Australian Government
Department of Health and Aged Care
Australian Industrial Chemicals Introduction Scheme

1H-Benzotriazole and its mono-substituted derivatives

Evaluation statement

1 October 2024

Draft

Table of contents

Contents

AICIS evaluation statement

Subject of the evaluation

1H-Benzotriazole and its mono-substituted derivatives

Chemicals in this evaluation

Name	CAS registry number
1H-Benzotriazole, 6-chloro-	94-97-3
1H-Benzotriazole	95-14-7
1H-Benzotriazole, 6-methyl-	136-85-6
1H-Benzotriazole, 6(or 7)-methyl-	29385-43-1
1H-Benzotriazole, 7-methyl-	29878-31-7
1H-Benzotriazole, 6-methyl-, sodium salt (1:1)	41253-36-5
1H-Benzotriazole, 6(or 7)-methyl-, sodium salt (1:1)	64665-57-2

Reason for the evaluation

Evaluation Selection Analysis indicated a potential human health risk.

Parameters of evaluation

Chemicals in this evaluation are a group of benzotriazole and its mono-substituted derivatives and their sodium salts that are listed on the Australian Inventory of Industrial Chemicals (the Inventory). These chemicals are grouped together based on their structural similarity and are expected to have similar hazard profiles.

This evaluation is a human health risk assessment for all identified industrial uses of these chemicals. In this evaluation, 1H-benozotriazole (CAS No. 95-14-7), 1H-benzotriazole, 6(or 7)-methyl (CAS No. 29385-43-1), 1H-benzotriazole, 6-methyl- (CAS No. 136-85-6) and 1H-benzotriazole, 6(or 7)-methyl-, sodium salt (1:1) (CAS No. 64665-57-2) will be referred to as benzotriazole, tolyltriazole and 5-tolyltriazole and sodium tolyltriazole, respectively throughout the report.

Summary of evaluation

Summary of introduction, use and end use

There is currently no specific information about the introduction, use and end use of these chemicals in Australia.

Based on international use information, these chemicals function as corrosion inhibitors and have a diverse range of commercial and domestic uses. Reported use categories include in anti-freeze and de-icing products, cleaning products, dishwashing products, paints and coatings, lubricants and greases and water treatment products. The predominant identified domestic uses are in dishwasher tablets and automotive care products (hydraulic fluid and lubricants). Uses in other products including spray cleaning products were identified. Typical use concentrations were reported as 1%.

Three chemicals in this group have site-limited applications as intermediates in the manufacture of other chemicals. Benzotriazole (CAS No. 95-14-7) is used in cosmetic products. Available data does not indicate significant cosmetic use but it has been identified in nail enhancement products (for fake nail plate) at 1% concentration.

No use data are available for 1H-benzotriazole, 6-chloro- (CAS No. 94-97-3).

Human health

Summary of health hazards

The identified health hazards are based on available data for chemicals in this group. Based on the physicochemical properties, these chemicals are expected to be readily absorbed following exposure.

Based on the available data these chemicals are not:

- considered to be skin sensitisers
- expected to cause serious systemic health effects following repeated oral exposure
- considered to have genotoxic potential.

Based on the available data, these chemicals are expected to have moderate acute oral toxicity (median lethal dose (LD50) = 500─1980 mg/kg bw in rats) and moderate acute inhalation toxicity (median lethal concentration (LC50) = 1.7–1.9 mg/L in rats).

Based on the available data for sodium tolyltriazole, the sodium salts are expected to be corrosive to the skin due to their alkalinity (pH 11.5–12 at 25 °C). In a dermal irritation study in rabbits, sodium tolyltriazole caused corrosive effects (visible necrosis) following exposure for 4 hours. Corrosive chemicals are also considered to cause serious eye damage. The other chemicals (non-salts) are expected to have slight skin and eye irritation potential.

The incidence of tumours in rats and mice following exposure to relatively high doses is suggestive of a possible carcinogenic effect of benzotriazole. However, the conclusions of the available studies were equivocal.

Based on the available data, these chemicals may have potential to cause specific adverse effects on foetal development warranting classification. An increase in post-implantation loss

was observed in guideline pre-natal development studies with benzotriazole and tolyltriazole. In addition, effects on foetal body weights and increase in soft tissue and skeletal variations were observed with some high concern malformations observed in a limited number of foetuses. Changes in anogenital distance (although not consistent) were observed at the highest doses. Although maternal toxicity was observed at the highest dose in these studies, some developmental effects were observed at lower doses with minimal maternal toxicity. Sufficient data are not available to draw conclusions regarding effects on fertility.

For further details of the health hazard information see **Supporting information**.

Hazard classifications relevant for worker health and safety

These chemicals satisfy the criteria for classification according to the Globally Harmonized System of Classification and Labelling of Chemicals (GHS) (UNECE 2017) for hazard classes relevant for work health and safety as follows. This evaluation does not consider classification of physical hazards and environmental hazards.

The classification for acute oral and inhalation toxicity and reproductive toxicity applies to all chemicals in this group. The classification for corrosion only applies to sodium salts (CAS Nos. 41253-36-5 and 64665-57-2).

Health hazards	Hazard category	Hazard statement
Acute toxicity	Acute Tox. 4	H302: Harmful if swallowed
Acute toxicity	Acute Tox. 4	H332: Harmful if inhaled
Skin corrosion/irritation	Skin Corr. 1	H314: Causes severe skin burns and eye damage
Reproductive toxicity	Repr. 2	H361d: Suspected of damaging the unborn child

Summary of health risk

Public

Based on the available use information, the public may be exposed to these chemicals:

- by direct application of these chemicals to the nails
- by incidental skin and eye contact with these chemicals during use of domestic products
- by inhaling aerosols during spray applications

Negligible exposure is expected when these chemicals are used in dishwasher tablets. Typical use concentrations in other products reported to be available to consumers is low (1%). This low concentration minimises the risk of local effects on the skin and eyes.

These chemicals have the potential to cause adverse effects on development. Using a worst-case scenario model, the margin of exposure (MOE) for the identified uses was >100, indicating that these chemicals are unlikely to pose a risk of adverse systemic effects.

Therefore, there are no identified risks to the public that require management.

Workers

During product formulation and packaging, dermal, ocular and inhalation exposure might occur, particularly where manual or open processes are used. These could include transfer and blending activities, quality control analysis, and cleaning and maintaining equipment. Worker exposure to these chemicals at lower concentrations could also occur while using formulated products containing these chemicals. The level and route of exposure will vary depending on the method of application and work practices employed. Good hygiene practices to minimise oral exposure are expected to be in place.

Given the local and systemic health effects, these chemicals could pose a risk to workers. Control measures to minimise dermal, ocular and inhalation exposure are needed to manage the risk to workers (see **Proposed means for managing risk** section).

Proposed means for managing risk

Workers

Recommendation to Safe Work Australia

It is recommended that Safe Work Australia (SWA) update the Hazardous Chemical Information System (HCIS) to include classifications relevant to work health and safety.

Information relating to safe introduction and use

The information in this statement including recommended hazard classifications, should be used by a person conducting a business or undertaking at a workplace (such as an employer) to determine the appropriate controls under the relevant jurisdiction Work Health and Safety laws.

Recommended control measures that could be implemented to manage the risk arising from oral, dermal, ocular and inhalation exposure to these chemicals include, but are not limited to:

- using local exhaust ventilation to prevent these chemicals from entering the breathing zone of any worker;
- minimising manual processes and work tasks through automating processes;
- adopting work procedures that minimise splashes and spills;
- cleaning equipment and work areas regularly; and
- using protective equipment that is designed, constructed, and operated to ensure that the worker does not come into contact with these chemicals.

