



Australian Government
Department of Health and Ageing
NICNAS

INVENTORY MULTI-TIERED ASSESSMENT AND PRIORITISATION (IMAP)



HUMAN HEALTH TIER II ASSESSMENT FOR
2-Cyclohexen-1-one, 3,5,5-trimethyl-
CAS Registry Number: 78-59-1

PREFACE

As part of the reform regarding assessment of Existing Chemicals, the National Industrial Chemicals Notification and Assessment Scheme (NICNAS) is implementing a new framework to address the human health and environmental impacts of industrial chemicals, not yet assessed, on the Australian Inventory of Chemical Substances (AICS).

The framework provides a more rapid, flexible and transparent approach for the assessment of existing chemicals.

The Inventory Multi-tiered Assessment and Prioritisation (IMAP) framework was developed, with significant input from stakeholders, and will be applied in stages.

Stage One of this program, which will take three years, started 1 July 2012 and is examining 3000 chemicals meeting characteristics identified by stakeholders as needing priority assessment. This includes chemicals for which NICNAS already holds exposure information, chemicals identified as a concern or for which regulatory action has been taken overseas, and chemicals detected in international studies analysing chemicals present in babies' umbilical cord blood.

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 three 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.

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

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

Disclaimer

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ACRONYMS & ABBREVIATIONS

ACToR	Aggregated Computational Toxicology Resource (US)
AICS	Australian Inventory of Chemical Substances
ASTDR	Agency for Toxic Substances and Disease Registry (US)
bw	bodyweight
CAS	Chemical Abstracts Service
CFR	Code of Federal Regulations (US)
CHO	Chinese hamster ovary
CosIng	Cosmetic Ingredients and Substances database (EU)
d	day
DNA	Deoxyribonucleic acid
EC	European Commission
EC3	Estimated concentration three
ECHA	European Chemicals Agency
ESIS	European Chemical Substances Information System
EU	European Union
EU RAR	European Union Risk Assessment Report
FDA	Food and Drug Administration (US)
FSANZ	Food Standards Australia and New Zealand
g	gram
g/mol	grams per mole
GHS	Globally Harmonized System of Classification and Labelling of Chemicals*
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
GPMT	Guinea Pig Maximisation Test
h	hour
HGPRT	hypoxanthine guanine phosphoribosyltransferase
HPV	high production volume
HSDB	Hazardous Substances Data Bank
HSIS	Hazardous Substances Information System
HVICL	High Volume Industrial Chemicals List
IARC	International Agency for Research on Cancer
INCHEM	International Programme on Chemical Safety (also known as IPCS)
INCI	International Nomenclature of Cosmetic Ingredients
ip	intraperitoneal
IRIS	Integrated Risk Information System (US)
IUCLID	International Uniform Chemical Information Database
iv	intravenous
kg	kilogram
L	litre
LC50	median lethal concentration
LD50	median lethal dose
LCLo	lowest published lethal concentration
LLNA	local lymph node assay
LOAEL	lowest observed adverse effect level
LOEL	lowest observed effect level
m ³	cubic metre
mg	milligram
mg/cm ³	milligrams per cubic centimetre
mg/kg bw/d	milligrams per kilogram bodyweight per day
min	minute
mL	millilitre
µg	microgram
µL	microlitre
(m)SDS	(material) Safety Data Sheet

NIOSH	National Institute for Occupational Safety and Health (US)
NOAEC	no observed adverse effect concentration
NOAEL	no observed adverse effect level
NOEC	no observed effect concentration
NOEL	no observed effect level
NOHSC	National Occupational Health and Safety Commission
NTP	National Toxicology Program (US)
OECD	Organisation for Economic Cooperation and Development
OEL	occupational exposure limit
PCBU	person conducting a business or undertaking
PEL	permissible exposure limit
PND	postnatal day
ppb	parts per billion
PPE	personal protective equipment
ppm	parts per million
REACH	Registration Evaluation Authorisation of Chemicals (ECHA)
SD	Sprague Dawley
SIAP	SIDS Initial Assessment Profile (OECD)
SIAR	SIDS Initial Assessment Report (OECD)
SIDS	Screening Information Data Set (OECD)
SMILES	simplified molecular-input line-entry system
SPIN	Substances in Preparations In the Nordic countries
STEL	short-term exposure limits
STV	short-term value
SUSMP	Standard for the Uniform Scheduling of Medicines and Poisons (The Poisons Standard**)
TCLo	lowest published toxic concentration
TEEL	temporary emergency exposure limits
TSCA	Toxic Substances Control Act (US EPA)
TG	test guideline
TGA	Therapeutic Goods Administration
TLV	threshold limit values
TWA	time weighted average
UN	United Nations
US	United States of America
US EPA	United States Environmental Protection Agency
WHS	Work, Health and Safety
wt	weight
w/w	weight per weight

