. OECD Publishing. Paris.

OECD. 2015. Test No. 241: The Larval Amphibian Growth and Development Assay (LAGDA). OECD Publishing. Paris.

Olker JH, Haselman JT, Kosian PA, Donnay KG, Korte JJ, Blanksma C, et al. Evaluating Iodide Recycling Inhibition as a Novel Molecular Initiating Event for Thyroid Axis Disruption in Amphibians. Toxicological sciences: an official journal of the Society of Toxicology 2018;166:318-31.

Conclusion

The fact that (i) iodine is an essential dietary trace element with a defined key role in the biosynthesis of the thyroid hormones T4 and T3 (T modality of the hormone system) and the point that (ii) dietary intake and even food supplementation are recommended to ensure a sufficient iodine intake show, that an entirely hazard based ED assessment as outlined in the “Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009” (ED GD) especially with focus on T modality is not meaningful for iodine and possibly beyond the scope of the Commission Delegated Regulation (EU) 2017/2100. If the ED properties of iodine and other essential micronutrients would be assessed from a scientific perspective, the assessment should consider not only the hazard, but also take into account the potency and exposure.

However, human health risk assessments for the approval of iodine as biocidal active substance and for the authorisation of iodine-based biocidal products

already consider the upper limit (UL) of iodine which takes into account the endocrine effects of iodine and represents a conservative basis for the risk assessment. For the environmental risk assessment the natural background levels for iodine were taken into account and it was assessed whether the calculated environmental concentrations based on the biocidal uses are within the natural background concentrations of iodine. Therefore, regulating iodine as endocrine disrupting chemical (EDC) would not provide any additional safety.

Rapporteur's comments

(Details of the rapporteur's assessment of the T-modality are available in the BPC Opinion on the Art. 15 request on (PVP-)iodine.)

According to the Commission Delegated Regulation (EU) No 2017/2100, a substance is identified as having endocrine-disrupting properties with respect to humans if it meets the following three criteria

(a) it shows an adverse effect in an intact organism or its progeny;

(b) it has an endocrine mode of action, i.e., it alters the function(s) of the endocrine system; and

(c) the adverse effect is a consequence of the endocrine mode of action.

Iodine in excess of physiological needs meets all the above three criteria – it shows adverse effects (thyroid disorders) in humans; it has an endocrine mode of action (disruption of thyroid hormones metabolism and hypothalamic-pituitary-thyroid axis); and the adverse effects are a consequence of the endocrine mode of action. Therefore, the biocidal active substances, iodine and PVP-iodine, are identified as having endocrine-disrupting properties with respect to humans.

Since iodine and PVP-iodine meet the criteria for endocrine disruption with respect to humans it can be concluded that the substances are endocrine disruptors also in non-target organisms. Furthermore, it is highly plausible that external administration of iodine via water or diet to amphibia interferes with endocrine mechanisms related to the thyroid and in excess of physiological needs subsequently leads to adverse effects in intact organisms, namely accelerated metamorphosis. This further supports the conclusions that the criteria for endocrine disruption in non-target species are met.

Note: The applicant should have assessed the T-modality according to the ECHA/EFSA ED guidance document based on a Lines of Evidence table including the data extracted from the CAR on (PVP-)iodine and from the systematic literature search also on relevant substances such as iodide salts (e.g., sodium iodide, potassium iodide).

Part B: Presentation of the assessment of estrogen, androgen and steroidogenic (EAS) mediated endocrine disrupting (ED) properties of iodine

Summary

The assessment of estrogen, androgen and steroidogenic (EAS) mediated endocrine disrupting (ED) properties of iodine was performed according to the "Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009" (ED GD; ECHA/EFSA, 2018) and further regulatory relevant documents in the assessment of ED chemicals (EDC). The focus of the present assessment was set on the scenario of excessive iodine intake.

Thyroid (T) mediated ED properties of iodine were not assessed in this part of the document. Referring to the T-modality, it is well-established that the micronutrient iodine plays a key role in thyroid-hormone biosynthesis. Accordingly iodine shows T-related activity (organification – incorporation of iodine into thyroglobulin) and T-related adversity (*e.g.* hypothyroidism) which are linked by an endocrine mode of action with human relevance; please refer to Part A of the document.

For the assessment of potential ED properties regarding EAS modalities of the active substance iodine, the Excel spreadsheet of Appendix E of the ED GD (ECHA/EFSA, 2018) was completed with relevant data and a literature search was performed and evaluated for toxicological endpoints. In addition, information from other sources including but not confined to open source databases was gathered. All available information relevant for ED-assessment is presented in the present document (data gathering). Based on the summarised information lines of evidence were established.

According to the ED GD (ECHA/EFSA, 2018) ED-related adverse effects as well as ED-related activity should be investigated and a link between both needs to be proven to consider a substance as endocrine disrupting chemical (EDC).

The analysis of the available toxicological information revealed (*in silico*, *in vitro* and *in vivo* mechanistic data), no relevant indications for EAS-related activity of iodine. There was no significant EAS-mediated adversity in connection with excessive intake of iodine when considering the available weight of evidence (WoE). Some indications for EAS-mediated adversity were seen in the toxicological dataset. However, the observed effects (changes in survival of the offspring and sperm morphology) are considered to be a consequence of the well-established T-related activity and T-mediated adversity of iodine and/or to other not EAS-mediated mechanisms (*e.g.* reactive oxygen species (ROS) formation). Of note, it is difficult to separate the T-mediated adverse effects of iodine from possible EAS-mediated adverse effects.