Measures required to eliminate, or manage risk arising from storing, handling and using hazardous chemicals depend on the physical form and the manner in which these chemicals are used.

These control measures may need to be supplemented with conducting health monitoring for any worker who is at significant risk of exposure to these chemicals, if valid techniques are available to monitor the effect on the worker's health.

Personal protective equipment should not solely be relied upon to control risk and should only be used when all other reasonably practicable control measures do not eliminate or sufficiently minimise risk.

Model codes of practice, available from the Safe Work Australia website, provide information on how to manage the risks of hazardous chemicals in the workplace, prepare an SDS and label containers of hazardous chemicals. Your Work Health and Safety regulator should be contacted for information on Work Health and Safety laws and relevant Codes of Practice in your jurisdiction.

Conclusions

The Executive Director proposes to be satisfied that the identified risks to human health from the introduction and use of the industrial chemicals can be managed.

Note:

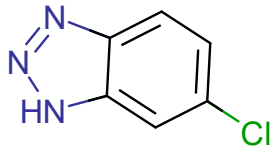
1. Obligations to report additional information about hazards under *Section 100* of the *Industrial Chemicals Act 2019* apply.
2. You should be aware of your obligations under environmental, workplace health and safety and poisons legislation as adopted by the relevant state or territory.

Supporting information

Grouping rationale

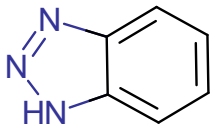
Chemicals in this group are benzotriazole (CAS No. 95-14-7), its derivatives with a methyl substituent on the benzene ring (CAS No. 136-85-6; 29385-43-1 and 29878-31-7) and their sodium salts (CAS No. 41253-36-5 and 64665-57-2). These chemicals are grouped together based on their structural similarity and are expected to have similar hazard profiles. The salts are expected to hydrolyse to form parent methyl benzotriazole in physiological solutions. The chemical 1H-benzotriazole, 6-chloro- (CAS No. 94-97-3) is also included in this group, based on having a similar in silico toxicological profile to other members of the group. No data are available for this chemical.

Chemical identity

CAS number	94-97-3
CAS name	1*H*-Benzotriazole, 6-chloro-
Molecular formula	$C_6H_4ClN_3$
Associated names	1*H*-Benzotriazole, 5-chloro-, 6-Chloro-1*H*-benzotriazole Benzotriazole, 5-chloro-
Molecular weight (g/mol)	153.57
SMILES (canonical)	ClC=1C=CC=2NN=NC2C1
Structural formula	

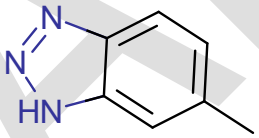
CAS number	95-14-7
CAS name	1*H*-Benzotriazole
Molecular formula	$C_6H_5N_3$
Associated names	1,2,3-Benzotriazole
Molecular weight (g/mol)	119.12
SMILES (canonical)	N1=NC=2C=CC=CC2N1

Structural formula



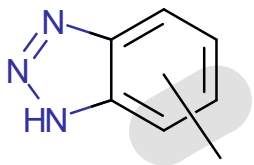
CAS number	136-85-6
CAS name	1*H*-Benzotriazole, 6-methyl-
Molecular formula	$C_7H_7N_3$
Associated names	1*H*-Benzotriazole, 5-methyl Benzotriazole, 5-methyl- 6-Methyl-1*H*-benzotriazole
Molecular weight (g/mol)	133.15
SMILES (canonical)	N1=NC=2C=C(C=CC2N1)C

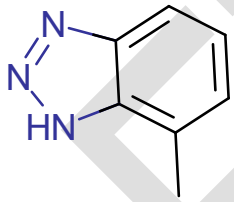
Structural formula

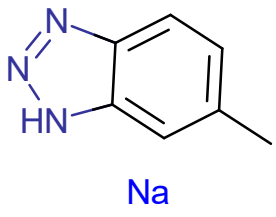


CAS number	29385-43-1
CAS name	1*H*-Benzotriazole, 6(or 7)-methyl-
Molecular formula	$C_7H_7N_3$
Associated names	6(or 7)-Methyl-1*H*-benzotriazole 1*H*-Benzotriazole, 4(or 5)-methyl- Tolyltriazole
Molecular weight (g/mol)	133.15
SMILES (canonical)	-

Structural formula

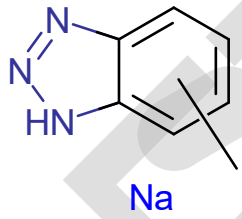


CAS number	29878-31-7
CAS name	1*H*-Benzotriazole, 7-methyl-
Molecular formula	$C_7H_7N_3$
Associated names	1*H*-Benzotriazole, 4-methyl-
	Benzotriazole, 4-methyl-
	7-Methyl-1*H*-benzotriazole
Molecular weight (g/mol)	133.15
SMILES (canonical)	N1=NC=2C=CC=C(C2N1)C
Structural formula	

CAS number	41253-36-5
CAS name	1*H*-Benzotriazole, 6-methyl-, sodium salt (1:1)
Molecular formula*	$C_7H_7N_3.Na$
Associated names	Tolutriazole, sodium salt
	1*H*-Benzotriazole, 5-methyl-, sodium salt
Molecular weight (g/mol)*	156.14
SMILES (canonical)*	[Na].N1=NC=2C=C(C=CC2N1)C
Representative structure*	

Additional chemical identity information

* This chemical is a salt and has been represented according to CAS nomenclature/identity convention.

CAS number	64665-57-2
CAS name	1*H*-Benzotriazole, 6(or 7)-methyl-, sodium salt (1:1)
Molecular formula*	$C_7H_7N_3.Na$
Associated names	Tolyltriazole, sodium salt 1*H*-Benzotriazole, 4(or 5)-methyl-, sodium salt
Molecular weight (g/mol)*	156.14
SMILES (canonical)	-
Representative structure*	

Additional chemical identity information

* This chemical is a salt and has been represented according to CAS nomenclature/identity convention.

Relevant physical and chemical properties

Chemicals in this group have molecular weights ranging between 119.1 and 156.1 g/mol. These chemicals are solid at ambient temperatures with melting point between 85–106 °C and boiling point between 297–320 °C. Based on the calculated vapour pressures <0.001 Pa at 25 °C (calculated with MPBPVPWIN), these chemicals are expected to have low volatility. The dissociation constant values (pKa) for these chemicals are 8.37–9.15 at 20 °C and log K_{ow} (calculated with EPI WSKOW) between 1.08–1.81 at 25 °C (Danish QSAR n.d.).

Introduction and use

Australia

There is currently no specific information about the introduction, use and end use of these chemicals in Australia.

International

The following international uses have been identified through:

- Galleria Chemica (Chemwatch n.d.)
- Substances in Preparations in Nordic Countries (SPIN) database (SPIN n.d.)
- European Union (EU) Registration, Evaluation and Authorisation of Chemicals (REACH) dossiers
- INCIpedia (Personal Care Products Council n.d.)

- the European Cosmetic Ingredient Database (CosIng) (EC n.d.)
- United States Environmental Protection Agency Chemical Data Reporting (CDR) (US EPA 2016, US EPA 2020)
- Draft Screening Assessment Benzotriazoles and Benzothiazoles Group, Environment and Climate Change Canada, Health Canada (Government of Canada 2021)
- Danish Environmental Protection Agency, Benzotriazole and Tolytriazole, Evaluation of health hazards and proposal of health based quality criteria for soil and drinking water (Danish EPA 2013)
- Consumer Products Information Database (DeLima Associates n.d.)