Glossary

NICNAS uses the IPCS Risk Assessment Terminology (IPCS, 2004) glossary, which includes:

Part 1: IPCS/OECD Key Generic Terms used in Chemical Hazard/Risk Assessment; and

Part 2: IPCS Glossary of Key Exposure Assessment Terminology.

The IPCS Risk Assessment Terminology can be accessed at:

<http://www.who.int/ipcs/methods/harmonization/areas/ipcsterminologyparts1and2.pdf>

*Globally Harmonised System of Classification and Labelling of Chemicals (GHS) United Nations, 2009.

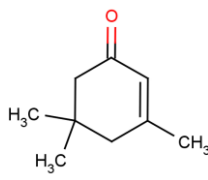
Third edition. Can be accessed at: http://www.unece.org/trans/danger/publi/ghs/ghs_rev03/03files_e.html

**The Poisons Standard (the SUSMP) can be accessed at: <http://www.tga.gov.au/industry/scheduling-poisons-standard.htm>

2-Cyclohexen-1-one, 3,5,5-trimethyl-

CAS No: 78-59-1

Chemical Identity

Synonyms	3,5,5-Trimethyl-2-cyclohexen-1-one Isophorone 2-Cyclohexen-1-one, 3,5,5-trimethyl- 3,5,5-Trimethylcyclohex-2-enone 1,1,3-Trimethyl-3-cyclohexene-5-one
Structural Formula	
Molecular Formula	C ₉ H ₁₄ O
Molecular Weight (g/mol)	138.21
Appearance and Odour (where available)	Colourless liquid with a peppermint like odour.
SMILES	C1(=O)C=C(C)CC(C)(C)C1

Import, Manufacture and Use**Australian**

No specific Australian use, import, or manufacture information have been identified.

International

The following international uses have been identified via European Union Registration, Evaluation and Authorisation of Chemicals (EU REACH) Dossiers, Canadian Assessments (Challenge List Batch 7), the Organisation for Economic Cooperation and Development Screening information data set International Assessment Reports (OECD SIAR), Galleria Chemica, Substances in Preparations in Nordic countries (SPIN) database and eChemPortal data sources including the US Environmental Protection Agency's Aggregated Computer Toxicology Resource (ACToR) and the US National Library of Medicine's Hazardous Substances Data Bank (HSDB).

The chemical has reported domestic use including:

- adhesives, binding agents;
- colouring agents; and
- paints, lacquers and varnishes.

The chemical has reported commercial use including:

- as a solvent for:
 - automotive and industrial coatings processes;
 - polyvinyl and polystyrene materials;
 - nitrocellulose resins and stoving lacquers; and
 - specialised paints and printing inks.

The chemical has reported site-limited use including:

- as a chemical intermediate for the synthesis of various organic chemicals such as alcohols; and
- raw material for 3,5-dimethylaniline.

Restrictions

Australian

This chemical is listed in the Poisons Standard (the Standard for the Uniform Scheduling of Medicines and Poisons (SUSMP)) in Schedule 5.

Schedule 5 chemicals are labeled with 'Caution'. These are substances with a low potential for causing harm, the extent of which can be reduced through using appropriate packaging with simple warnings and safety directions on the label.

International

- EU Cosmetic Directive 76/768/EEC Annex II: List of Substances which must not form part of the Composition of Cosmetic Products.
- New Zealand Cosmetic Products Group Standard - Schedule 4: Components Cosmetic Products Must Not Contain.

