Methylisothiazolinone preservatives and industrial biocides: Environment tier II assessment

16 June 2020

CAS Registry Numbers: 2682-20-4, 26172-54-3, 26172-55-4, 26530-03-0

Preface

This assessment was carried out by staff of the National Industrial Chemicals Notification and Assessment Scheme (NICNAS) using the Inventory Multi-tiered Assessment and Prioritisation (IMAP) framework.

The IMAP framework addresses the human health and environmental impacts of previously unassessed industrial chemicals listed on the Australian Inventory of Chemical Substances (the Inventory).

The framework was developed with significant input from stakeholders and provides a more rapid, flexible and transparent approach for the assessment of chemicals listed on the Inventory.

Stage One of the implementation of this framework, which lasted 4 years from 1 July 2012, examined 3000 chemicals meeting characteristics identified by stakeholders as needing priority assessment. This included chemicals for which NICNAS already held exposure information, chemicals identified as a concern or for which regulatory action had been taken overseas, and chemicals detected in international studies analysing chemicals present in babies' umbilical cord blood.

Stage Two of IMAP began in July 2016. We are continuing to assess chemicals on the Inventory, including chemicals identified as a concern for which action has been taken overseas and chemicals that can be rapidly identified and assessed by using

Stage One information. We are also continuing to publish information for chemicals on the Inventory that pose a low risk to human health or the environment or both. This work provides efficiencies and enables us to identify higher risk chemicals requiring assessment.

The IMAP framework is a science and risk-based model designed to align the assessment effort with the human health and environmental impacts of chemicals. It has 3 tiers of assessment, with the assessment effort increasing with each tier. The Tier I assessment is a high throughput approach using tabulated electronic data. The Tier II assessment is an evaluation of risk on a substance-by-substance or chemical category-by-category basis. Tier III assessments are conducted to address specific concerns that could not be resolved during the Tier II assessment.

These assessments are carried out by staff employed by the Australian Government Department of Health and the Australian Government Department of the Environment and Energy. The human health and environment risk assessments are conducted and published separately, using information available at the time, and may be undertaken at different tiers.

This chemical or group of chemicals are being assessed at Tier II because the Tier I assessment indicated that it needed further investigation.

For more detail on this program please visit: www.nicnas.gov.au.

Disclaimer

NICNAS has made every effort to assure the quality of information available in this report. However, before relying on it for a specific purpose, users should obtain advice relevant to their particular circumstances. This report has been prepared by NICNAS using a range of sources, including information from databases maintained by third parties, which include data supplied by industry. NICNAS has not verified and cannot guarantee the correctness of all information obtained from those databases. Reproduction or further distribution of this information may be subject to copyright protection. Use of this information without obtaining the permission from the owner(s) of the respective information might violate the rights of the owner. NICNAS does not take any responsibility whatsoever for any copyright or other infringements that may be caused by using this information.

Acronyms & Abbreviations

Grouping Rationale

This Tier II assessment considers the environmental risks associated with the industrial uses of a group of methylisothiazolinones. Chemicals in this group are widely used as preservatives in consumer products, including personal care products and cosmetics. They are also used as industrial biocides in facilities such as fuel storage systems and industrial air-conditioning plants.

The known industrial uses for the chemicals in this group will lead to their release into sewers which may result in their emission to the aquatic environment in the treated effluents produced by sewage treatment plants (STPs). This is of potential environmental concern because chemicals with preservative and biocidal properties are often very toxic to aquatic life. This assessment will evaluate the potential for emissions of methylisothiazolinones to the aquatic environment in Australia and whether risk reduction measures are required for industrial uses of these chemicals.

The methylisothiazolinones in this assessment belong to a larger group of preservatives and industrial biocides which all have an isothiazolinone heterocyclic ring system. The other members of this group that are listed on the Inventory are the two octylisothiazolinones, 2-octyl-3-isothiazolinone (OIT, CAS RN 26530-20-1) and 4,5-dichloro-2-octyl-3-isothiazolinone (DCOIT, CAS RN 64359-81-5), and 1,2-benzisothiazolinone (BIT, CAS RN 2634-33-5). These 3 other isothiazolinone preservatives and industrial biocides will be assessed separately.

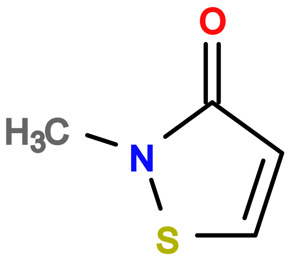
Chemical Identity

The chemicals in this group have a common unsaturated 5-membered heterocyclic ring which has an *endo*-cyclic sulfur atom bonded to an *endo*-cyclic methylated nitrogen atom. The group includes the parent chemical, 3-isothiazolone, 2-methyl- (MIT), and its monochloro- derivative (CMIT). The hydrochloride salts of MIT and CMIT have industrial uses and they are also included in this group.

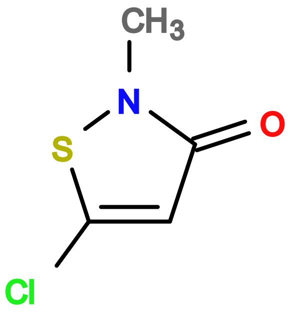
Methylisothiazolinones are made industrially by oxidative cyclisation of the linear organic di-sulfide, *N*,*N′*-dimethyl-3,3′-dithiodipropionamide (CAS RN 999-72-4), in a process that uses chlorine as the oxidant (Burnett, et al., 2010, Lewis, et al., 1978). This manufacturing process inevitably produces a mixture of MIT and CMIT, as well as a small amount of the dichloro-derivative (DCMIT; CAS RN 26542-23-4). These mixtures are generally not separated into their constituent chemicals and CMIT is not commercially available except as a mixture with MIT (TGA, 2015); DCMIT is an incidental impurity in these mixtures (Alvarez-Sanchez, et al., 2003) and it has no identified industrial uses. It is noted that the Chemical Abstracts Service has assigned a CAS RN (55965-84-9) to mixtures of MIT and CMIT. This substance is not listed on the Inventory.