These chemicals have a diverse range of uses based on their function of corrosion inhibitors. All chemicals except 1H-benzotriazole, 6-chloro- have reported commercial uses, including:

- industrial cleaning and washing products
- cutting fluids and lubricants
- anti-freezing agents
- corrosion inhibitors
- surface treatment
- flame retardants and extinguishing agents (reported use concentration of <1%)
- water treatment products.

All chemicals except 1H-benzotriazole, 6-chloro- have reported domestic uses. The predominant identified domestic uses are in dishwasher tablets and automotive care products (hydraulic fluid and lubricants). Other identified uses include:

- liquid impression pen (reported use concentration of 1%)
- spray cleaning products (reported use concentration of <1%)
- cooling system repair (reported use concentration of 1%)
- scented candles.

Benzotriazole has reported uses in cosmetics with a reported function as a preservative (Personal Care Products Council n.d.). Data available does not indicate significant use but it has been identified as used in nail enhancement product for fake nail plate (reported use concentration of 1%) (Government of Canada 2021).

Benzotriazole, tolyltriazole and sodium tolyltriazole have reported site-limited uses, including as intermediates in product formulation and chemical synthesis.

Benzotriazole, tolyltriazole and sodium tolyltriazole have reported non-industrial uses as pesticides.

Existing Australian regulatory controls

AICIS

No specific controls are currently available for these chemicals.

Public

No specific controls are currently available for these chemicals.

Workers

These chemicals are not listed on the Hazardous Chemical Information System (HCIS) (Safe Work Australia, SWA).

No exposure standards are available for these chemicals in Australia (SWA n.d.).

In 2019, SWA reviewed and made recommendations regarding workplace exposure standards for 1H-benzotriazole (CAS No. 95-14-7).

The SWA review stated: “A workplace exposure standard is not recommended as the available data is considered insufficient to support a health based recommendation. An evaluation of additional sources, including dermal studies, are recommended at the next scheduled review” (SWA 2019).

International regulatory status

Exposure standards

The following exposure standards were identified (Chemwatch n.d.):

- Time weighted average (TWA): 5 mg/m^3, in Latvia and Russia (CAS No. 95-14-7; 136-85-6)
- Short-term Exposure Limit (STEL): 5 mg/m^3, in Belarus (CAS Nos. 95-14-7; 136-85-6).

Human exposure

Public

The public may also be exposed to these chemicals as they are present in a range of domestic products including cleaning products, dishwashing products and automotive care products. When used in dishwasher tablets negligible public exposure is expected. To determine the daily systemic exposure to benzotriazole from cleaning spray products, a worst case scenario estimate was determined using ConsExpo Web (RIVM n.d.).

A default dermal and inhalation absorption value of 100% was assumed and default adult body weight of 60 kg were assumed for these estimates. The typical use scenario was spraying an all-purpose cleaning spray containing the chemical (at 1% concentration) on a kitchen worktop and then wiping down the surface with a wet cloth, once daily. Therefore, the estimate accounted for both dermal and inhalation exposure to benzotriazole. The parameters used for the models were based on the scenario for ‘All-purpose cleaning spray’ presented in the RIVM Cleaning products Fact Sheet (RIVM 2018).

The aggregate worst case daily systemic exposure to benzotriazole from this cleaning spray use scenario was 0.068 mg/kg bw/day. The estimate was based on 3 exposures as described below: 1) inhalation of aerosolised spray, 2) incidental dermal contact with spray and 3) dermal contact with cloths used when wiping down the surface.

For the inhalation of aerosols during spraying, the daily systemic exposure was estimated to be 0.012 mg/kg bw/day. This estimate was based on the following parameters and assumptions (RIVM 2018):

- Exposure model: Exposure to spray – spraying
- Spray duration: 0.23 minutes
- Exposure duration: 60 minutes
- Room volume: 15 m^3
- Room height: 2.5 m
- Ventilation rate: 2.5 per hour
- Inhalation rate: 1.49 m^3 per hour
- Mass generation rate: 1.6 g/s
- Airborne fraction: 0.1
- Density non-volatile: 1 g/cm^3
- Inhalation cut-off diameter: 15 µm
- Aerosol diameter (Log-normal): median diameter of 2.4 µm with coefficient of variation 0.37 and maximum diameter 50 µm.

For incidental dermal exposure to the cleaning spray, the daily systemic exposure was estimated to be 0.0036 mg/kg bw/day. This estimate was based on the following parameters and assumptions (RIVM 2018):

- Exposure model: Direct product contact – Constant rate loading
- Exposed area: 2,200 cm^2
- Contact rate: 46 mg/min
- Release duration: 28 seconds.

For dermal exposure on the hands when using a cloth to wipe the excess spray off the worktop, the daily systemic exposure was estimated to be 0.052 mg/kg bw/day. This estimate was based on the following parameters and assumptions (RIVM 2018):

- Exposure model: Direct product contact – Instant application
- Exposed area: 225 cm^2
- Product amount: 0.31 g
- Retention factor: 1.

Estimates of daily systemic exposure as a result of uses in automotive cooling system repair (1%) are 0.027 mg/kg bw/day (Government of Canada 2021).

Limited use of benzotriazole in cosmetic nail products has been identified (1% concentration). Estimates of daily systemic exposure for teenagers are 0.027 mg/kg bw/day (Government of Canada 2021).

Health hazard information

Toxicokinetics

No data are available for this group of chemicals.

Based on the molecular weights and estimated log K_{ow} values (see relevant physico-chemical properties), chemicals in this group are expected to be readily absorbed following oral and dermal exposure. Based on calculated vapour pressures, inhalation exposure is not

expected unless dusts/aerosols are formed. Benzotriazole undergoes hydroxylation (phase I metabolic reaction) at a slow rate when incubated in rat liver microsome at a final concentration of 0.2 mg/mL. In an hour, <5% of benzotriazole was metabolised into 4- and 5-hydroxybenzotriazole (0.32 and 1.6%, respectively). These chemicals are not likely to undergo phase II metabolic reactions and are expected to be excreted mainly via urine or faecal routes (REACH n.d.-a).

Acute toxicity

Oral

Based on the available data, chemicals in this group are considered to have moderate acute toxicity following oral exposure, warranting hazard classification.

In a GLP compliant acute oral toxicity study conducted in accordance with OECD Test Guideline (TG) 423, Sprague Dawley (SD) rats (3/dose) were treated with benzotriazole at 300 or 2000 mg/kg bw via gavage. All animals dosed at 2000 mg/kg bw died whereas all animals dosed at 300 mg/kg bw survived. Therefore, the median lethal dose LD50 is in the range 300–2000 mg/kg bw. Reported sublethal signs of toxicity included decrease or absence in spontaneous activity, in muscle tone, in righting reflex and in Preyer's reflex, pupil dilation, lacrimation, bradypnoea and partial ptosis (REACH n.d.-a).

In a GLP compliant acute oral toxicity study similar to OECD TG 401, Bor:WISW rats (5/sex/dose) were treated with tolyltriazole at 500, 630, 730, 800, 1000 and 1300 mg/kg bw via gavage. The reported LD50 was 720 mg/kg bw. Reported sublethal signs of toxicity included body weight loss, sedation, paralysis of rear extremities and bloody mouth (REACH n.d.-c).