Not all parameters required for EAS-related activity according to the ED GD (ECHA/EFSA, 2018) were reported *in sensu stricto*. However the available data set regarding EAS-related activity was considered to be sufficient and adequate for a reliable and scientifically sound assessment of EAS-related activity of iodine.

All available data were assessed regarding the EAS-mediated adversity of iodine. However, based on the ED GD (ECHA/EFSA, 2018) EAS-mediated adversity cannot be considered as sufficiently investigated in the case of iodine. Nevertheless , this does not represent a relevant data gap, since the EAS–related activity can be considered as sufficiently investigated based on the available data set (*in silico, in vitro* and *in vivo* mechanistic) and revealed no relevant indications for EAS-related activity of iodine.

Based on the data gathering and evaluation it was concluded that iodine possesses no ED properties regarding the EAS modalities due to the lack of relevant indications for EAS-related activity and no strong indication of the EAS-mediated adversity (Scenario 2a (ii) of the ED GD). Therefore, no mode of action (MoA) analysis needed to be conducted.

In a WoE approach it was considered that no EAS-related activity is attributable to iodine. Additionally no toxicologically significant EAS-mediated adversity was observed in the available data set. Therefore, the "ED criteria regarding EAS modalities are not met" for iodine based on the present assessment.

1. Gather all relevant information

For the toxicological data gathering, all available information was summarised using the data matrix template provided in Appendix E of the ED GD (ECHA/EFSA, 2018), where applicable, and key excerpts are presented in the following tables.

Rapporteur's comments

- The applicant's literature search is confined to iodine and PVP-iodine. It should also cover iodide salts (for e.g., sodium iodide and potassium iodide) as these are relevant to assess the toxicity of iodine.
- A relevant study available in the REACH registration dossier for iodine is missing in the applicant's assessment. It's an OECD 422 study (combined repeated dose/reproductive toxicity screening study) with iodine. The study is GLP compliant but with deviations (not listed in the summary) from the text guideline. No treatment-related adverse reproductive effects were reported in this study. The very limited study summary is accessible via the ECHA dissemination webpage https://echa.europa.eu/registration-dossier/-/registered-dossier/15294/7/9/2, last accessed in March 2022.

1.1. Scientific data generated from *in vivo* studies

The most reliable and relevant data to assess possible ED properties of iodine besides human epidemiological data are *in vivo* data from available internationally standardised level 4 and level 5 test methods for mammals, categorized according to the OECD Conceptual Framework (CF) (OECD, 2018) for testing and assessment of potential EDC. All relevant *in vivo* studies based on information requirements for biocidal active substances were assessed and the ED relevant information

gathered and analysed in a data summary/data matrix fully compliant with Appendix E of the ED GD (ECHA/EFSA, 2018).

The Competent Authority Report (CAR; Schweden, 2013) and Assessment Report of iodine (including PVP-iodine) for PT 1, 3, 4 and 22 (AR, 2013) comprises studies on iodine and PVP iodine since PVP iodine was not considered as a separate active substance but rather as a iodine complex to accomplish a controlled release of iodine. Therefore, in addition to the available data set on iodine (CAS no. 7553-56-2) also available data on PVP-iodine (CAS-No 25655-41-8) was used in this assessment.

Key studies summarized in the Table B-1 had already been assessed during the active substance evaluation under the BPR. Information on the results and methods from Doc IIA/Doc IIIA of the CAR for PT 1, 3, 4 and 22 (Sweden, 2013) and from the study reports were used for the present assessment. Specific information regarding endocrine properties which is not included in the summaries of the CAR is provided in addition in the Appendix E and in the lines of evidence tables. The data derived from the literature search were included in Table B-1 as well.

Table B-1: Summary of the *in vivo* studies included in the present assessment

Study Type Reliability, Key / supportive study	**Species**	**Test substance Purity**	**Dose levels or concentrations**	**Reference**	**Study ID (App. E)**
Combined teratogenicity/reproduction toxicity study Reliability 2 Key study	Rat	Potassium iodide -	0, 2500 ppm (exp. 1,3,4) 0, 500, 1000, 1500, 2000 (exp. 2)	Ammerman *et al.* (1964) Doc. No. 592-011	**1**
Prenatal developmental toxicity study Reliability 2 Key study	Rabbit	PVP-iodine -	0, 16, 35, 75 mg/kg bw/day	Siegemund *et al.* (1987) Doc. No. 592-066;	**2**
Subchronic oral toxicity study Reliability 1 Key study	Rat	Iodine -	0,1, 3, 10, 100 mg/L; equivalent to 0.14, 0.42, 1.4, 14 mg/kg bw/day	Sherer *et al.* (1991) Doc. No. 592-027	**3**
Prenatal developmental toxicity study Reliability 2 Key study (as part of the "toxic effects on livestock and pets" study)	Rat, rabbit, hamster	Potassium iodide or Sodium iodide -	0, 250, 500 and 1000 ppm 0 and 2500 ppm	Arrington *et al.* (1965) Doc No 592-012	**10**
In vivo mechanistic study Reliability 2 supportive study	Mare	PVP-iodine -	intrauterine infusions on days 0 and 2 using 1000 mL of a 1% povidone-iodine solution	Kalpokas *et al.* (2010) Outcome of literature search	**14**
Adult male assay Reliability 2 supportive study	Rat	Potassium iodide -	0, 0.7, 3.5 mg/kg bw/day	Chandra *et al.* (2017) Outcome of literature search	**15**

-: not indicated, App.: appendix, ID: identification

1.2. Relevant human health data and epidemiological data

Based on the ED GD (ECHA and EFSA, 2018) "*any available epidemiological studies should be considered as supportive evidence for the evaluation of whether an ED is likely to have adverse effects for humans. However, they cannot be used to override or dismiss evidence of adversity found in laboratory studies*".