MIT is available for industrial uses separate from its two chlorinated derivatives. It is obtained by purification of technical mixtures (Dow, 2015).

Due to the rapid conversion of the hydrochloride salts to the neutral compounds at environmental pH, these salts are not separately considered in this assessment. The exposure assessment of the neutral species include the contribution from use of the salts.

CAS RN	2682-20-4
Chemical Name	3-Isothiazolone, 2-methyl-
Synonyms	MIT MI 2-methyl-4-isothiazolin-3-one
Structural Formula	
Molecular Formula	C_4H_5NOS
Molecular Weight (g/mol)	115.15
SMILES	CN1C(=O)C=CS1

CAS RN	26172-54-3
Chemical Name	3(2*H*)-Isothiazolone, 2-methyl-, hydrochloride
Synonyms	MIT.HCl
Molecular Formula	C_4H_6ClNOS

CAS RN	26172-55-4
Chemical Name	3(2*H*)-Isothiazolone, 5-chloro-2-methyl-
Synonyms	CMIT MCI 5-chloro-2-methyl-4-isothiazolin-3-one
Structural Formula	
Molecular Formula	C_4H_4ClNOS
Molecular Weight (g/mol)	149.6
SMILES	CN1C(=O)C=C(S1)Cl

CAS RN	26530-03-0
Chemical Name	3(2*H*)-Isothiazolone, 5-chloro-2-methyl-, hydrochloride
Synonyms	CMIT.HCl
Molecular Formula	$C_4H_5Cl_2NOS$

Physical and Chemical Properties

The information on physical form and the experimental values for the octanol-water partition coefficient (K_{ow}) of MIT and CMIT were taken from the peer-reviewed scientific literature (Arning, et al., 2009). An experimental value for the water solubility of CMIT was obtained from a European Commission report (SCCS, 2009), whereas the solubility of MIT was calculated using a standard quantitative structure-property relationship (QSPR) (US EPA, 2008). The remaining physical and chemical property data for MIT and CMIT were retrieved from the Hazardous Substances Data Bank (HSDB) of the United States (US) National Library of Medicine (HSDB, 2015a, b):

Chemical	MIT	CMIT
Physical Form	solid	solid
Melting Point	50–51°C (exp.)	54–55°C (exp.)
Boiling Point	93°C at 4 Pa (exp.)	262°C (calc.)
Vapour Pressure	8.26 Pa (exp.)	2.40 Pa (exp.)
Water Solubility	959 g/L (calc.)	706–751 g/L (exp.)
Ionisable in the Environment?	no	no
log K_{ow}	-0.49 (exp.)	0.53 (exp.)

The methylisothiazolinones in this group are very weak organic bases (Lindner, 1986). The free-base forms of the 2 chemicals in this group (MIT and CMIT) are not expected to be ionised in environmental surface waters where the pH is typically in the range from 4 to 9.

MIT and CMIT are volatile organic chemicals which are readily soluble in water based on measured or calculated values for these properties. The low measured K_{ow} values for both MIT and CMIT indicate that both chemicals have a low tendency to partition from water into octanol and that they are not lipophilic.

Import, Manufacture and Use

Australia

The volume of CMIT introduced annually is in the range of 100 to 1000 tonnes based on information reported to NICNAS. No specific introduction volume information has been identified for the remaining chemicals in this group.

MIT and CMIT are used as preservatives to prevent or inhibit microbial growth in consumer products. They have been reported to be widely used in Australia as biocidal preservatives in cosmetics, personal care products, baby wipes, cleaning and laundry products (NICNAS, 2018b). MIT and CMIT has reported uses in consumer and industrial sealants and in marine and automotive aftermarket products such as waxes, polishes and coatings. Domestic calls for information have revealed that MIT is used in car wash soap and floor protection and finish coatings, and industrial coatings (NICNAS, 2018a).

According to industry information, MIT, CMIT and CMIT.HCl are used as preservatives in paint formulations. MIT and CMIT are also used in sealants (consumer and industrial) and in marine and automotive aftermarket products, such as waxes, polishes and coatings.

Commercial mixtures of MIT and CMIT are available for use as biocides in industrial circulating cooling water systems, and as preservatives in papermaking, leather treatment and cutting fluids (Lonza Water Treatment, 2018).

MIT and CMIT are used in domestic coal seam gas (CSG) applications (Commonwealth of Australia, 2014). However, release from this use is beyond the scope of this assessment.

MIT and CMIT are used as excipients in therapeutic goods such as biological products, devices, export only products, listed medicines, over-the-counter and prescription medicines. They are also used as active pharmaceutical ingredients in biological products and prescription medicines (TGA, 2017). These non-industrial uses are outside the scope of this assessment.

The Therapeutic Goods Administration has listed MIT and CMIT on Schedule 6 of the Poisons Standard (TGA, 2019). The listing for methylchloroisothiazolininone (CMIT) includes exemptions for rinse-off cosmetics containing 0.0015% or less of MIT and CMIT in total and non-topical preparations containing 0.1% or less of MIT and CMIT in total. The listing for methylisothiazolinone (MIT) includes exemptions for topical rinse-off cosmetics containing 0.01% or less of MIT and non-topical preparations containing 0.1% or less of MIT; however, the exemption for use of MIT in topical rinse-off cosmetics is due to be amended to 0.0015% or less of MIT by 1 October 2019 (TGA, 2017).