In a GLP compliant acute oral toxicity study similar to OECD TG 401, SD rats (5/sex/dose) were treated with sodium tolyltriazole at 362–1080 mg/kg bw via gavage. The reported LD50 was 735 and 930 mg/kg bw for females and males, respectively. Reported sublethal signs of toxicity included decreased physical activity at lower doses to ataxia at higher doses (REACH n.d.-d).

The following additional oral LD50 values were reported:

- 500–964 mg/kg bw in rats and 615–831 mg/kg bw in mice for benzotriazole (CCOHS 2023a; Danish EPA 2013)
- >1600 mg/kg bw in rats for 5-tolyltriazole (CCOHS 2023b; REACH n.d.-b)
- 675–1830 mg/kg bw in rats and 800 mg/kg bw in mice for tolyltriazole (CCOHS 2023c; Danish EPA 2013)
- 640–1980 mg/kg bw in rats for sodium tolyltriazole (CCOHS 2023d).

Dermal

Based on the available data, chemicals in this group are considered to have low acute toxicity following dermal exposure.

In a GLP compliant acute dermal toxicity study conducted similarly to OECD TG 402, New Zealand white (NZW) rabbits (5/sex) were treated with a single dose of the chemical reported as a mixture of tolyltriazole and sodium tolyltriazole at 2000 mg/kg bw of under occlusive conditions. The reported median LD50 was >2000 mg/kg bw. No mortality occurred. Reported clinical signs of toxicity included ataxia, hypersalivation and nasal discharge.

Oedema was noted only on day 1. Erythema and discolouration of the back were apparent from day 1 to 14 (REACH n.d.-c; REACH n.d.-d).

In an acute dermal toxicity study in rabbits (n=5), the reported LD50 was >2000 mg/kg bw for benzotriazole. No signs of toxicity were reported other than irritation of the skin (Danish EPA 2013; REACH n.d.-a).

The following additional dermal LD50 values were reported:

- >2000 mg/kg bw in rabbits and rats for benzotriazole (CCOHS 2023a)
- >2000 and >4000 mg/kg bw in rabbits and >4000 mg/kg bw in rats tolyltriazole (CCOHS 2023c)
- >2000 mg/kg bw in rabbits for sodium tolyltriazole (CCOHS 2023d).

Inhalation

Based on the available data, chemicals in this group may have moderate acute toxicity following inhalation exposure, warranting hazard classification.

In an acute inhalation toxicity study in male SD rats (n=10), animals were exposed (whole body) to benzotriazole (aerosol) at 780, 1460, 2030, 2230 or 2710 mg/m^3 for 3 hours. To aerosolise the chemical was heated to 200°C. The reported median lethal concentration (LC50) was 1910 mg/m^3 (1.9 mg/L) for benzotriazole. Reported clinical signs of toxicity included deep abdominal breathing with open mouth gasping. Accumulation of white frothy fluid in the trachea and moderate to severe incidence of dark red haemorrhagic areas in the lungs were reported. There is uncertainty whether mortalities in this study were directly attributable to the test material or the heat of the aerosol (Danish EPA 2013; REACH n.d.-a).

In rats exposed to tolyltriazole (aerosol) at 95 to 323 mg/m^3 for 3 hours, respiratory irritation, bleeding of the respiratory tract due to local irritation, and liver and kidney effects have been reported (Danish EPA 2013).

In rats exposed for 1 hour to tolyltriazole (aerosol) at 1.73 g/m^3 and then observed for a period of 14 days, no toxic effects were reported. Haemorrhagic areas were detected in the lungs (Danish EPA 2013).

Corrosion/Irritation

Skin irritation

Based on the available data, the sodium salts are expected to be corrosive to the skin due to their alkalinity. Sodium tolyltriazole is synthesised via reactions with strong inorganic bases of sodium hydroxide NaOH and has pH of 11.5–12 at 25 °C (REACH n.d.-d).

In a non-GLP compliant skin irritation study conducted in accordance with OECD TG 404, the skin of 3 NZW rabbits (sex not specified) was applied with sodium tolyltriazole (50% solution Preventol CI 7-50, 0.5 mL) for 4 hours under semi-occlusive conditions. The following mean scores were reported for observations at 24, 48 and 72 hours: 1.66, 4, 4 for erythema and 2, 2.3, 2.3 for oedema, respectively. An erythema was considered not reversible in all animals. Signs of corrosive effects include grey black skin discolouration in 2/3 animals after 1 hour and bloody eschar formation (resulting from necrosis) in 2/3 animals after 24 hours (CCOHS 2023d; REACH n.d.-d).

Only mild signs of irritation were reported for sodium tolyltriazole in the acute dermal toxicity study (see **Acute toxicity – Dermal** section); however, this may be a result of the chemical being used in a mixture with tolyltriazole (at unknown concentration).

Based on the available data, the remaining chemicals in this group, are slightly irritating to the skin. They do not warrant hazard classification.

In a GLP compliant skin irritation study conducted in accordance with OECD TG 404, benzotriazole (500 mg pasted in water) was applied to the skin of 3 NZW rabbits (sex not specified) for 4 hours under semi-occlusive conditions. Observations were recorded at 24, 48 and 72 hours after patch removal. The mean erythema and oedema scores were 0/4 and 0/4, across all time points. There were no reported signs of dermal irritation in any animal (REACH n.d.-a).

Application of benzotriazole (500 mg) to the intact and abraded skin of 12 rabbits induced no irritation (Danish EPA 2013; USEPA 1977). Signs of skin irritation were reported in 3/5 rabbits after 2000 mg/kg bw benzotriazole was applied for 24 hours to abraded skin (Danish EPA 2013). Application of benzotriazole (50% in ethanol) induced slight irritation in guinea pigs (Danish EPA 2013). No further study details were available.

In an in vitro skin corrosion assay conducted in accordance with OECD TG 431, 5-tolyltriazole was applied to reconstructed human skin model EpiDerm™ for 3 and 60 minutes. The mean tissue viability was reported as ≥9.7% to ≤104.8% (duration of exposure not specified) (REACH n.d.-b). No comparison with the classification criteria could be made based on available data.

Application of undiluted 5-tolyltriazole induced skin irritation in rabbits and guinea pigs after 24 hours (Danish EPA 2013). No further study details were available.

In a non-GLP compliant skin irritation study conducted in accordance with OECD TG 404, the skin of 3 NZW rabbits (sex not specified) was applied with tolyltriazole (500 mg pasted in water) for 4 hours under semi-occlusive conditions. Observations were recorded at 24, 48 and 72 hours and at 7 days after patch removal. The mean erythema and oedema scores were 0/4 and 0/4, across all time points. There were no reported signs of dermal irritation in any animal (REACH n.d.-c).

Application of undiluted tolyltriazole to intact or abraded skin of rabbits induced slight or no irritation (Danish EPA 2013). Application of tolyltriazole (10% in petrolatum) induced no skin irritation in guinea pigs (Danish EPA 2013). No further study details were available.

Eye irritation

Based on the available data for benzotriazole and tolyltriazole, chemicals in this group, with the exception of the salts, are slightly irritating to eyes. In two guideline studies, only slight fully reversible irritant effects were observed. The sodium salts are expected to be corrosive to skin. Corrosive chemicals are also presumed to cause irreversible effects on the eyes. No information was available for studies reporting severe irritation.