No relevant human health data and epidemiological data on EAS-mediated adversity or activity for excessive iodine are available.

1.3. Scientific data selected applying systematic review methodology (Non-guideline studies)

A literature search for the active substance iodine on potential EAS-mediated ED properties of the active substance iodine was performed as requested by the ED GD (ECHA/EFSA, 2018).

The literature search (Reisinger, 2020) was conducted in accordance to the provisions of the EFSA Guidance "Submission of scientific peer-reviewed open literature for the approval of pesticide active substances under Regulation (EC) 1107/2009".

The objective of the literature search was the assessment of scientific peer-reviewed open literature dealing on potential EAS-mediated ED properties of the active substance iodine.

This report summarises the search and selection process of the literature search performed.

Literature was searched accessing the databases: AGRICOLA, BIOSIS, CABA, EMBASE, ESBIOBASE, HCAPLUS, MEDLINE, PQSCITECH, TOXCENTER via the service provider STN-International.

In total, 381 records were retrieved from bibliographic databases and were screened by expert reviewers for relevance. Based on the evaluation of the summary records (titles/abstracts) 358 publications were assessed as obviously not relevant for the assessment of potential endocrine disruptive properties (EAS) of the active substance iodine.

23 full-text documents were assessed in detail. Four of these publications provided relevant information on the potential endocrine disruptive properties (EAS) of the active substance iodine.

One full-text document (Pearce, 2018) represents a review document and was considered as background information. Three full-text documents (Kalpokas *et al.,* 2010, Chandra *et al.,* 2017 and Stoddard *et al*., 2008) were included in the data matrix template provided in Appendix E of the ED GD (ECHA/EFSA, 2018).

1.4. Data *of in vitro* methods, from databases or obtained with *in silico* tools with information relevant to EDC identification.

The available open access databases listed in Appendix D.1. of the ED GD (ECHA/EFSA, 2018) were searched for information on a putative ED potential of iodine. No information was obtained which would be indicative of endocrine activity or endocrine-mediated adversity of iodine.

Furthermore, Q(SAR) tools were used in accordance to Appendix D.2. of the ED GD (ECHA/EFSA, 2018) to provide predictions concerning putative endocrine activity of iodine. No data on endocrine endpoints were found in the databases EDKB, EADB, NURSA, AOP Knowledge Base, COSMOS DB and the (Q)SAR Data Bank. The Toxicity Reference Database (ToxRefDB) identified one relevant developmental toxicity study (Arrington, LR *et al*. 1965, Doc No 592-012). This study was already included in the iodine CAR (Sweden, 2013) (Section A6.13/01-07) and is considered in the present ED assessment.

OECD (Q)SAR Toolbox (OECD, ECHA)

In silico prediction using OECD (Q)SAR Toolbox (Version 4.4., 2020) revealed no estrogen binding activity for iodine (CAS no.7553-56-2). Iodine was predicted to be non-binder for the estrogen receptor because of the absence of a cyclic structure. In addition, using the rainbow trout estrogen receptor(s) (rtER)- Expert System model, integrated in the OECD (Q)SAR Toolbox no alerts were found for iodine.

Table B-2: Outcome of the in silico prediction using OECD (Q)SAR Toolbox (OECD, ECHA)

CAS Number	7553-56-2
Chemical name(s)	**I-I; Iodine**
SMILES	II
Molecular formula	I_2
Predefined substance type	Mono constituent
Additional Ids	EC Number:2314424
CAS-SMILES relation	High
Estrogen Receptor Binding	Non binder, non cyclic structure
rtER Expert System - USEPA	No alert found

Endocrine Disruptome

The prediction for the endocrine activities of iodine was performed using the respective SMILES Code: II. Low binding probabilities (“green class”) were predicted for androgen receptor agonist and antagonist (AR, AR an), estrogen receptor alpha and beta (ERα and ERβ) and glucocorticoid receptor (GR and GRα) (Endocrine Disruptome, 2020). Taking together no EAS-related activity was predicted for iodine using “Endocrine disruptome”.

Figure B-1: **Docking prediction for iodine by Endocrine Disruptome**. (MW = Molecular weight. HBA = Hydrogen bond acceptors. HBD = Hydrogen bond donors. LogP = Logarithm of partition coefficient. TPSA = Topological polar surface area. Rot = Rotatable bonds).

EDSP21 Dashboard (US-EPA)

To obtain *in vitro* mechanistic information for potential EAS-related activity data of iodine the U.S. EPA CompTox Chemistry Dashboard and the EDSP21 Dashboard were evaluated and the outcome included in Appendix E.

According to the EDSP21 Dashboard, iodine was found non-active in the 12 tested assays for estrogen receptor (ER) related activity, in the 10 tested assays for androgen receptor (AR) related activity and 1 of 2 tested assays for steroidogenesis-related activity.