International

Mixtures of MIT and CMIT are used as preservatives in many consumer products, including personal care products, disinfectants, and perfumes and fragrances (REACH, 2018). They are also used for this purpose in paints, lacquers, adhesives, surface treatments, fillers, corrosion inhibitors, construction materials, colouring agents, photochemicals, pH regulators, absorbents, surface active agents, lubricants and additives (Nordic Council of Ministers, 2016). Mixtures of MIT and CMIT are also used as a biocide in fuel storage systems and as a preservative in fuel to prevent microbial spoilage (Dow, 2009).

MIT and CMIT are listed on the US EPA and OECD High Production Volume chemical lists indicating production of over 454 tonnes per year (US) or 1000 tonnes per year (OECD) (OECD, 2009, US EPA, 2013). MIT and CMIT are each imported or manufactured in the European Union in volumes in the range of 10–100 tonnes per year (REACH, 2018).

MIT and CMIT are also used in agricultural pesticides and products. Such non-industrial uses are outside the scope of this assessment.

Environmental Regulatory Status

Australia

The use of the chemicals in this group is not subject to any specific national environmental regulations.

Industrial chemicals used as biocides in cooling systems are subject to regulation in some states. Isothiazolones are used in cooling tower water and large air-conditioning systems to inhibit the growth of potentially harmful microbes such as the *Legionella* family of bacteria which cause Legionnaires' disease in humans. Treated cooling tower wastewater is considered a liquid trade waste and its disposal is subject to state government guidelines and authorisation (WA Department of Water, 2007). The guidelines specify that cooling tower water should not be disposed into waterways or stormwater drainage but into the sewer. Approval from the relevant water authority is required before discharge to sewer.

United Nations

No chemicals in this group are currently identified as a Persistent Organic Pollutant (UNEP, 2001), an ozone depleting substance (UNEP, 1987), or hazardous substance for the purpose of international trade (UNEP & FAO, 1998).

OECD

MIT and CMIT have both been identified as high-production volume chemicals by the OECD, which indicates that more than 1000 tonnes of each chemical are produced annually in at least 1 member country (OECD, 2009).

No chemicals in this group have been sponsored for assessment under the Cooperative Chemicals Assessment Programme (OECD, 2017).

Canada

MIT, CMIT and their hydrochloride salts are listed on the Canadian Domestic Substances List (DSL) as existing substances already in commerce in Canada (Environment and Climate Change Canada, 2018).

European Union

MIT and MIT.HCl are registered under the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) legislation (ECHA, 2017c). CMIT and CMIT.HCl are pre-registered under REACH (ECHA, 2017b).

MIT is listed on the Public Activities Coordination Tool (PACT) as appropriate for informal hazard assessment and/or risk management option analysis under the SVHC Roadmap due to its potential as a skin sensitiser (ECHA, 2017a).

MIT and CMIT are approved for use under the EU Biocidal Products Regulation (ECHA, 2018). The regulation authorises the manufacture and use of biocidal and preservative chemicals on the European market, with the aim of high level consumer and environmental protection. These 2 chemicals are currently approved for use in the following product types: PT11: preservatives in liquid cooling and processing systems; PT12: slimicides; and PT13: metal working fluids. MIT and CMIT are under review for use in the following product type: PT6: in-can preservatives.

MIT and mixtures of MIT/CMIT are listed on the European Commission's Cosmetic Ingredient database (CosIng) and on Annex V with the restriction to be used at a maximum concentration of 0.0015% in rinse off cosmetic products (European Commission, 2017).

United States of America

MIT and CMIT are both listed on the United States Environmental Protection Agency (US EPA) Chemical Substance Inventory, established under the Toxic Substances Control Act 1976. Both chemicals are registered as ‘active’ on the Chemical Substance Inventory, which indicates that they have recently been manufactured, imported or processed by industry in the USA (US EPA, 2018a). They are also both listed on the US EPA Safer Chemical Ingredients List (US EPA, 2018b). This list identifies chemicals deemed to be safer alternatives to other chemicals employed for the same functional use. The criteria for adding chemicals to the list cover a broad range of human health and environmental toxicological effects.

Environmental Exposure

MIT and CMIT are synthetic chemicals and their occurrence in the environment results exclusively from human activity. They can be emitted to the environment as the result of both industrial and non-industrial uses. Emissions to the environment from industrial uses can occur from point sources such as STPs and from diffuse sources. The diffuse sources include external hard surfaces of buildings that were coated with paints that contain these chemicals as preservatives. These readily water soluble preservatives can be released from coated surfaces as a result of leaching by rainwater.

Methylisothiazolinones used as preservatives in consumer products such as personal care and cosmetic products will be released down the drain to sewers in domestic wastewater as a normal part of this use pattern. Treatment of this wastewater in sewage treatment plants will remove some fraction of the quantity of these chemicals in influents, depending on the efficiency of various degradation and partitioning processes. Based on the very high water solubilities of MIT and CMIT, these chemicals are unlikely to be removed in STPs by partitioning process (volatilisation or adsorption to sludge). However, up to 67% may be removed by biodegradation (Struijs, 1996). Additional quantities of these chemicals may be removed by abiotic degradation processes that are not included in standard models. Hence, the removal efficiency of these chemicals by standard sewage treatment processes is likely to be greater than the 67% based on biodegradation alone; however, the overall removal efficiency cannot be reliably estimated. For the purposes of this assessment it is assumed that that these chemicals are not completely removed from wastewater and that emissions to surface waters in treated effluents are possible.