In an eye irritation study conducted in accordance with OECD TG 405, 0.1 mL of benzotriazole was instilled into the conjunctival sac of one eye each of 3 NZW rabbits. The eyes were rinsed after 24 hours. Observations were recorded at 1, 24, 48, 72 hours, and at 7 days. Individual animal scores were not provided. The following mean scores (mean of 3 animals) were reported at 24, 48 and 72 hours: corneal opacity 1/4, 0.66/4, 0/4, iritis 0.33/2,

0.33/2, 0.33/2, conjunctival redness 1.33/3, 1.66/3, 0.66/3 and chemosis 1.33/4, 1033/4, 0/4. The observed effects were fully reversible in all animals within 7 days (REACH n.d.-a).

Application of benzotriazole (100 mg, dry powder) induce severe eye irritation in 6 NSW rabbits (Danish EPA 2013). No further study details were available.

In a non-GLP compliant eye irritation study conducted in accordance with OECD TG 405, 0.1 mL of tolyltriazole was instilled into the conjunctival sac of one eye each of 3 NZW rabbits. The eyes were rinsed after 24 hours. Observations were recorded at 1, 24, 48, 72 hours, and at 7 days. The following mean scores were reported at 24, 48 and 72 hours: corneal opacity 0/4, 0.66/4, 0/4, iritis 0/2,0/2,0/2, conjunctival redness 1/3, 1/3, 0.66/3, and chemosis 0.33/4, 0.66/4, 0.33/4. The observed effects were fully reversible in all animals within 7 days (REACH n.d.-c).

Application of tolyltriazole (10 mg or 100 mg or 0.1 mL of a 35% solution in isopropanol, ~35 mg) induced slight to moderate eye irritation in rabbits (Danish EPA 2013). No further study details were available.

Sensitisation

Skin sensitisation

Based on the data available for benzotriazole and tolyltriazole, chemicals in this group are not considered to be potent skin sensitisers. Weak sensitisation reactions to commercial grade chemical cannot be ruled out.

In a GLP compliant guinea pig maximisation test (GPMT) conducted according to OECD TG 406, male DHPW guinea pigs (n=20) were intradermally injected with benzotriazole at 5% in propylene glycol and topically induced with the chemical at 25% in propylene glycol. After being challenged with the chemical at 12% in propylene glycol, positive reactions were observed in 0.5% (1/20) of the animals (REACH n.d.-a).

In an in vivo skin sensitisation study described as being a GPMT, intradermal injection was performed on guinea pigs (Pirbright-Hartley, 20/sex/dose) using benzotriazole (commercial grade or purified) at 1% in physiological saline and saline adjuvant mixture and epidermal induction with the chemical at 30% in petrolatum. Challenge with the chemical (commercial grade) at 30% in petrolatum resulted in positive reactions in 15% (3/20) of the animals. No skin reactions were observed in any of the animals challenged with purified benzotriazole (REACH n.d.-a).

In a non-guideline sensitisation study described as Maurer optimisation test, intradermal injection was performed on guinea pigs (Pirbright-Hartley; 20/sex/dose) using benzotriazole (commercial grade or purified) at 0.1% in physiological saline and in saline complete adjuvant Freund mixture. Epidermal challenge with the chemical at 30% in petrolatum resulted in positive reactions in 25% (5/20) and 15% (3/20) of the animals for commercial grade and purified benzotriazole, respectively. No skin sensitisation reactions were observed in epidermally challenged animals (Danish EPA 2013; REACH n.d.-a).

In a GLP compliant GPMT conducted according to OECD TG 406, intradermal injection was performed on male Dunkin-Hartley guinea pigs (n=20) using tolyltriazole at 5% in propylene glycol and topical induction with the chemical at 50% in propylene glycol. No skin sensitisation reactions were observed in any of the animals challenged with the chemical at 6% in propylene (REACH n.d.-c).

No skin sensitisation reaction was observed in a non-guideline sensitisation test in guinea pigs (10/sex) treated with tolyltriazole at 10% in petrolatum (Danish EPA 2013).

Observation in humans

In 4 human patch tests, positive reactions were observed in 4 subjects with contact dermatitis when treated with products containing benzotriazole. In another human patch test, there was no evidence of skin sensitisation for benzotriazole at 1% in petrolatum in 145 subjects (car mechanics and metal workers with contact dermatitis) (Danish EPA 2013).

In silico

These chemicals have no structural alerts for protein binding based on the mechanistic profiling functionality of the Organisation for Economic Co-operation and Development (OECD) QSAR Application Toolbox (OECD QSAR Toolbox v4.5). These chemicals are predicted to be non-sensitising using OASIS–TIMES (Optimised Approach based on Structural Indices Set–Tissue Metabolism Simulator; version 2.31) (OASIS LMC), and the knowledge-based expert system Deductive Estimation of Risk from Existing Knowledge (DEREK) Nexus version 6.0.1 (Lhasa Limited).

Repeat dose toxicity

Oral

Based on the available data for benzotriazole and tolyltriazole, chemicals in this group are not expected to cause serious damage to health following repeated oral exposure. The severity of the adverse effects or doses at which effects were observed in various organs of rats and mice is not sufficient to warrant hazard classification.

In two sub-chronic repeated dose toxicity studies, F344 rats and B1C3F1 mice (5/sex/dose) were administered benzotriazole in feed at 300, 1000, 3000, 10000, or 30000 ppm (equivalent to 15, 50, 150, 500 and 1500 mg/kg bw/day in rats; 45, 150, 450, 1500 and 4500 mg/kg bw/day in mice) 7 days/week for 8 weeks. No mortality occurred during the study. Mean body weight depressions of ≤12% were observed at 300 to 10000 ppm in male and female rats, and 40% and 34% of male and female rats, respectively at the highest dose. In mice, a slight weight depression of approximately 5% was observed at 10000 ppm in both sexes. No mortality or other treatment related clinical signs of toxicity were reported (Danish EPA 2013; NTP 1978). The dose range identified from these studies were used to conduct the following chronic combined repeated dose and carcinogenicity studies in rats and mice.

In a chronic combined repeated dose and carcinogenicity study similar to OECD TG 451, Fischer 344 rats (50/sex/dose) were administered benzotriazole (>99% purity) in feed at 6700 or 12100 ppm (equivalent to 335 and 605 mg/kg bw/day) for 78 weeks followed by 27 weeks observation. Mortality was observed in dosed and control groups (14–36%), although it was reported to be non-treatment related. Mean body weights were lower in both male and female rats at all doses compared to the control group. Reported non-cancer effects included prostate and uterine inflammation, cytoplasmic changes in the liver cells (clear cell, eosinophilic and basophilic alterations) and kidney epithelial hyperplasia and nephrosis. However, these incidences did not show a dose related trend. Based on the data, the lowest observed adverse effect level (LOAEL) was determined to be 335 mg/kg bw/day in rats (Danish EPA 2013; NTP 1978; REACH n.d.-a).

In a chronic combined repeated dose and carcinogenicity study similar to OECD TG 451, B6C3F1 mice (50/sex/dose) were administered benzotriazole (>99% purity) in feed at 11700 or 23500 ppm (equivalent to 1755 and 3525 mg/kg bw/day) for 104 weeks followed by a 2 week observation period. Mortality was observed in dosed and control groups (6–40%), although it was reported to be non-treatment related. Mean body weights were lower in both male and female mice at all doses compared to the control group. Reported non-cancer effects included decreased growth and damage to the bone marrow, lymph nodes and lungs, kidney nephrosis and hyperplasia in the spleen. However, these incidences did not show a dose related trend. Based on the data, the LOAEL was determined to be 1755 mg/kg bw/day in mice (Danish EPA 2013; NTP 1978; REACH n.d.-a).