Existing Worker Health And Safety Controls

Hazard classification

The chemical is classified as hazardous with the following risk phrases for human health in the Hazardous Substances Information System (HSIS) (Safe Work Australia):

Carc. Cat. 3; R40 (Carcinogenicity)
Xn; R21/22 (Acute toxicity)
Xi; R36/37 (Irritation).

Exposure standards

Australian

Time Weighted Average (TWA): 28 mg/m³ (5 ppm) [peak limitation].

International

TWA: 11 mg/m³ (2 ppm) [Austria (MAK), Switzerland (MAK)]
TWA: 23 mg/m³ (4 ppm) [USA (Alaska, California, Hawaii, Michigan, Minnesota, Nt Carolina, Tennessee, Vermont)]
TWA: 25 mg/m³ (5 ppm) [Argentina, Canada(Yukon), Greece]
TWA: 55 mg/m³ (10 ppm) [USA (Oregon)]
TWA: 140 mg/m³ (25 ppm) [USA (Idaho, Wyoming)].

Short-Term Exposure Limit (STEL): 11 mg/m³ (2 ppm) [Austria (MAK)]
STEL: 22 mg/m³ (4 ppm) [Switzerland (MAK)]
STEL: 25 mg/m³ (5 ppm) [Argentina, Greece, Mexico, South Africa]
STEL: 28 mg/m³ (5 ppm) [Belgium, Singapore]
STEL: 29 mg/m³ (5 ppm) [United Kingdom].

Ceiling: 28 mg/m³ (5 ppm) [China (Hong Kong), Canada (Alberta, NW Territories), Chile, Korea (South), Malaysia, New Zealand, Peru]
Ceiling: 30 mg/m³ (5 ppm) [Sweden].

Health Hazard Information

Toxicokinetics

It is reported that on administration via the oral and inhalation route, the chemical is well absorbed and rapidly distributed through the body of rats and rabbits (OECD 2003). While some of the absorbed

chemical is excreted unchanged via urine and exhaled air, metabolites are mainly excreted as glucuronides. The tendency of the chemical to bioaccumulate is very low, since within 24 hours after administration more than 93% of orally administered chemical was excreted by rats.

Acute Toxicity

Oral

The chemical is classified as hazardous with the risk phrase 'Harmful if swallowed' (Xn; R22) in HSIS (Safe Work Australia). The data available support this classification.

Oral median lethal doses (LD50s) of 1500–3450 mg/kg bw were reported in rats (OCED 2003). Clinical signs of toxicity reported at doses of ≥ 1450 mg/kg bw included general apathy, weariness (leading to coma), ptosis, lacrimation and laboured respiration. At doses of ≥ 5000 mg/kg bw, congestion of lungs and kidneys were found at necropsy, in addition to pancreas and liver lesions.

Dermal

The chemical is classified as hazardous with the risk phrase 'Harmful in contact with skin' (Xn; R21) in HSIS (Safe Work Australia). The data available support this classification.

The dermal LD50 in rats was reported to be 1700 mg/kg bw (OECD 2012). Clinical signs reported were general apathy, loss in body mass, tremors, lacrimation, either accelerated or laboured respiration, prostration, narcosis and in some instances coma. At necropsy, uniform thickening of the cutaneous stomach mucosa and cases of pulmonary emphysema, oedema or hyperaemia were reported.

Inhalation

The chemical showed very low acute toxicity with a median lethal concentration (LC50) value of 7000 mg/m³ reported in rats (OECD 2003).

Clinical signs of toxicity included nose and eye irritation, accelerated or laboured respiration and, in some cases, coma at doses ≥ 5000 mg/m³ (OCED 2003). At necropsy, congestion of the lungs and reported liver and stomach congestion were found at high exposure concentrations.

Corrosion / Irritation

Skin irritation

The chemical was reported as not irritating to the skin of rabbits when tested for 4 hours under semi-occlusive conditions according to OECD Test Guideline 404 (OECD 2003).

Eye irritation

The chemical is classified as hazardous with the risk phrase 'Irritating to eyes' (Xi; R36) in HSIS (Safe Work Australia). The data available support this classification.