There is also potential for more concentrated releases of methylisothiazolinones to wastewater as a result of their use as industrial biocides in cooling systems. Treated cooling water is used to dissipate heat in a range of industrial processes (Dow, 2019, WA Department of Water, 2007). In closed system cooling towers there is often a need to expel a portion of the cooling water to remove dissolved solids and other impurities. This expelled volume of liquid is called ‘blow-down’ and forms a possible emission pathway for isothiazolinone biocides. Treated water is also used to flush closed hydraulic systems to eliminate microbial build-up as part of regular maintenance (Australian Standard, 2011). The typical methylisothiazolinone exposure concentrations that are required in these applications are significantly higher than those which are required for use in consumer products, and spent treated cooling water is therefore expected to be more hazardous to the aquatic environment. However, management of spent treated cooling water should be according to disposal guidelines, which directs users to apply for authorisation to dispose of the water to the sewer (WA Department of Water, 2007). As disposal of spent cooling water in stormwater drains or directly to waterways is prohibited, environmental emissions of the chemicals in this group directly to surface waters from their use as biocides in cooling water systems is expected to be limited.

Isothiazolinone preservatives will slowly leach from painted surfaces and some surface treated construction materials exposed to the weather through the action of rainfall (Bollmann, et al., 2014). The surface run-off containing these leached chemicals can be discharged directly onto soil or indirectly into surface waters through the discharge of stormwater from drainage systems. Emissions from these sources cannot be quantified currently, but they will contribute to cumulative diffuse emissions of these chemicals into the environment.

Environmental Fate

Partitioning

The chemicals in this group partition to water or soil when released into the environment.

The chemicals in this group are neutral organic chemicals that are readily soluble in water. The calculated Henry's Law constants for the partitioning of these chemicals between air and water are 0.0036 and 0.005 Pa-m^3/mol for CMIT and MIT respectively (US EPA, 2008). These low values indicate that both chemicals are only very slightly volatile from water and moist soil. The calculated organic carbon normalised soil adsorption coefficients (K_{oc}) for both chemicals are low (log K_{oc} = 1.08 and 1.28 for MIT and CMIT respectively) (US EPA, 2008). These very low K_{oc} values indicate that both chemicals will have very high mobility in soil, assuming that there are no specific interactions between the chemicals and soil.

Calculations with a standard multimedia partitioning (fugacity) model assuming equal and continuous distributions to air, water and soil compartments (Level III approach) predict that the chemicals will mainly partition to the soil (65% MIT, 61% CMIT) and water (34% MIT, 38% CMIT) compartments in the environment. However, in a scenario where release only occurs to water, 99% of the chemical remains in the water compartment (US EPA, 2008).

Degradation

The chemicals in this group are biodegradable at expected environmental exposure concentrations.

Biodegradation studies on MIT and CMIT have been hindered by their toxicity to bacteria (Madsen, et al., 2001). Tests conducted on CMIT with OECD Test Guideline (TG) 301C for ready biodegradability at a test concentration of 100 mg/L showed 0% degradation of the chemical after 28 days (NITE, 2017). A test on MIT.HCl using OECD TG 301B at a test concentration of 31.6 mg/L showed 0% degradation over 29 days (REACH, 2018).

The apparent non-degradability of these biocides in standard biodegradability screening tests is a result of their toxicity to microbes in activated sludge innocula. When the tests are conducted at lower exposure concentrations the chemicals are biodegraded. A respirometric CO_2 evolution test (OECD TG 301B) on MIT at test concentrations of 0.1, 0.03 and 0.01 mg/L showed 54%, 56% and 48% mineralisation over 29 days, respectively (Madsen, et al., 2001). The same test with CMIT at test concentrations of 0.3, 0.1 and 0.03 mg/L showed 39%, 55% and 62% mineralisation, respectively (Madsen, et al., 2001). The rate of biodegradation in these tests does not satisfy the OECD criterion for readily biodegradability (60% in a 10 day window), but the results do show that these chemical are biodegradable at more realistic environmental exposure concentrations.

Degradation studies on MIT in soil showed rapid primary biodegradation. Monitoring of the lifetime and degradation products of MIT in soil showed that the primary degradation half-life is 6.7 h at a test concentration of 2 micrograms (µg) of biocide per gram of soil (Bollmann, et al., 2017). The study also measured rates of degradation for 2-octylisothiazolone (OIT) and 4,5-dichloro-2-octylisothiazolone (DCOIT) which both underwent primary degradation with half-lives under 10 days. It is expected that CMIT will also undergo rapid primary biodegradation in soil based on the findings for these three isothiazolinones.

The principal environmental degradation pathway for MIT and CMIT involves ring opening and loss of sulfur and chlorine atoms to form *N*-methylmalonamic acid (CAS RN 42105-98-6) as a metabolite (Krzeminski, et al., 1975). A similar ring opening mechanism has been proposed for the degradation pathway of OIT in soil (Bollmann, et al., 2017).

MIT and CMIT are hydrolytically stable based on hydrolysis measurements made using OECD TG 111. The hydrolysis half-lives at 25°C and pH = 7.4 for MIT and CMIT are >80 and 69 days respectively (Arning, et al., 2009).

Although the methylisothiazolinones in this group are stable towards hydrolysis, they can react with a range of naturally occurring nucleophilic chemicals such as thiols and other chemicals with a reduced sulfur functional group (Alvarez-Sanchez, et al., 2003, Williams and McGinley, 2010). This abiotic degradation pathway is likely to be significant in natural waters and sewage treatment plants where thiols are abundant (Krzeminski, et al., 1975).

Bioaccumulation

The chemicals in this group are not expected to bioaccumulate.