Iodine was found to be active in 1 steroidogenesis-related assay (TOX21_Aromatase_Inhibition). However, the AC50 of 40.89 µL was clearly above

the cytotoxicity limit (6.40 µL) for this assay. Moreover the reported assay was flagged as with efficacy values less than 50%. Therefore the biological significance of the observed activity in this assay is highly questionable. The outcomes of individual assays were reported in Appendix E.

Conclusion on data of *in vitro* methods, from databases or obtained with *in silico* tools

No toxicologically relevant concern is given indicating that iodine might comprise EAS-related activity.

1.5. Relevance and reliability

In accordance with the ED GD (ECHA/EFSA, 2018), relevance and reliability were assessed for the gathered information and considered adequate. Non-relevant and/or non-reliable data have not been included in the present analysis. The assessments of relevance and reliability were integrated in Appendix E.

1.6. Summary of data gathering according to the Guidance Document for the identification of endocrine disruptors (ECHA/EFSA, 2018)

As a result of the data gathering according to Appendix E, the effects as listed in Table B-3 –B-6 were observed for the assessed data.

Table B-3: Summary table of extracted data on *in vitro* and *in vivo* mechanistic information for effects of iodine according to data gathering with the ED GD (ECHA/EFSA, 2018), Appendix E

	Effect	No Effect
Androgen receptor (*in vitro*)	0	3
CYP19 (*in vitro*)	1	0
Estrogen receptor (ER) (*in vitro*)	0	2
Estrogen related receptor (*in vitro*)	1	0
Nuclear receptor (ER) (*in vivo*)	0	1
Nuclear receptor progesterone receptor (*in vivo*)	1	0

Table B-4: Summary table of extracted data on EAS-mediated parameters for effects of iodine according to data gathering with the ED GD (ECHA/EFSA, 2018), Appendix E

	Effect	No Effect
Mammary gland histopathology (female)	1	1
Seminal vesicles histopathology	1	0
Sperm morphology	1	0
Testis weight	0	2

Table B-5: Summary table of extracted data on parameters which are sensitive to, but not diagnostic of, EAS for effects of iodine according to data gathering with the ED GD (ECHA/EFSA, 2018), Appendix E

	Effect	No Effect
Brain weight	0	2
Fertility (mammals)	0	1
Gestation length	0	3
Litter size	0	4
Litter viability	5	0
Litter/pup weight	4	2
Number of implantations, corpora lutea	0	2
Number of live births	0	1
Numbers of embryonic or foetal deaths and viable foetuses	0	1
Post implantation loss	0	1
Pre implantation loss	0	1
Presence of anomalies (external, visceral, skeletal)	0	1
Pup development	0	1
Pup survival index	0	1
Reproduction	0	2

Table B-6: **Summary table of target organ and systemic toxicity for effects of iodine according to data gathering with the ED GD (ECHA/EFSA, 2018), Appendix E**

	Effect	No Effect
Kidney weight	1	2
Liver weight	1	2
Body weight	5	1
Food consumption	4	0
Mortality	0	2

2. ED assessment for humans

2.1. ED assessment for EAS-modalities

2.1.1. Have EAS-mediated parameters been sufficiently investigated?

With regard to the available toxicology studies, EAS-mediated adversity was not sufficiently investigated according to the ED GD (ECHA/EFSA, 2018). The EAS-related endocrine activity was *in sensu stricto* not sufficiently investigated according to the ED GD (ECHA/EFSA, 2018). However, taking into account the available data set regarding EAS-related activity (*in silico*, *in vitro* and *in vivo* mechanistic) the EAS-related activity was considered to be sufficient and adequate for a reliable and scientifically sound assessment.

	Sufficiently investigated
EAS-mediated parameters	**Yes** for activity (based on availability of the following data) *In silico* prediction *In vitro* ToxCast data *In vitro* estrogen assay (Stoddard *et al.,* 2008) *In vivo* mechanistic study in mares (Kalpokas *et al.,* 2010) **No** for adversity (based on availability of the following studies) Combined teratogenicity/reprotoxicity study in rat (Ammerman *et al*. (1964)) Prenatal developmental toxicity study in rabbit (Siegemund *et al*., 1987) Subchronic oral toxicity test in rat (Sherer *et al.,* 1991) Prenatal developmental toxicity study in rat, rabbit and hamster (Arrington *et al.,* 1965) Adult male assay in mice (Chandra *et al*., 2017)

2.1.2. Lines of evidence for adverse effects and endocrine activity related to EAS-modalities

According to the ED GD (ECHA/EFSA, 2018) "*the assembling of lines of evidence should take into consideration all the available evidence (positive and negative) that have been evaluated as relevant and reliable*" during the data gathering. These parameters were assessed to determine "*whether and how they contribute to the lines of evidence for adversity and/or endocrine activity".*

The integrated lines of evidence for EAS-related activity and EAS-mediated adversity are reported in Table B-7. The data comprise *in silico* prediction, *in vitro* mechanistic studies, *in vivo* mechanistic studies, data on organ weight, histopathological evaluations and reproduction and developmental parameters.