The chemical was reported to be an eye irritant in a guideline study on rabbits (OECD 2003). Marked irritation was observed, which reversed within 14 days but was still present after 7 days. The 3-day average scores for cornea/iris/conjunctiva were reported as 14/80, 0/10, 6/20.

Respiratory irritation

The chemical is classified as hazardous with the risk phrase 'Irritating to the respiratory system' (Xi; R7) in HSIS (Safe Work Australia). The data from available acute inhalation studies and observations in humans support this classification.

Observation in humans

Irritating effects in eyes, nose and throat were reported after 15 minutes' exposure to 12 volunteers at vapour concentrations above 144 mg/m³. The odour threshold of the chemical is reported to be 1.15 mg/m³ (OECD 2003).

Sensitisation

Skin sensitisation

The chemical was not found to induce dermal sensitisation in guinea pigs when tested according to OECD Guideline 406 (OECD 2003).

Repeat Dose Toxicity

Oral

Repeat dose oral toxicity studies have reported no observed adverse effect levels (NOAELs) ranging from 102.5 to 500 mg/kg bw/day.

In a repeated dose oral toxicity study in rats, 750, 1000, or 3000 ppm of the chemical was administered via the diet to 20 animals per sex per dose for 13 weeks (OECD 2003, REACH 2012). The actual doses received (based on the mean daily compound consumption) were 57.0, 102.5 and 233.8 mg/kg bw/day (males) and 73.9, 163.8 and 311.8 mg/kg bw/day (females). The only effect reported was a reduction in body weight gain (−12 % to −13 %) in male rats from the 233.8 mg/kg bw/day group from 6–11 weeks on. The NOAEL from this study was determined to be 102.5 mg/kg bw/day (males) and 311.8 mg/kg bw/day (females).

In a repeated dose oral toxicity study in rats (Fischer), the chemical was administered via gavage at 0, 125, 250, 500, 1000 and 2000 mg/kg bw/day to five animals per sex per dose for 16 days (OECD 2003). Five deaths were reported (one male and four females) from the 2000 mg/kg bw/day group. A reduction in body weight gain was reported in male (−13.9 %) and female (−6.7 %) rats from the 1000 mg/kg bw/day group.

In an additional study in repeated oral dose toxicity in rats (Fischer), the chemical was administered via gavage to 10 males and 10 females at 0, 62.5, 125, 250, 500 and 1000 mg/kg bw/day for 13 weeks. It was reported that one female died in the 1000 mg/kg bw/day group. A reduction in body weight gain was only seen in the male rats (−5.1 %) from the 1000 mg/kg bw/day group. The reported NOAEL for these two studies was 500 mg/kg bw/day.

Dermal

No data available.

Inhalation

The no observed adverse effect concentration (NOAEC) for the chemical was determined to be < 208 mg/m³.

In a repeated dose inhalation toxicity study in rats (Charles River CD), the chemical was administered to 10 males and 10 females at 0 or 250 mg/m³ (whole body exposure) for 6 hours/day, 5 days/week for 4 weeks (REACH, 2012). Prior to gross necropsy at 4 weeks, no mortality within the treatment group was reported. Histological examination was performed in three males and three females per group. Transient nasal bleeding, increased percentages of lymphocytes, decreased percentages of neutrophils and increased haemoglobin concentration in males and females were reported. Significantly lower terminal body weights and significantly decreased absolute and relative liver weights of exposed males, compared with controls, were reported. The reported average daily exposure to the chemical was 208 mg/m³.

In another repeated dose inhalation toxicity study using rats (Wistar), the chemical was administered to 10 males and 10 females at 2873 mg/m³ (500 ppm whole body exposure) for 6 hours/day, 5 days/week for 6 months (males) and 4 months (females) (REACH 2012). Three cases of mortality were reported from those exposed to the chemical (two males and one female). No mortality was reported in the control animals.

Genotoxicity

Together with the negative in vitro and in vivo results, the chemical is not expected to cause genotoxic effects (OECD 2003).

In vitro studies

The chemical did not exhibit mutagenic activity in several Ames tests using *Salmonella typhimurium* (with and without S9 activation).