Studies of the bioconcentration of MIT and CMIT in bluegill sunfish (*Lepomis macrochirus*) at an exposure concentration of 0.12 mg/L showed bioconcentration factors (BCF) in this species of 2.3 and 114 L/kg respectively (Madsen, et al., 2001). These BCF values are less than the domestic categorisation criterion for bioaccumulation (BCF ≥ 2000 L/kg). The octanol-water partition coefficient for these hydrophilic chemicals are also less than the domestic categorisation threshold for bioaccumulation hazards in aquatic organisms (log $K_{ow} \geq 4.2$).

The low bioconcentration potential, hydrophilicity, and the reactivity of both chemicals with biomolecules indicate that they will not biomagnify in aquatic or terrestrial food webs.

Transport

The chemicals in this group are not expected to undergo long range transport based on their short-half lives in the environment.

Predicted Environmental Concentration (PEC)

The estimated environmental concentration of MIT and CMIT in effluents from STPs are 0.04 and 0.03 µg/L respectively. No Australian environmental monitoring data were identified for these chemicals.

The exposure scenarios considered in this assessment involve emissions of MIT and CMIT to surface waters in STP effluent and leaching of these chemicals from the surfaces of buildings. International monitoring studies indicate that the removal of these chemicals from wastewater in STPs is highly effective (Raforth, et al., 2007, Speksnijder, et al., 2010). These studies show that methylisothiazolinones could not be detected in STP effluent above the respective limits of detection for the analytical method, which are 0.04 µg/L for MIT and 0.03 µg/L for CMIT. In the absence of other information, these limits of detection have been taken to provide a conservative upper estimate of the maximum concentrations of methylisothiazolinones in effluents from STPs in Australia.

International monitoring of MIT concentrations in stormwater run-off indicate the potential for leaching of this chemical from building facades. In 1 study, for every rain event, an average of 11 µg of MIT was emitted per house in the suburban catchment area considered in the study (Bollmann, et al., 2014). Emission rates fluctuated with rain intensity and the highest measured MIT concentration was 0.162 micrograms per litre (µg/L) in stormwater. However, emissions into stormwater are irregular and dependent on rain volumes and other weather effects. MIT was detected in stormwater on only a few occasions which suggests that if leaching from buildings does occur in Australia it will be an intermittent and diffuse source of low level emissions of these biocides to surface waters in urban areas.

Environmental Effects

Effects on Aquatic Life

MIT and CMIT are very toxic to unicellular organisms including algae and bacteria.

The majority of the publically available ecotoxicity data for methylisothiazolinones is for Kathon (i.e., mixtures of MIT and CMIT), which is a commercial formulation that typically contains stabilising additives (e.g., magnesium, calcium or copper salts) which are not always disclosed (HSDB, 2015a, US EPA, 1992). The effects of these additives which can include ecotoxicologically significant copper(2+) ions cannot be separated from the combined toxicity of MIT and CMIT. The ecotoxicity of these commercial formulations is more relevant to considerations of the environmental effects of mixtures of MIT and CMIT used as pesticides and has, therefore, not been relied on in this assessment.

Acute toxicity

The following measured median effective concentration (EC50) values for freshwater model organisms across two trophic levels were taken from peer-reviewed scientific publications (Arning, et al., 2009, Kresmann, et al., 2018):

Taxon	Endpoint	Method
Invertebrates	MIT: 48 h EC50 = 0.51 mg/L	Experimental *Daphnia magna* OECD TG 202
Algae	MIT: 72 h EC50 = 0.56 mg/L CMIT: 72 h EC50 = 0.089 mg/L	Experimental *Scenedesmus vacuolatus* (green algae) ISO 8692:1989 Freshwater algal growth inhibition
Bacteria	MIT: EC50 = 1.61 mg/L CMIT: EC50 = 0.086 mg/L	Experimental *Vibrio fischeri* (bioluminescent bacteria) DIN EN ISO 11348-2:2007 Freshwater bacterial luminescence inhibition

MIT and CMIT have a specific mode of toxicity related to their electrophilic reactivity. The isothiazolinone moiety common to all chemicals in this group undergoes a ring opening reaction at the electrophilic sulfur atom with a range of biological nucleophiles (Alvarez-Sanchez, et al., 2003). The chemicals diffuse across cellular membranes where they react with nucleophilic thiol-containing proteins or thiol-containing biomolecules like glutathione. Reaction with proteins inhibits the enzymes critical to respiration which slows or stops cellular growth (Williams, 2007). Reaction with glutathione can rapidly disrupt the cellular reduction potential leading to oxidative stress and reduced cellular viability (Arning, et al., 2009, Williams, 2007). Unicellular organisms are more susceptible to these stressors than higher order organisms. The most sensitive trophic level is therefore expected to be algae and other unicellular organisms.

CMIT is more toxic to algae and bacteria than MIT. This greater toxicity has been linked to both the higher reactivity of CMIT to nucleophiles (Arning, et al., 2009), and to the formation of an additional highly reactive thioacyl chloride intermediate, which is reactive with cellular nucleophiles other than thiols (Alvarez-Sanchez, et al., 2003).

Chronic toxicity

No reliable chronic toxicity data were available for MIT or CMIT.

Predicted No-Effect Concentration (PNEC)

The PNECs for MIT and CMIT are 0.51 and 0.089 µg/L respectively.

The invertebrate 48 h EC50 value of 0.51 mg/L for MIT was used to derive the PNEC for this chemical. For CMIT, the 72 h EC50 value of 0.089 mg/L for algae was used. An assessment factor of 1000 was used in both cases as there are incomplete acute aquatic toxicity data available for both chemicals.

Categorisation of Environmental Hazard

The categorisation of the environmental hazards of the chemicals in this group according to domestic environmental hazard thresholds is presented below (EPHC, 2009, NICNAS, 2017):

Persistence

Not Persistent (Not P). Based on the rates of abiotic and biotic degradation of MIT and CMIT at environmentally relevant exposure concentrations, they are both categorised as Not Persistent.