Table B-7: Assessment of the integrated lines of evidence related to EAS-modalities

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
Integrated line of evidence for endocrine activity	*In silico* prediction	(Q)SAR prediction: Endocrine Disruptome	n.a.	n.a.	n.a.	n.a.	No effect	Low binding probabilities ("green class") were predicted	Docking predictions revealed no indications for ER, AR and GR binding probabilities	**Sufficient** Overall, no convincing evidence for E, A, S-related activity	E,A,S
		OECD (Q)SAR Toolbox (Version 4.4., 2020): Estrogen receptor	n.a.	n.a.	n.a.	n.a.	n.a.	Predicted to be non - binder for ER because of the absence of a cyclic structure	Supporting evidence for non-receptor binding (ER)		E
		OECD (Q)SAR Toolbox (Version 4.4., 2020) (rtER-Expert System) Estrogen Receptor	n.a.	n.a.	n.a.	n.a.	n.a.	No alert found			E

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
Integrated line of evidence for endocrine activity	*In vitro* mechanistic	Androgen receptor	human	24h	Uptake from the medium	LC50=0 µM	No effect	Non active as agonist	ToxCast *in vitro* AR. No agonistic and antagonistic binding activity.		A
		Androgen receptor	human	24h	Uptake from the medium	LC50=0 µM	No effect	Non active as antagonist			
		Androgen receptor	human	24h	Uptake from the medium	LC50=0 µM	No effect	Non active as agonist			
		CYP19 (ToxCast Aromatase inhibition)	human	24h	Uptake from the medium	LC50 = 40.98 µM	Change	AC50 clearly above the cytotoxicity limit (6.40 µM). Assay was flagged as with efficacy values less than 50%.	Very weak evidence for S (receptor binding)		S
		Estrogen receptor	human	24h	Uptake from the medium	LC50=0 µM	No effect	Non active as agonist	ToxCast *in vitro* ER. No agonistic and antagonistic binding activity.		E
		Estrogen receptor	human	24h	Uptake from the medium	LC50=0 µM	No effect	Non active as antagonist			E
		Estrogen related receptor	human	48h	Uptake from the medium	1mM Iodine/Iodide	Change	Increased mRNA levels of	Indication of possible indirect		E

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
								several genes involved in estrogen metabolism	mechanism on estrogen metabolism.		
	In vivo mechanistic	Nuclear receptor ER	mare	-	Direct		No effect	Does not affect the expression of ERa receptor	No evidence (for ER activity		E
		Nuclear receptor (progesterone receptor)	mare		Direct		Change	The number of PR positive cells (PPC) on day 6 was reduced. However the infusion process *per se* may have promoted the disturbance in the PR expression that could not be	Some indication of S related activity, however without appropriate control the mechanism is questionable.		S

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
								tested since the control mares were not infused with saline.			
		Progesterone	mare		Direct		Decrease	Specific data points show tendency, without statistical significance.			S
Integrated line of evidence for EAS-mediated adversity		Mammary gland histopathology (female)	Rat	32 "interrupted"; 42"continuous"	Days	2500 ppm	No effect	Histological examination of mammary tissue from females fed iodine revealed epithelial development comparable to	Effects on milk production due to the thyroid (T) related activity. Not a direct evidence of EAS adversity.	Some evidence of possible EAS-mediated adversity. However the influence of other factors(T related activity and adversity)	E, A, S

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
								control rats.		, ROS building)	
	EAS-mediated		Rat	32 "interrupted"; 42"continuous"	Days	2500 ppm	Change	Milk secretion was absent or markedly diminished (probably due to the effects in thyroid gland), but not the development of mammary tissue.		is considered relevant	E, A, S
		Seminal vesicles histopathology	Rat	60 days	Oral	7 mg/kg bw/day (100 X excess of iodine)	Change	Marked degenerative changes in the surface morphology of seminiferous tubules with shrinkage and appreciabl	Evidence of EAS adversity, however by indirect (non-endocrine) mechanism (generation of ROS)		E,A,S

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
								e decrease in the tubular areas.			
		Sperm morphology	Rat	60 days	Oral	7 mg/kg bw/day (100 X excess of iodine)	Change	Exposures to 7 mg/kg bw/day (60D), 35mg/kg bw/day (30D and 60 D) resulted in a significantly high number of FITC-PSA positive ceils indicating disintegration of acrosomal status.			E, A, S
		Testis weight	Rat	100	Days	Oral	No effect	Nominal weight	No effects on testis weight		E,A,S
		Testis weight	Rat	100	Days	Oral	No effect	Relative to the body weight			E,A,S
		Litter size	Rat	68 Days	Oral	-	No effect				N

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
	Sensitive to, but not diagnostic of, EAS		Rat	65 -75 Days	Oral	-	No effect		No strong evidence for endocrine adversity		
			Rabbit	12 Days	Direct (percutaneous injection)	-	Decrease	The effect was observed at the doses causing slight systemic toxic effects in dams			
			Hamster	12 Days	Oral	-	No effect				
		Litter viability	Rat	32 "interrupted"; 42"continuous" Days	Oral	2500	Decrease	Decreased number of surviving young is due to reduced or absent lactation (mammary secretion) and not due to embryo / developmental toxicity.	Overall decreases of surviving of offspring: secondary effect due to the reduced milk production		
			Rat	68 Days	Oral	2500	Decrease				
			Rat	65 -75 Days	Oral	2500	Decrease				
			Rat	0-8 Days	Oral	2500 pm	Decrease				
			Rabbit	-	Oral	250 ppm	Decrease				