The mouse lymphoma tests were primarily negative, with one positive result recorded in the presence of metabolic activation. Without metabolic activation some studies gave negative results, others positive results. However, positive results were only observed at reduced relative total growth (RTG) values.

In a cytogenetic assay with Chinese hamster ovary (CHO) cells, no significant increase in chromosomal aberrations was observed. A further chromosomal aberration assay, performed on Chinese hamster lung (CHL) cells, was positive with metabolic activation only after a modified treatment of the cells (cells were treated for six hours and then cultured in fresh medium for another 18 hours). Increased chromosome aberrations without metabolic activation were only observed at cytotoxic concentrations. It was reported that a significant increase in sister chromatid exchange (SCE) frequency at concentrations of 500–1000 mg/L was induced by the chemical only in the absence of S9 mix (no increase in the presence of Aroclor 1254-induced rat liver S9 mix). As these high concentrations were cytostatic, increased SCE frequencies could only be detected after delayed harvest.

The chemical was tested for the induction of unscheduled DNA synthesis (UDS) in rat primary hepatocytes. Concentrations ranged from 0.005–0.4 µl/ml, the highest concentration being toxic. No increase in the mean nuclear grain count (as compared to controls), or in the incidence of cells undergoing repair, was detected at any dose level.

In vivo studies

In a DNA binding study, male and female F344 rats and B6C3F1 mice (25 animals per group) were administered 500 mg/kg of the chemical. The liver and kidneys were examined after 24 hours. There was no reported binding of the chemical or its metabolites to DNA of these organs.

Carcinogenicity

The chemical is currently classified as hazardous as a Category 3 carcinogen with the risk phrase 'Limited evidence of carcinogenic effect' (Xn; R40) in HSIS (Safe Work Australia). The data available support this classification.

There is reported evidence of carcinogenicity of the chemical in male rats (kidney tumours, preputial gland carcinomas). The kidney tumours can be attributed to a male rate-specific α₂µ-globulin associated mechanism and therefore not relevant to other species. As the preputium is only investigated histopathologically when gross lesions are found, neither true tumour incidences from this study nor from historical controls are available. There was equivocal evidence of carcinogenicity for male mice (liver tumours, mesenchymal tumours of the integumentary system). There was no evidence of carcinogenicity of the chemical in female rats and mice (OECD 2003; REACH 2012).

Reproductive and developmental toxicity

The chemical is reported to not cause adverse reproductive or developmental effects (OECD 2003).

In a one generation reproductive toxicity study, 10 male and 10 female rats (Wistar) were exposed to the chemical at 2872 mg/m³ (500 ppm) in air for three months. Mating occurred between groups of five exposed males with five control and five exposed females. In addition, groups of five exposed females were mated with five control and five exposed males. Exposure to the chemical continued for females throughout gestation and they were allowed to deliver. Treatment with the chemical did not influence pregnancy rates or litter sizes, nor were there any abnormalities reported in the pups.

In another study, pregnant rats (Fischer 344) were exposed to the chemical through day 6 to day 15 of gestation, at concentrations of 144, 289, or 664 mg/m³ (22 animals per dose level). A significant reduction in food consumption was reported in the highest dose group. Loss in body weight was also reported in the the highest dose group (-6.1% at gestation day 12 and -6.8% at gestation day 15). A dose related increase in alopecia was observed, as well as a discolouration of the cervical and anogenital region. No adverse effects in the foetuses were reported. The NOAEL for maternal toxicity in this study was reported to be 289 mg/m³.

Risk Characterisation

Critical Health Effects

The chemical has moderate acute oral and dermal toxicity and is irritating to the eyes and respiratory system. Although there is limited evidence of carcinogenicity in animal tests, the data do not provide sufficient evidence to regard the chemical as a likely human carcinogen.

Public Risk Characterisation

While the use of this chemical in domestic products in Australia is not known, the chemical is reported to be used in a number of domestic products overseas.

In Australia this chemical is currently listed in the SUSMP in Schedule 5. Products containing the chemical must be labelled in accordance with the SUSMP requirements and this should be sufficient to mitigate risks from such products.