Bioaccumulation

Not Bioaccumulative (Not B). Based on the low bioconcentration factors for MIT and CMIT in fish they are categorised as Not Bioaccumulative.

Toxicity

Toxic (T). MIT and CMIT have acute toxicity values below 1 mg/L indicating they are toxic to aquatic organisms.

Summary

3-Isothiazolone, 2-methyl- (MIT); 3(2*H*)-isothiazolone, 2-methyl-, hydrochloride (MIT.HCl); 3(2*H*)-isothiazolone, 5-chloro-2-methyl- (CMIT); and 3(2*H*)-isothiazolone, 5-chloro-2-methyl-, hydrochloride (CMIT.HCl) are categorised as:

- Not P
- Not B
- T

Risk Characterisation

Methylisothiazolinones are very toxic to unicellular organisms including aquatic primary producers such as algae. This toxicity is necessary for their functional use as preservatives and industrial biocides which involves controlling the growth of unicellular organisms such as bacteria, fungi and algae. These industrial uses also have the potential to release these biocides directly or indirectly to the aquatic environment which is of potential concern. The highest concern scenario involves discharge of large volumes of spent industrial cooling water that has been treated with these biocides.

Methylisothiazolinones can potentially enter the aquatic environment in STP effluent as a result of their use as preservatives in personal care products and through use as biocides in industrial cooling systems. Based on the PEC and PNEC values determined above, the following Risk Quotients (RQ = PEC ÷ PNEC) have been calculated for release of MIT and CMIT into the riverine environment:

Chemical	PEC (µg/L)	PNEC (µg/L)	RQ
MIT	0.04	0.51	0.078
CMIT	0.03	0.089	0.337

An RQ of less than 1 indicates that MIT and CMIT are not expected to pose an unreasonable risk to the environment, as environmental concentrations are below the levels likely to cause harmful effects. There is uncertainty in these calculations as the PECs are conservative upper estimates based on very limited international environmental monitoring data. An additional uncertainty is the total quantity of both MIT and CMIT that is used annually in Australia for industrial uses. It is noted that forthcoming changes to the Poisons Standard will decrease the allowable concentrations of MIT in some consumer products. This may eventually result in an overall decline in releases of this chemical to sewers and a reduction in environmental emissions from STPs in Australia.

The disposal of spent cooling water treated with biocides is of potentially high concern for the aquatic environment because the typical concentrations of MIT and CMIT used in these applications are up to 15 mg/L, which is more than 160 times the lethal concentration for algae and bacteria. Hence, direct release of cooling waters to natural waterways poses a risk of severe adverse effects on aquatic ecosystems.

The environmental risks associated with the use of biocides in industrial cooling water systems are recognised by state governments and they prohibit unauthorised release of treated water to surface waters and stormwater drains. Cooling waters must, instead, be released to sewers after appropriate approvals have been obtained. Additional approvals are required from wastewater managers as high concentrations of biocides can impact the biological processes operating in STPs. Adherence to these guidelines and requirements for approval to appropriately dispose of these biocides as trade waste is expected to be sufficient to manage the risk both to the operation of STPs and to the aquatic environment.

Key Findings

Methylisothiazolinones are preservatives and biocides. They are used as preservatives in a wide range of household products and cosmetics on the Australian market. They are also used as industrial biocides for cooling water in industrial air conditioning plants.

MIT and CMIT are very toxic to aquatic primary producers such as algae and they pose a risk to aquatic ecosystems if emitted in high concentrations. The risks to the aquatic environment associated with indirect emissions of these chemicals in treated effluent from STPs is not considered to be of concern based on the available information.

The risks posed to the aquatic environment by the use of MIT and CMIT as industrial biocides in cooling water is expected to be managed by current state government water authority guidelines on liquid trade waste disposal. Standard practice is that spent cooling water treated with isothiazolinone biocides are disposed to sewer after permits are obtained from the relevant waste water authority. Adherence to these guidelines and the use of additional precautions where appropriate such as the use of specific deactivation protocols recommended by manufacturers is expected to be sufficient to manage any risks to STPs and the aquatic environment.

The chemicals in this group are not PBT substances according to domestic environmental hazard criteria.

Recommendations

No further assessment is currently required under the IMAP Framework.

Environmental Hazard Classification

In addition to the categorisation of environmental hazards according to domestic environmental thresholds presented above, the classification of the environmental hazards of 3-isothiazolone, 2-methyl- (MIT); 3(2*H*)-isothiazolone, 2-methyl-, hydrochloride (MIT.HCl); 3(2*H*)-isothiazolone, 5-chloro-2-methyl- (CMIT); and 3(2*H*)-isothiazolone, 5-chloro-2-methyl-, hydrochloride (CMIT.HCl) according to the third edition of the United Nations' Globally Harmonised System of Classification and Labelling of Chemicals (GHS) is presented below (UNECE, 2009):

Hazard	GHS Classification (Code)	Hazard Statement
Acute Aquatic	Category 1 (H400)	Very toxic to aquatic life

Hazard	GHS Classification (Code)	Hazard Statement
Chronic Aquatic	Category 1 (H410)	Very toxic to aquatic life with long lasting effects

The classification of the aquatic hazards of the chemicals in this group was performed based on the acute toxicity data presented in this assessment. The EC50 values available for MIT and CMIT are < 1 mg/L and they are not considered to be rapidly degradable in aquatic ecosystems for the purposes of this aquatic hazard classification.

References

Alvarez-Sanchez R, Basketter D, Pease C and Lepoittevin J-P (2003). Studies of Chemical Selectivity of Hapten, Reactivity, and Skin Sensitization Potency. 3. Synthesis and Studies on the Reactivity toward Model Nucleophiles of the 13C-Labeled Skin Sensitizers, 5-Chloro-2-methylisothiazol-3-one (MCI) and 2-Methylisothiazol-3-one (MI). *Chemical Research in Toxicology*, **16**, pp 627-636.