In a reproductive and developmental toxicity study, SD rats (12/sex/dose) were administered benzotriazole via gavage at doses of 0, 30, 100 or 300 mg/kg bw/day (duration of treatment not available in the study). Regeneration of proximal tubules in the kidneys was reported in adult female rats at 100 mg/kg bw/day. Reversible and irreversible haematological effects were observed in adult males and females at the highest dose. The reported no observed adverse effect level (NOAEL) was 30 mg/kg bw/day in rats based on kidney effects at the higher dose level (Government of Canada 2021).

In a 28 day repeated dose oral toxicity study, Wistar rats (6/sex/dose) were administered tolyltriazole via gavage at doses of 0, 50, 150 or 450 mg/kg bw/day. Signs of potential liver toxicity were reported in the study. At the highest dose, a decrease in plasma proteins and an increase in the activities of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) was reported in both sexes, and reduced levels of erythrocytes, haemoglobin and haematocrit in males. There were no changes to organ weights or histopathological findings. The reported NOAEL was 150 mg/kg bw/day in rats based on haematological and biochemical effects (Government of Canada 2021). No further study details were available.

In a non-guideline repeated dose oral toxicity study, 15 male rats (strain not specified) were administered tolyltriazole at 0.5% in diet (equivalent to 375 mg/kg bw/day) for 8 weeks followed by 8 weeks observation. No mortality was reported in the study (Danish EPA 2013). No further study details were available.

Genotoxicity

Based on the available data, chemicals in this group are not expected to be genotoxic.

In vitro

The available in vitro data for these chemicals were mostly negative.

The following results were reported for benzotriazole:

- negative in 2 bacterial reverse mutation assays (OECD TG 471) in Salmonella typhimurium strains TA 97a, TA 98, TA 100, TA 102 and TA 1535 with and without metabolic activation (S9) at concentrations of 50–5000 µg/plate (REACH n.d.-a)
- negative in a bacterial reverse mutation assay (OECD TG 471) in S. typhimurium strains TA 97, TA 98, TA 100 and TA 1535 with and without metabolic activation at concentrations of 0–1666 µg/plate (NTP 1978)
- negative in a mammalian cell gene mutation study (OECD TG 476) in hypoxanthine-guanine phophoribosyl transferase (HPRT) gene in Chinese hamster ovary (CHO) cells

with and without metabolic activation at concentrations of 50–1000 µg/mL (REACH n.d.-a)
- negative without metabolic activation and positive with metabolic activation in a mammalian chromosome aberration assay in CHO cells at concentrations of 0–2500 µg/mL (NTP 1978)
- positive without metabolic activation and negative with metabolic activation in 2 sister chromatid exchange (SCE) assays in CHO cells at concentrations of 0–3000 µg/mL (NTP 1978).

The following results were reported for tolyltriazole:

- negative in a bacterial reverse mutation assay (OECD TG 471) in *S. typhimurium* strains TA 98, TA 100, TA 1535 and TA 1537 with and without metabolic activation at concentrations of 20–2000 µg/plate (REACH n.d.-c)
- negative in a bacterial reverse mutation assay (OECD TG 471) in *S. typhimurium* strains TA 97, TA 98, and TA 1535 with and without metabolic activation, and negative without metabolic activation and equivocal with metabolic activation in *S. typhimurium* TA 100 at concentrations of 0–6666 µg/plate (NTP 1978)
- negative in the transformation of mouse cells or DNA damage in human lung cells (no further study details were available) (Danish EPA 2013).

The following results were reported for 5-tolyltriazole:

- negative in a bacterial reverse mutation assay (OECD TG 471) in *S. typhimurium* strains TA 97a, TA 98, TA 100, TA 102 and TA 1535 with and without metabolic activation at concentrations of 15–5000 µg/plate (REACH n.d.-b)
- negative in a CA assay in hamster cells with and without metabolic activation (no concentrations available) (Danish EPA 2013).

In vivo

Limited in vivo data are available for chemicals in this group due to the mostly negative in vitro results obtained. The available data are negative for genotoxicity.

In a GLP compliant mammalian erythrocyte micronucleus test conducted in accordance with OECD TG 474, NMRI mice (5/sex) were administered benzotriazole by gavage at a single dose of 800 mg/kg bw in polyethylene glycol 400. The incidence of micronuclei in bone marrow polychromatic erythrocytes did not increase in any of the treated groups, indicating a lack of clastogenicity (REACH n.d.-a).

In a GLP compliant mammalian erythrocyte micronucleus test similar to OECD TG 474, NMRI mice (5/sex) were administered tolyltriazole by gavage at a single dose of 600 mg/kg bw in polyethylene glycol. The incidence of micronuclei in bone marrow polychromatic erythrocytes did not increase in any of the treated groups, indicating a lack of clastogenicity (REACH n.d.-c).

In silico

Chemicals in this group present alerts for in vivo mutagenicity (micronucleus) based on the molecular structure as profiled by the OECD QSAR Toolbox v4.5 (OECD 2020). As a hydrogen receptor, these chemicals have potential to interact with DNA and proteins via non-covalent binding. The expert rule based system, SARAH Nexus version 3.1.0 (Lhasa Limited n.d.) identified alerting group (mutagenicity in vitro) for benzotriazole, but no

structural alert presented for the rest of the group. These chemicals were predicted to be in vitro Ames negative in OASIS TIMES (OASIS LMC).

Carcinogenicity

The incidence of tumours in rats and mice following exposure to relatively high doses is suggests a possible carcinogenic effect of benzotriazole although overall evidence is considered to be equivocal. In the absence of more comprehensive information, carcinogenicity classification is not warranted for this group of chemicals.

In a chronic combined repeated dose and carcinogenicity study similar to OECD TG 451 (see **Repeated Dose Toxicity: Oral** section), Fischer 344 rats (50/sex/dose) were administered benzotriazole (>99% purity) in feed at 6700 or 12100 ppm (equivalent to 335 and 605 mg/kg bw/day). At the high dose, neoplastic nodules of the liver were observed in 5/45 (11%) males and in 2/50 (4%) females. However, it was noted in the study that the incidences of tumours could not be clearly associated with the administration of the chemical as similar incidences had been observed in historical controls at the same laboratory. Brain tumours (oligodendrogliomas and gliomas) were observed in 3/44 (7%) males at low dose, and in 1/50 (2%) females at high dose. The observation of these rare tumours suggests a potential carcinogenic effect of the chemical. In female rats, incidences of endometrial stromal polyps or endometrial stromal sarcomas were reported in control 4/48 (8%), at low dose 10/45 (22%) and at high dose 9/50 (18%). However, it was concluded that there was no evidence that under the conditions of this bioassay that the chemical was carcinogenic (Danish EPA 2013; NTP 1978; REACH n.d.-a).

In a chronic combined repeated dose and carcinogenicity study similar to OECD TG 451 (see **Repeated Dose Toxicity: Oral** section), B6C3F1 mice (50/sex/dose) were administered benzotriazole (>99% purity) in feed at 11700 or 23500 ppm (equivalent to 1755 and 3525 mg/kg bw/day). An increased incidence of alveolar/bronchiolar carcinomas was reported in female mice (control 0/49, low dose 9/49 (18%), high dose 3/49 (6%)). However, it was noted in the study that the incidences did not show a dose related trend and that a similar incidence had been observed in historical controls in the same laboratory. No statistically significant tumour incidences were reported in male mice at both doses. It was concluded that there was no convincing evidence that under the conditions of this bioassay that the chemical was carcinogenic (Danish EPA 2013; NTP 1978; REACH n.d.-a).

In silico

Chemicals in this group have no alert for carcinogenicity (genotoxic and non-genotoxic) based on the molecular structure as profiled by the OECD QSAR Toolbox v4.5 (OECD 2020).