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
		Litter/pup weight	Rat	32 "interrupted"; 42"continuous" Days	Oral	2500 pm	Decrease		No sufficient evidence for endocrine adversity		
			Rat	65 -75 days	Oral		No effect				
			Rabbit	12 Days	Direct (percutaneous injection)	-	No effect				
			Rabbit	-	Oral	500 ppm	Decrease				
			Rat	710 day	Oral	2500 ppm	Decrease				
			Hamster	12 Days	Oral	-	No effect				
		Pup survival index	Hamster	12 Days	Oral	-	No effect		No evidence for endocrine adversity		
		Number of ovarian follicles	Rat	35 Days	Oral	-	No effect				
		Number of implantations, corpora lutea	Rat	32 "interrupted"; 42"continuous"	Oral	-	No effect				
			Rabbit	12 Days	Direct	-	No effect				
		Numbers of embryonic or foetal	Rabbit	12 Days	Direct	-	No effect				

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
		deaths and viable foetuses									
		Presence of anomalies (external, visceral, skeletal	Rabbit	12 Days	Direct (percutaneous injection)	-	No effect				
		Post implantation loss	Rabbit	12 Days	Direct (percutaneous injection)	-	No effect				
		Pre implantation loss	Rabbit	12 Days	Direct (percutaneous injection)	-	No effect				
		Fertility (mammals)	Rabbit	12 Days	Direct (percutaneous injection)	-	No effect				
		Pup development	Rabbit		Oral	-	No effect				
		Gestation length	Rabbit		Oral	-	No effect				
			Rat	65-75	Oral		No effect				
			Hamster	12 Days	Oral	-	No effect				
		Number of live births	Hamster	12 Days	Oral		No effect				
		Reproduction	Hamster	12 Days	Oral	-	No effect				
			Rat	710 days	Oral		No effect				

	Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence	Assessment on the integrated line of evidence	Modality
		Brain weight	Rat	100 Days	Oral		No effect				

- = negative evidence.. Abs. = absolute. an = antagonist. AR = androgen receptor. bw = body weight. EAS = estrogen, androgen, steroidogenic. ER = estrogen receptor. F = Females. LC50 = half-maximal inhibitory concentration. M = Males. N.a. = not applicable/not available. NR = nuclear receptor; D = day, ROS = reactive oxygen species, QSAR= Quantitative structure–activity relationship, N = Endpoints potentially sensitive to, but not diagnostic of, EATS modalities

Table B-8: Assessment of the integrated lines of evidence for target organ and general toxicity

Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence
Target organ toxicity	Liver weight	Rat	100 days	oral	-	No effect	M	The effects were observed only in F and w/o correction for the body weight
	Liver weight	Rat	100 days	oral	14	Increase	F	
	Kidney weight	Rat	100 days	oral	-	No effect	M	
	Kidney weight	Rat	100 days	oral	14	Increase	F	
Systemic toxicity	Body weight	Rat	32 "interrupted"; 42"continuous days"	Oral	2500 ppm	Decrease		Dose-related effects on food consumption and body weight observed indicative for systemic toxicity
		Rat	68 days	Oral	2500 ppm	Decrease		
		Rat	65-67 days	Oral	500 ppm	Decrease		
		Rat	100 days	oral	-	No effect		
		Rabbit	12 days	Direct	75	Decrease	The dose dependent ↓ of relative bw gain was observed upon the treatment. However the statistically significant ↓ of bw gain was observed only at 75 mg/kg bw/day.	
		Hamster	100 days	oral	2500 ppm	Decrease	F1 (at D21) ↓ due to the food intake by the pregnant	

Grouping	Line(s) of evidence	Species	Duration of exposure	Route of administration	Effect dose [mg/kg bw/d] or as indicated	Effect direction	Observed effect (positive and negative)	Assessment of each line of evidence
							and lactating hamsters.	
	Food consumption	Rat	68	Oral	2500	Decrease	Overall food consumption ↓	
		Rat	65-67 days	Oral	500 ppm	Decrease		
		Rat	710 days	Oral	1000 ppm	Decrease		
		Hamster	12 days	oral	2500 ppm	Decrease		
	Clinical signs	Rabbit	12 days	Direct	Direct	No effect		
	Mortality	Rabbit	12 Days	Direct	-	No effect		
		Rat	100 days	oral		No effect		

bw = body weight. F = Females.. N.a. = not applicable/not available.↓ = Decrease. ↑ = Increase. F1 = first filial generation. w/o =with out

2.1.2.1. Assessment of the integrated lines of evidence and weight of evidence for EAS-mediated adversity and endocrine activity