Arning J, Matzke M, Stolte S, Nehen F, Bottin-Weber U, Boschen A, Abdulkarim S, Jastorff B and Ranke J (2009). Analyzing Cytotoxic Effects of Selected Isothiazol-3-one Biocides Using the Toxic Ratio Concept and Structure-Activity Relationship Considerations. *Chemical Research in Toxicology*, **22**, pp 1954-1961.

Australian Standard (2011). *Australian/New Zealand Standard 3666.2 - Air-handling and water systems of buildings – Microbial control, Part 2: Operation and maintenance*.

Bollmann U, Vollertsen J, Carmeliet J and Bester K (2014). Dynamics of Biocide Emissions From Buildings in a Suburban Stormwater Catchment - Concentrations, Mass Loads and Emission Processes. *Water Research*, **56**, pp 66-76.

Bollmann U, Ferna´ndez-Calvin~o D, Brandt K, Storgaard M, Sanderson H and Bester K (2017). Biocide Runoff from Building Facades: Degradation Kinetics in Soil. *Environmental Science and Technology*, **51**, pp 3694-3702.

Burnett C, Bergfeld W, Belsito D, Klaassen C, Marks J, Shank R, Slaga T, Snyder P and Anderson A (2010). Final Report of the Safety Assessment of Methylisothiazolinone. *International Journal of Toxicology*, **29**, pp 1875-2135.

Commonwealth of Australia (2014). Technical Report number 14: Environmental Risks Associated With Surface Handling of Chemicals Used in Coal Seam Gas Extraction in Australia. Department of the Environment and Energy, Canberra. http://www.environment.gov.au.

Dow (2009). *Kathon FP 1.5 Fuel Biocide MSDS*. The Dow Chemical Company, Accessed at http://msdssearch.dow.com.

Dow (2015). *DOW™ Methylisothiazolinone (MIT) Antimicrobial Products*. The Dow Chemical Company, Accessed at http://msdssearch.dow.com.

Dow (2019). KATHON™ WT Water Treatment Microbicide. The Dow Chemical Company. Accessed 1 March 2019 at http://msdssearch.dow.com.

ECHA (2017a). PACT - RMOA and hazard assessment activities. European Chemicals Agency, Helsinki, Finland. Accessed 10 November 2017 at https://echa.europa.eu.

ECHA (2017b). Pre-registered substances. European Chemicals Agency, Helsinki, Finland. Accessed 10 November 2017 at https://echa.europa.eu.

ECHA (2017c). Registered substances. European Chemicals Agency, Helsinki, Finland. Accessed 10 November 2017 at https://echa.europa.eu.

ECHA (2018). Biocidal Products Directive: Information on Biocides. European Chemicals Agency, Helsinki, Finland. Accessed 21 December 2018 at https://echa.europa.eu.

Environment and Climate Change Canada (2018). Search Engine for the Results of DSL Categorisation. Environment and Climate Change Canada, Gatineau, Quebec, Canada. Accessed 18 April 2018 https://pollution-waste.canada.ca.

EPHC (2009). Environmental Risk Assessment Guidance Manual for Industrial Chemicals. The Department of Environment, Canberra, Australia. Accessed 10 November 2017 at http://www.nepc.gov.au.

European Commission (2017). Cosmetic Ingredient Database (CosIng). European Commission, Brussels, Belgium. Accessed 10 November 2017 at http://ec.europa.eu.

HSDB (2015a). Hazardous Substances DataBank: 5-CHLORO-2-METHYL-4-ISOTHIAZOLIN-3-ONE. US National Library of Medicine, Bethesda, MD 20894. Accessed 29 November 2018 at https://toxnet.nlm.nih.gov.

HSDB (2015b). Hazardous Substances DataBank: METHYLISOTHIAZOLINONE. US National Library of Medicine Bethesda, MD 20894. Accessed 29 November 2018 at https://toxnet.nlm.nih.gov.

Kresmann S, Arokia A, Koch C and Sures B (2018). Ecotoxicological Potential of the Biocides Terbutryn, Octhilione and Methylisothiazolinone: Underestimated Risk from Biocidal Pathways? *Science of the Environment*, **625**, pp 900-908.

Krzeminski S, Brackett C, Fisher J and Spinnler J (1975). Fate of Microbiocidal 3-Isothiazolone Compounds in the Environment: Products of Degradation. *Journal of Agricultural and Food Chemistry*, **23**(6), pp 1068-1075.

Lewis S, Miller G and Law A (1978), *3-Isothiazolones as Biocides*. Rohm and Haas Company, USA. Accessed March 2019.

Lindner W (1986), *Patent EP0271761A1: Process for the isolation of isothiazolinone derivatives*. European Patent Office, Germany. Accessed 16 March 2020 at https://patents.google.com.

Lonza Water Treatment (2018). Isothiazolinones. Lonza Water Treatment. Accessed 29 November 2018 at http://www.lonzawatertreatment.com.au.

Madsen T, Buchardt Boyd H, Nylén D, Rathmann Pedersen A, Petersen G and Simonsen F (2001). *Environmental and Health Assessment of Substances in Household Detergents and Cosmetic Detergent Products*. DEP Agency, Copenhagen, Denmark. Accessed at https://www2.mst.dk.

NICNAS (2017). Inventory Multi-tiered Assessment and Prioritisation (IMAP) Framework. National Industrial Chemicals Notification and Assessment Scheme, Sydney, Australia. Accessed 27 November 2017 at https://www.nicnas.gov.au.