Reproductive and development toxicity

Based on the available data, chemicals in this group may have potential to cause specific adverse effects on foetal development warranting classification. An increase in post-implantation loss was observed in guideline pre-natal development studies with benzotriazole and tolyltriazole. In addition, effects on foetal body weights and increase in soft tissue and skeletal variations were observed with some high concern malformations observed in a limited number of foetuses. Changes in anogenital distance (although not consistent) were observed at the highest doses. Although maternal toxicity was observed at the highest dose in these studies, some developmental effects were observed at lower doses

with minimal maternal toxicity. Sufficient data are not available to draw conclusions regarding effects on fertility.

In a GLP compliant combined reproduction/developmental toxicity screening test conducted in accordance with OECD TG 421, Wistar rats (12/sex/dose) were administered benzotriazole (99.87% purity) in polyethylene glycol by gavage at concentrations 0, 12.5, 50 or 200 mg/kg bw/day. The animals were treated from 14 days pre-mating to between day 8 and 14 of lactation for females, and between 39 and 50 days for males. No treatment related mortality was reported. No treatment related effects were observed in both F0 and F1 generations up to the highest dose tested. Based on these observations, the NOAEL value was determined to be 200 mg/kg bw/day for reproductive and developmental toxicity (Government of Canada 2021; REACH n.d.-a). Since this study was performed before the update of OECD TG 421, anogenital distance (AGD), nipple retention, and thyroid hormone or thyroid-stimulating hormone levels, or thyroid histology were not analysed.

In a GLP compliant prenatal developmental toxicity study conducted in accordance with OECD TG 414, pregnant SD rats (n = 24–26/dose) were administered benzotriazole (vehicle: Kollisolv® PEG E400) by gavage once daily at 36, 120 or 330 mg/kg bw/day on gestational days (GD) 5–19. Dams were sacrificed on GD 20 and the developmental and reproductive toxicity parameters were examined. No treatment related mortality was reported. Clinical signs of toxicity were observed at the highest dose including hypokinesia (decreased motor activity) (12 females), staggered gait (11 females) and flattened posture (6 females).

Treatment related thyroid C-cell hyperplasia (minimal severity) was observed at 120 mg/kg bw/day (3/24) and at 330 mg/kg bw/day (4/23). No significant changes in the level of thyroid hormones (T3, T4 and TSH) were reported. There is no information on calcitonin/calcium homeostasis.

No significant changes in the number of implantation sites, corpora lutea and pre-implantation loss was reported. The relative post-implantation loss (associated with early resorptions) was statistically significantly increased in the 120 and 330 mg/kg bw/day dose groups compared to the control ($p < 0.01$ and $p < 0.05$, respectively). A trend for lower litter size (13.0, 13.2, 12.6, and 12.2 at 0, 36, 120, and 330 mg/kg bw/day, respectively). In addition, the total number of females with post-implantation loss was increased (but not statistically significant) in the 120 and 330 mg/kg bw/day dose groups. As specific numbers for post-implantation loss were not available it is not possible to compare effects with historical control data.

The mean body weight of male and female foetuses, and mean weight of litter was significantly decreased by 11.2%, 11.8% and 11.5%, respectively in the highest dose group. The normalized anogenital distance was statistically significantly increased in female foetuses. The anogenital distance was not changed in male foetuses. Nipple retention was not examined in the study. At 120 mg/kg bw/day, inward rotated hindlimbs, domed head and dilated ventricles of the brain (in one foetus) was observed in three foetuses from one litter. However, no skeletal and visceral malformations were reported for these foetuses, although altered ossifications and soft disintegrating consistency of skeleton was observed in one foetus. There was no significant increase in foetal and litter incidence of skeletal observations. At 330 mg/kg bw/day, external malformations (inward rotated hindlimbs, absent of some digits on the left hindlimb, thread-like tail, and absent of anus) was reported in one foetus. Incidence of altered ossification (bone formation) and soft tissue variations in the liver (pale spotted), kidney (discolouration) and uterus (thin uterine horns) were significant increased at this dose (REACH n.d.-a). The NOAEL values are considered to be 36 mg/kg bw/day for maternal toxicity based on clinical signs and C-cell hyperplasia and 36

mg/kg bw/day for developmental toxicity based on post-implantation loss, changes in foetal weight and litter size, external and skeletal malformations. An increased anogenital distance in female foetuses was also noted at highest dose.

In a GLP compliant prenatal developmental toxicity study conducted in accordance with OECD TG 414, pregnant SD rats (n=24–25/dose) were administered tolyltriazole (vehicle: Kollisolv® PEG E40ture) by gavage once daily at 30, 90 or 300 mg/kg bw/day on GD 5–19. Dams were sacrificed on GD 20 and the developmental and reproductive toxicity parameters were examined. No treatment-related mortality, and statistically significant body or uterine weight change was reported in all dosed groups. Clinical signs of toxicity were observed at the highest dose including hypokinesia (all females), staggering gait (13 females) and flattened posture (9 females). The final maternal body weight without a gravid uterus did not statistically differ from the body weight of control female.

At 300 mg/kg bw/day, the mean level of thyroid stimulating hormone was decreased (24.5% compared to the control), although there were no histological alterations in the thyroid gland.

No significant changes in the number of implantation sites, corpora lutea and pre-implantation loss was reported. The relative post-implantation loss (associated with early resorptions) was increased in the 300 mg/kg bw/day dose group compared to the control. The number of females with post-implantation loss was statistically increased in this group (17/23) compared to the control group (8/22).

Mean body weight of male and female foetuses was significantly decreased (11.5% and 12.7%, respectively) in the highest dose group. Although not significant, a slight tendency to decrease in foetal body weight was observed in the 90 mg/kg bw/day dose group (by 4.9% males, and 4.1% females).

The absolute and normalised anogenital distance was statistically significantly reduced in male foetuses (2.5%) in the highest dose group. The anogenital distance was not significantly changed in female foetuses. Nipple retention was not examined in the study. The foetal and litter incidence of small foetuses was 5.7 % and 23.8 %, respectively at 90 mg/kg bw/day, and 1.7 % and 14.7 %, respectively at 300 mg/kg bw/day compared to control. No test item related external and visceral malformations were reported. Skeletal malformations (absence of one 13th rib) in two foetuses from two litters, and skeletal variations (supernumerary thoracolumbar ribs) in 4 foetuses from two litters were observed at 300 mg/kg bw/day.

Foetal and/or litter incidence of skeletal and cartilage variation was significantly increased in all test groups compared to control. The incidence of total affected foetuses with variations and other alterations in soft tissues was increased in the 90 mg/kg bw/day dose group ($p < 0.05$, foetal and litter incidence) and in the 300 mg/kg bw/day dose group ($p < 0.05$ for litter incidence). Reported significant incidence of soft tissue variations include soft tissue variations (liver, at 90 and 300 mg/kg bw/day), testes malposition (at 90 mg/kg bw/day), and increased percentage of intra-abdominal haemorrhage and enlarged blood-filled heart atrium (at 300 mg/kg bw/day). The reported NOAEL for maternal toxicity was determined to be 90 mg/kg bw/day based on clinical signs. The reported LOAEL for foetal effect was 30 mg/kg bw/day based on skeletal variation (REACH n.d.-c).