Table B-9: WoE for EAS-mediated adversity

• Some weak indications for EAS-mediated adversity were seen in the toxicological dataset. However the observed effects can be assigned to the well-established T related activity of iodine and/or to the other not EAS-mediated mechanisms (*e.g.* ROS formation).
• The survival of the offspring of dams that received iodine was clearly decreased in the combined teratogenicity/reproduction toxicity study conducted on rats (Study ID 1). This effect ware considered as secondary effect to a lactation defect in the dams and is not related to teratogenic or reprotoxic properties of iodine. The effect was dose dependent and associated with low incidences of mortality at 500 ppm and higher incidences of mortality at 1000 ppm and 2500 ppm.
• Mammary secretion was affected at ≥500 ppm of iodine. However the histological examination of mammary tissue from females fed iodine revealed epithelial development comparable to control rats in this study (Study ID 1).
• No teratogenic effects were observed even with doses causing slight systemic toxic effects in dams in the teratogenicity study conducted on rabbits (Study ID 2). Compared with the controls, the average foetus weights were statistically significantly decreased ($P < 0.05$) but the effects were not dose-dependent.
• Pup survival index and litter/pup weight was not affected by treatment in hamster (Study ID 11).
• No effects on testis weight were observed in the study conducted on rats (Study ID 3).
• Sperm morphology (disintegration of acrosomal status) and morphology of seminiferous tubules were affected by excess iodine treatment in the study conducted on rats (Study ID 15). However the possible mechanism of observed effects is elevated formation of reactive oxygen species (ROS) due to the oxidizing properties of iodine and not a direct endocrine mechanism.
• No other developmental or reproductive parameters investigated in the studies conducted on rats, rabbits and hamster (number of ovarian follicles, number of implantations, corpora lutea, numbers of embryonic or foetal deaths and viable foetuses, pre and post- implantation loss, gestation length, pup development and *etc.)* were affected by treatment with iodine.
• No significant effects on target organ toxicity (liver and kidney) were observed in the study conducted on rats. (Study ID 3).

EAS-mediated adversity was considered to be not sufficiently investigated with regard to the toxicology studies.

Rapporteur's comments

According to the ECHA/EFSA ED Guidance document (2018), the EAS-mediated adversity is not sufficiently investigated for iodine since a two-generation (OECD TG 416; ver. 2001) or an extended-one generation (OECD TG 443) study is lacking.

There is human data on iodine-induced thyroid disorders leading to secondary adverse reproductive effects (WHO, 2020). However, the available animal toxicological dataset is not sufficient to conclude that the adverse reproductive effects of iodine are solely due to T-modality or non-ED mechanisms (e.g., ROS formation). According to the Guidance document, the A- and S-modalities are not sufficiently investigated for iodine..

WHO (World Health Organisation). 2020. Iodine in drinking-water. Background document for development of WHO *Guidelines for drinking-water quality*. WHO/HEP/ECH/WSH/2020.5.

Table B-10: WoE for EAS-related activity

- *In vitro* mechanistic and *in silico* predictions: No convincing evidence for E, A, S related activity
  - Endocrine Disruptome *in silico* data showed no probabilities for receptor binding activity for ER, AR and GR.
  - According to profiling with the OECD QSAR Toolbox, iodine was predicted as non-binder for ER because of the absence of a cyclic structure. No alerts were found using rtER- Expert System for ER activity.
  - ToxCast *in vitro* AR assays: non-active as agonist and antagonist.
  - ToxCast *in vitro* ER assays: non-active as agonist and antagonist.
  - *In vitro* assay using MCF-7 breast cancer cell line: increased mRNA levels of several genes involved in estrogen metabolism. Indirect effects, no measurements of receptor activity.
  - ToxCast *in vitro* SR assay (CYP19): LC50 = 40.98 µM; The AC50 was above the cytotoxicity limit (6.40 µM) for this assay. Moreover the reported assay was flagged as with efficacy values less than 50%.
- *In vivo* mechanistic: no convincing evidence for E, or S related activity.
  - *In vivo* ER: expression of ERα receptor was not affected by topical application of the reference substance PVP-iodine in mares (Study ID 14).
  - *In vivo* (progesterone receptor): the number of PR positive cells (PPC) on Day 6 was reduced. However the infusion process per se may have promoted the disturbance in the PR expression that could not be tested since the control mares were not infused with saline. The progesterone level decrease was not statistically significant.

The WoE suggests that iodine comprises no EAS-related activity. Overall, no toxicologically relevant EAS-related activity was observed.

Rapporteur's comments

Iodine showed anti-estrogenic activity in-vitro in the MCF-7 breast cancer cell line (Stoddard et al., 2008). ER ToxCast bioactivity model outputs are not available for iodine, PVP-iodine or potassium iodide. However, it is available for sodium iodide and the score reported is zero. Thus, the E-modality can be considered as sufficiently investigated according to the ECHA/EFSA ED Guidance document. Overall, the E-modality can be considered as negative.

According to the Guidance document, the A- and S-modalities are not sufficiently investigated for iodine since a Hershberger bioassay (OECD TG 441) and a steroidogenesis assay (OECD TG 456) are lacking. Iodine reduced the expression of progesterone receptors in-vivo in mares (Kalpokas et al., 2010) but the exposure in this study was via intrauterine infusion. Therefore, this study is of limited relevance. The in-silico data presented by the applicant lacks details; for e.g., there is no information on the applicability domain of the predictions made. Overall, the available in-silico and in-vitro data is inadequate to conclude on the A- and S-modalities.

Effects secondary to other toxicities:

According to the ED GD "*adverse effects that are nonspecific secondary consequences of other toxic effects shall not be considered for the identification of the substance as endocrine disruptor*" (ECHA/EFSA, 2018).

For this reason, additional information on systemic general toxicity is depicted in Table B-8 in order to contextualize the presence of an adverse effect potentially linked to an endocrine activity and to allow the assessment of putative secondary effects.

Some effects noted in the toxicity studies with iodine occurred at high doses only in the presence of general adversity, as seen in reduced body weight and decreased food consumption, representing the key adverse effects.