Occupational Risk Characterisation

There is concern for worker health and safety given the critical health effects of the chemical.

The chemical is currently classified on HSIS and the appropriate labelling should be applied to ensure that a person conducting a business or undertaking (PCBU), or an employee at a workplace, has adequate information to adhere to appropriate controls.

NICNAS Recommendation

Current risk management measures are considered adequate for the protection of public and workers' health and safety, provided that all requirements are met under workplace health and safety and poisons legislation as adopted by the relevant state or territory. No further assessment is required.

Regulatory Control

Public Health

Products containing the chemical should be labelled in accordance with state and territory legislation (SUSMP).

Occupational Health and Safety

The chemical is classified and labelled under the current Approved Criteria and adopted GHS as recommended below (SWA GHS, 2012). This does not consider classification of physical hazards and environmental hazards.

	Approved Criteria (HSIS)^a	GHS Classification
Acute Toxicity	Harmful if swallowed (Xn; R22)* Harmful in contact with skin (Xn; R21)*	Harmful if swallowed - Cat. 4 (H302) Harmful in contact with skin - Cat. 4 (H312)
Irritation / Corrosivity	Irritating to eyes (Xi; R36)* Irritating to respiratory system (Xi; R37)*	Causes serious eye irritation - Cat. 2A (H319) May cause respiratory irritation - Specific target organ tox, single exp Cat. 3 (H335)
Carcinogenicity	Carc. Cat 3 - Limited evidence of a carcinogenic effect (Xn; R40)*	Suspected of causing cancer - Cat. 2 (H351)

^a Approved Criteria for Classifying Hazardous Substances [NOHSC:1008(2004)].

* Existing Hazard Classification. No change recommended to this classification.

Advice for consumers

Products containing the chemical should be used according to label instructions.

Advice for industry

Control measures

Control measures to minimise the risk from dermal, ocular and inhalation exposure to the chemical should be implemented in accordance with the hierarchy of controls. Approaches to minimise risk include substitution, isolation and engineering controls. Measures required to eliminate or minimise risk arising from storage, handling and use of a hazardous chemical are dependent on the physical form and the manner in which the chemical is used. Examples of control measures which may minimise the risk include, but are not limited to:

- use of closed systems or isolation of operations;
- use of local exhaust ventilation to prevent the chemical from entering the breathing zone of any worker;
- health monitoring for any worker who is at risk of exposure to the chemical, if valid techniques are available to monitor the effect on the worker's health;
- air monitoring to ensure control measures in place are working effectively and continue to do so;
- minimisation of manual processes and work tasks through automation of processes;
- work procedures that minimise splashes and spills;
- regular cleaning of equipment and work areas; and
- use of protective equipment that is designed, constructed, and operated to ensure that the worker does not come into contact with the chemical.

Guidance on managing risks from hazardous chemicals are provided in the *Managing Risks of Hazardous Chemicals in the Workplace—Code of Practice* available on the Safe Work Australia website.

Personal protective equipment should not be relied upon on its own to control risk and should only be used when all other reasonably practicable control measures do not eliminate or sufficiently minimise risk. Guidance in selection of personal protective equipment can be obtained from Australian, Australian/New Zealand or other approved standards.

Obligations under workplace health and safety legislation

Information in this report should be taken into account to assist with meeting obligations under workplace health and safety legislation as adopted by the relevant state or territory. This includes but is not limited to:

- ensuring that hazardous chemicals are correctly classified and labelled;
- ensuring that (material) safety data sheets ((m)SDS) containing accurate information about the hazards (relating to both health and physicochemical (physical) hazards) of chemicals are prepared; and
- management of risks arising from storage, handling and use of a hazardous chemical.

Your work health and safety regulator should be contacted for information on the work health and safety laws in your jurisdiction.

Information on how to prepare an (m)SDS and how to label containers of hazardous chemicals are provided in relevant Codes of Practice such as the *Preparation of Safety Data Sheets for Hazardous Chemicals— Code of Practice* and *Labelling of Workplace Hazardous Chemicals—Code of Practice*, respectively. These Codes of Practice are available from the Safe Work Australia website.

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