NICNAS (2018a). 3-Isothiazolone, 2-methyl-: Human health tier II assessment. Accessed 20 December 2018 at https://www.nicnas.gov.au.

NICNAS (2018b). 3(2H)-Isothiazolone, 5-chloro-2-methyl-: Human health tier II assessment. Accessed 29 November 2018 at https://www.nicnas.gov.au.

NITE (2017). Japan CHEmicals Collaborative Knowledge Database (J-CHECK). National Institute of Technology and Evaluation, Tokyo, Japan. Accessed 6 July 2017 http://www.safe.nite.go.jp.

Nordic Council of Ministers (2016). Substances in Preparations in Nordic Countries (SPIN). Chemical Group, Nordic Council of Ministers, Copenhagen, Denmark. Accessed 1 March 2016 at http://www.spin2000.net.

OECD (2009). *The 2009 OECD List of High Production Volume Chemicals*. Paris. Accessed at http://www.oecd.org.

OECD (2017). OECD Existing Chemicals Database. Organisation for Economic Cooperation and Development, Paris, France. Accessed 10 November 2017 at http://webnet.oecd.org.

Raforth A, Gabriel S, Sacher F and Brauch H-J (2007). Analysis of Isothiazolinones in Environmental Waters by Gas Chromatography–mass Spectrometry. *Journal of Chromotography A*, **1164**, pp 74-81.

REACH (2018). Registration, Evaluation, Authorisation and Restriction of Chemicals Breif Profile: 2-methyl-2H-isothiazol-3-one, 2-methyl-2H-isothiazol-3-one hydrochloride and 5-chloro-2-methyl-2H-isothiazol-3-one. European Chemicals Agency, Helsinki, Finland. Accessed 29 November 2018 at https://echa.europa.eu.

SCCS (2009). Scientific Committee on Consumer Safety Opinion on the Mixture of 5-chloro-2-methylisothiazolin-3(2H)-one and 2-methylisothiazolin-3(2H)-one. European Commission Health & Consumers Directorate C: Public Health and Risk Assessment Brussels. Accessed 16 January 2019 at http://ec.europa.eu.

Speksnijder P, van Ravestijn J and de Voogt P (2010). Trace Analysis of Isothiazolinones in Water Samples by Large-volume Direct Injection Liquid Chromatography Tandem Mass Spectrometry. *Journal of Chromotography A*, **1217**, pp 5184-5189.

Struijs J (1996). *SimpleTreat 3.0: a model to predict the distribution and elimination of chemicals by sewage treatment plants*. Bilthoven, The Netherlands.

TGA (2015). Public Submission on Scheduling Matters Referred to the Joint ACCS/ACMS #11. Department of Health, Therapeutic Goods Administration, Canberra, ACT. Accessed 23 November 2018 at https://www.tga.gov.au.

TGA (2017). Scheduling delegate's final decisions, October 2017: 3.6 Methylisothiazolinone. Australian Government Department of Health, Therapeutic Goods Administration, Canberra, Australia. Accessed 21 December 2018 at https://www.tga.gov.au.

TGA (2019). *Poisons Standard February 2019*. Canberra, Australia. at https://www.legislation.gov.au.

UNECE (2009). *Globally Harmonised System of Classification and Labelling of Chemicals (GHS), 3rd Revised Edition*. United Nations Economic Commission for Europe, Geneva, Switzerland. Accessed 10 November 2017 at http://www.unece.org.

UNEP (1987). The Montreal Protocol on Substances that Deplete the Ozone Layer. United Nations Environment Programme, Ozone Secretariat, Nairobi, Kenya. Accessed July 2017 at http://ozone.unep.org.

UNEP (2001). *The Stockholm Convention on Persistent Organic Pollutants*. United Nations Environment Programme Secretariat of the Stockholm Convention, Châtelaine, Switzerland. Accessed 10 November 2017 at http://www.pops.int.

UNEP & FAO (1998). *The Rotterdam Convention on the Prior Informed Consent Procedure for Certain Hazardous Chemicals and Pesticides in International Trade*. United Nations Environment Programme and Food and Agriculture Organization of the United Nations, Châtelaine, Switzerland. Accessed 30 March 2016 at http://www.pic.int.

US EPA (1992). Pesticide Ecotoxicity Database (formally Environmental Fate and Effects Database (EEDB)). Environmental Fate and Effects Division, US EPA,, Washington, DC. Accessed 8 January 2019 at http://www.ipmcenters.org.

US EPA (2008). *Estimation Programs Interface (EPI) Suite^{TM} for Microsoft Windows®*, v 4.10. United States Environmental Protection Agency, Washington DC, USA. Available at https://www.epa.gov.

US EPA (2013). High Production Volume (HPV) Challenge. United States Environmental Protection Agency, Washington DC, USA. Accessed 21 December 2018 at http://www.epa.gov.

US EPA (2018a). TSCA Chemical Substance Inventory. United States Environmental Protection Agency, Washington DC,USA. Accessed 21 December 2018 at https://www.epa.gov.

US EPA (2018b). Safer Chemical Ingredients List. United States Environmental Protection Agency, Washington DC, USA. Accessed 21 December 2018 at https://www.epa.gov.

WA Department of Water (2007). Cooling Tower Wastewater Management and Disposal. Government of Western Australia, Perth. Accessed 15 January 2019 at https://www.water.wa.gov.au.

Williams T (2007). The Mechanism of Action of Isothiazolone Biocides. *PowerPlant Chemistry*, **9**(1), pp 14-22.

Williams T and McGinley H (2010). Deactivation of Industrial Water Treatment Biocides. *Corrosion 2010,* San Antonio, Texas. Accessed 29 January 2019 at https://www.onepetro.org.