2.1.3. Initial analysis of the evidence and identification of relevant scenario for the ED assessment of EAS-modalities

Table B-11: Selection of relevant scenario

Adversity based on EAS-mediated parameters	Positive mechanistic OECD CF level 2 / 3 Test	Scenario	Next step of the assessment	Scenario selected
No (sufficiently investigated)	Yes/No	1a	Conclude: ED criteria not met because there is not "EAS-mediated" adversity	
Yes (sufficiently investigated)	Yes/No	1b	Perform MoA analysis	
No (not sufficiently investigated)	Yes	2a (i)	Perform MoA analysis (additional information may be needed for the analysis)	
No (not sufficiently investigated)	No (sufficiently investigated)	2a (ii)	Conclude: ED criteria not met because no EAS-mediated endocrine activity observed	X[a]
No (not sufficiently investigated)	No (not sufficiently investigated)	2a (iii)	Generate missing level 2 and 3 information. Alternatively, generate missing "EATS-mediated" parameters. Depending on the outcome move to corresponding scenario	
Yes (not sufficiently investigated)	Yes/No	2b	Perform MoA analysis	

[a] Not all parameters required for EAS-related activity according to the ED GD (ECHA/EFSA, 2018) were reported *in sensu stricto*. However, the available data set regarding EAS-related activity was considered to be sufficient and adequate for a reliable and scientifically sound assessment of EAS-related activity of iodine.

2.1.4. MoA analysis for EAS-modalities

In accordance with the selected "scenario 2a (ii)" (Table B-11), no MoA analysis is required for the EAS-modalities. The "*ED criteria for iodine for EAS-modalities are not met*".

2.1.5. Conclusion of the assessment of EAS-modalities

The present assessment was based on a weight of evidence approach and revealed that no EAS-related activity is attributable to iodine. Additionally no toxicologically

significant EAS–mediated adversity was observed in the available data set. Therefore "*ED criteria regarding EAS modalities are not met*" for iodine.

Rapporteur's comments

EAS-related activity:

ER ToxCast bioactivity model outputs are not available for iodine, PVP-iodine or potassium iodide. However, it is available for sodium iodide and the score reported is zero. Thus, the E-modality can be considered as sufficiently investigated according to the ECHA/EFSA ED Guidance document. Overall, the E-modality can be considered as negative.

According to the Guidance document, the A- and S-modalities are not sufficiently investigated for iodine since a Hershberger bioassay (OECD TG 441) and a steroidogenesis assay (OECD TG 456) are lacking.

EAS-mediated adversity:

The EAS-mediated adversity is not sufficiently investigated for iodine since a two-generation (OECD TG 416; ver. 2001) or an extended-one generation (OECD TG 443) study is lacking.

3. ED assessment for non-target organisms

3.1. ED assessment for EAS-modalities

Currently, EAS-related endocrine activity in the form of specific hormone measurements was not investigated *in vivo* in the available ecotoxicological data set presented in the Assessment Report of iodine (including PVP-iodine) for PT 1, 3, 4 and 22 (AR, 2013). The overall conclusion on the ED assessment on humans revealed that based on the available data set the ED criteria are not met, since iodine displays neither EAS-mediated adversity nor activity. Because of the high level of conservation of the endocrine system across taxonomic groups, the available mammalian data set is considered sufficient and relevant for non-target organisms as well to demonstrate that neither EAS-related activity nor EAS-mediated adversity is attributable to iodine in non-target organisms. Furthermore, with reference to chapter 1.3 of part B, in the literature search no information was identified which might indicate a potential EAS mode of action for non-target organisms. Therefore, for animal safety, further vertebrate studies on non-target organisms are not required.

3.1.1. Conclusion of the assessment of EAS-modalities

Based on the available data, the results from the EAS assessment for mammals and the evaluated scientific literature, for the natural occurring element iodine, the ED criteria for EAS-modalities are not met for non-target organisms. There is no need to generate further information.

Rapporteur's comments

The applicant's literature search did not retrieve any relevant information on potential endocrine disrupting mechanisms or effects related to the EAS modalities with respect to non-target organisms. Neither do the REACH registration dossier or the biocidal assessment report (CAR; 2013) contain any relevant information in that respect. The rapporteur is not aware of any published information related to effects related to the EAS modalities from iodine, PVP iodine or any other iodine containing compound in non-mammalian species. Thus, it is not possible to conclude whether the criteria are met for the EAS modalities in non-target organisms.

Further information will need to be generated if the criteria for endocrine disruption are not met in humans.

Conclusion

In order to answer the question whether iodine possesses estrogen, androgen and steroidogenic (EAS)-mediated endocrine disrupting (ED) properties relevant toxicological and ecotoxicological information was assessed using a weight of evidence (WoE) approach according to the "Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009" (ED GD; (ECHA/EFSA, 2018) and under consideration of the revised GD 150 (OECD, 2018).

The analysis of the toxicological information including *in silico* and *in vitro* mechanistic information revealed no EAS-related activity (sufficiently investigated for a reliable and scientifically sound assessment) attributable to iodine. Moreover no toxicologically significant EAS–mediated adversity was observed in the available data set.

Taking into account the toxicological information and evaluated scientific publications there are no indications that iodine could have any impact on the endocrine system regarding EAS modalities for non-target organisms. Therefore, the ED criteria for EAS-modalities for non-target organisms are not met for the natural occurring element iodine.

Taking together iodine does not compromise EAS-mediated endocrine disrupting properties in humans or non-target organism.

Rapporteur's comments

For conclusion with respect to humans, please see the comment under section 2.1.5.

For non-target organisms, please see the comment under section 3.1.1.

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