In a reproductive and developmental toxicity study (see **Repeated Dose Toxicity: Oral** section), SD rats (12/sex/dose) were administered benzotriazole via gavage at doses of 0, 30, 100 or 300 mg/kg bw/day (duration of treatment not available in the study). No effect on reproductive performances, implantation number, sex ratio and viability of pups was reported. The reported NOAEL values were determined to be 30 mg/kg bw/day for maternal

toxicity based on kidney effects at the higher dose, and 300 mg/kg bw/day for developmental toxicity (Government of Canada 2021).

Endocrine activity

A change in anogenital distance (although not consistent) was observed in prenatal development studies which can represent a biomarker for endocrine activity (although this may occur due to other causes such as body weight changes). Benzotriazole did not show receptor mediated oestrogenic and androgenic activity in in vitro studies. Although there was some evidence of anti-oestrogenic and anti-androgenic activity no conclusions can be made (ECHA 2023).

Human health risk characterisation

Critical health effects

The critical health effects for risk characterisation for these chemicals are potential developmental effects with a LOAEL of 30 mg/kg bw/day. By applying an assessment factor of 3 to account for LOAEL to NOAEL extrapolation an estimated NOAEL for risk characterisation is 10 mg/kg bw/day.

Public risk

The MOE methodology is commonly used to characterise risks to human health associated with exposure to chemicals (ECB 2003).

The MOE risk estimate provides a measure of the likelihood that a particular adverse health effect will occur under the conditions of exposure. As the MOE increases, the risk of potential adverse effects decreases. To decide whether the MOE is of sufficient magnitude, expert judgment is required. Such judgments are usually made on a case-by-case basis and should consider uncertainties arising in the risk assessment process such as the completeness and quality of available data, the nature and severity of effect(s) and intra/inter species variability. In general, an MOE value greater than or equal to 100 is considered acceptable to account for intra- and inter-species differences.

Based on the estimated daily systemic exposures (see **Human Exposure – Public**), the calculated MOE are:

- 370 from use of benzotriazole (nail products)
- 370 from automotive cooling system repair
- 147 from use in cleaning products.

References

CCOHS (Canadian Centre for Occupational Health and Safety) (2023a) *Record for 1H-Benzotriazole, CAS No. 95-14-7*, CCOHS, accessed 4 December 2023.

CCOHS (Canadian Centre for Occupational Health and Safety) (2023b) *Record for 1H-Benzotriazole, 5-methyl-, CAS No. 136-85-6*, CCOHS, accessed 4 December 2023.

CCOHS (Canadian Centre for Occupational Health and Safety) (2023c) *Record for* 1H-Benzotriazole, methyl-, *CAS No. 29385-43-1*, CCOHS, accessed 4 December 2023.

CCOHS (Canadian Centre for Occupational Health and Safety) (2023d) *Record for* 1H-Benzotriazole, 4(or 5)-methyl-, sodium salt, *CAS No. 64665-57-2*, CCOHS, accessed 4 December 2023.

Chemwatch (n.d.) *Galleria Chemica*, Chemwatch website, accessed 23 October 2023.

Danish EPA (Danish Environmental Protection Agency) (2013), *Benzotriazole and Tolytriazole, Evaluation of health hazards and proposal of health based quality criteria for soil and drinking water*, Danish EPA, accessed 23 October 2023.

Danish QSAR (Danish Environmental Protection Agency) (n.d.) *Danish QSAR*, accessed 23 October 2023.

DeLima Associates (n.d.) *Consumer Product Information Database*, DeLima Associates website, accessed 24 August 2024.

EC (European Commission) (n.d.) *CosIng*, EC website, accessed 23 October 2023.

ECB (European Chemicals Bureau) (2003) *Technical Guidance Document on Risk Assessment Part I*, ECB, accessed 17 August 2023.

ECHA (European Chemicals Agency) (2023) *Substance Evaluation Conclusion – Benzotriazole CAS No. 95-14-7*, ECHA, accessed 23 September 2024.

Government of Canada (2021) *Draft Screening Assessment Benzotriazoles and Benzothiazoles Group*, Environment and Climate Change Canada, Government of Canada, accessed 20 November 2023.

Lhasa Limited DEREK Nexus (version 6.0.1), [Computer software], Lhasa Limited, accessed 18 December 2023.

Lhasa Limited SARAH Nexus (version 3.1.0), [Computer software], Lhasa Limited, accessed 18 December 2023.

NTP (National Toxicology Program) (1978) *Bioassay of 1H-benzotriazole for possible carcinogenicity CAS No. 95-14-7*, NTP, accessed 20 November 2023.

OASIS LMC (Laboratory of Mathematical Chemistry) Optimised Approach based on Structural Indices Set–Tissue Metabolism Simulator (OASIS–TIMES) (Version 2.31), [Computer software], LMC, accessed 17 August 2024.

OECD (Organisation for Economic Co-operation and Development) (2020) *Quantitative Structure-Activity Relationship Toolbox (Version 4.5)* [Computer software], OECD website, assessed 18 December 2023.

Personal Care Products Council (n.d.) *Cosmetic Ingredient Identification Database*, Personal Care Products Council website, accessed 20 November 2023.

REACHa (Registration, Evaluation, Authorisation and Restriction of Chemicals) (n.d.) *Registered dossier for benzotriazole, CAS No. 95-14-7*, European Chemicals Agency website, accessed 20 November 2023.

REACHb (Registration, Evaluation, Authorisation and Restriction of Chemicals) (n.d.) *Registered dossier for 6-methylbenzotriazole, CAS No. 136-85-6*, European Chemicals Agency website, accessed 20 November 2023.

REACHc (Registration, Evaluation, Authorisation and Restriction of Chemicals) (n.d.) *Registered dossier for methyl-1H-benzotriazole, CAS No. 29385-43-1*, European Chemicals Agency website, accessed 20 November 2023.

REACHd (Registration, Evaluation, Authorisation and Restriction of Chemicals) (n.d.) *Registered dossier for sodium 4(or 5)-methyl-1H-benzotriazolide, CAS No. 64665-57-2*, European Chemicals Agency website, accessed 20 November 2023.

RIVM (The Dutch National Institute for Public Health and Environment) (n.d.) *ConsExpo*, RIVM website, accessed 23 September 2024.

RIVM (National Institute for Public Health and the Environment – Netherlands) (2018) *Cleaning Products Fact Sheet, Default parameters for estimating consumer exposure*, RIVM, accessed 23 September 2024.

SPIN (Substances in Preparation in Nordic Countries) (n.d.) *SPIN Database*, SPIN website, accessed 4 December 2023.

SWA (Safe Work Australia) (n.d.) *Hazardous Chemicals Information System*, SWA website, accessed 4 December 2023.

SWA (Safe Work Austalia) (2019) *Draft evaluation reports and recommendations – 1H benzotriazole*, SWA website, accessed 4 December 2023.

TGA (Therapeutic Goods Administration) (2024) *Therapeutic Goods (Poisons Standard—June 2024) Instrument 2024*, TGA, accessed 4 December 2023.

UNECE (United Nations Economic Commission for Europe) (2017) *Globally Harmonized System of Classification and Labelling of Chemicals (GHS), Seventh Revised Edition*, UNECE, accessed 19 December 2023.

US EPA (United States Environmental Protection Agency) (1977) *Investigation of Selected Potential Environmental Contaminants: Benzotriazoles – Final Report*, US EPA, accessed 23 October 2023.

US EPA (United States Environmental Protection Agency) (2016) *Access Chemical Data Reporting Data (CDR data)*, US EPA website, accessed 23 October 2023.

US EPA (United States Environmental Protection Agency) (2020), *Access Chemical Data Reporting Data (CDR data)*, US EPA website, accessed 17 August 2024.