File No: STD/1614

November 2017

NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

PUBLIC REPORT

Acetic acid, 2-(2-benzothiazolylthio)-

This Assessment has been compiled in accordance with the provisions of the *Industrial Chemicals (Notification and Assessment) Act 1989* (the Act) and Regulations. This legislation is an Act of the Commonwealth of Australia. The National Industrial Chemicals Notification and Assessment Scheme (NICNAS) is administered by the Department of Health, and conducts the risk assessment for public health and occupational health and safety. The assessment of environmental risk is conducted by the Department of the Environment and Energy.

This Public Report is available for viewing and downloading from the NICNAS website or available on request, free of charge, by contacting NICNAS. For requests and enquiries please contact the NICNAS Administration Coordinator at:

Street Address:	Level 7, 260 Elizabeth Street, SURRY HILLS NSW 2010, AUSTRALIA.
Postal Address:	GPO Box 58, SYDNEY NSW 2001, AUSTRALIA.
TEL:	+ 61 2 8577 8800
FAX:	+ 61 2 8577 8888
Website:	www.nicnas.gov.au

Director
NICNAS

TABLE OF CONTENTS

SUMMARY

The following details will be published in the NICNAS *Chemical Gazette:*

ASSESSMENT REFERENCE	APPLICANT(S)	CHEMICAL OR TRADE NAME	HAZARDOUS CHEMICAL	INTRODUCTION VOLUME	USE
STD/1614	Clariant (Australia) Pty Ltd	Acetic acid, 2-(2-benzothiazolylthio)-	Yes	≤ 6 tonnes per annum	Component of coolants and metalworking fluid

CONCLUSIONS AND REGULATORY OBLIGATIONS

Hazard classification
Based on the available information, the notified chemical is recommended for hazard classification according to the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia. The recommended hazard classification is presented in the following table.

Hazard classification	*Hazard statement*
Acute Toxicity (Oral) Category 4	H302 – Harmful if swallowed
Serious Eye Damage/Eye Irritation Category 1	H318 – Causes serious eye damage

The environmental hazard classification according to the *Globally Harmonised System of Classification and Labelling of Chemicals* (GHS) is presented below. Environmental classification under the GHS is not mandated in Australia and carries no legal status but is presented for information purposes.

Hazard classification	*Hazard statement*
Acute Category 3	H402 – Harmful to aquatic life
Chronic Category 3	H412 – Harmful to aquatic life with long lasting effects

Human health risk assessment
Provided that the recommended controls are being adhered to, under the conditions of the occupational settings described, the notified chemical is not considered to pose an unreasonable risk to the health of workers.

When used in the proposed manner, the notified chemical is not considered to pose an unreasonable risk to public health.

Environmental risk assessment
On the basis of the PEC/PNEC ratio and the reported use pattern, the notified chemical is not considered to pose an unreasonable risk to the environment.

Recommendations

REGULATORY CONTROLS

Hazard Classification and Labelling

- The notified chemical should be classified as follows:
  - Acute Toxicity (Oral) Category 4: H302 – Harmful if swallowed
  - Serious Eye Damage/Eye Irritation Category 1: H318 – Causes serious eye damage

The above should be used for products/mixtures containing the notified chemical, if applicable, based on the concentration of the notified chemical present and the intended use/exposure scenario.

CONTROL MEASURES

Occupational Health and Safety

- A person conducting a business or undertaking at a workplace should implement the following engineering controls to minimise occupational exposure to the notified chemical during reformulation:
  - Enclosed and automated system
  - Fume extractor for weighing
  - Local exhaust ventilation

- A person conducting a business or undertaking at a workplace should implement the following safe work practices to minimise occupational exposure during handling of the notified chemical:
  - Avoid contact with eyes
  - Avoid inhalation of any dust or aerosol

- A person conducting a business or undertaking at a workplace should ensure that the following personal protective equipment is used by workers to minimise occupational exposure to the notified chemical:
  - Protective clothing
  - Impervious gloves
  - Eye protection
  - Respiratory protection if dusts or aerosols are expected

  Guidance in selection of personal protective equipment can be obtained from Australian, Australian/New Zealand or other approved standards.

- A copy of the SDS should be easily accessible to employees.

- If products and mixtures containing the notified chemical are classified as hazardous to health in accordance with the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS)* as adopted for industrial chemicals in Australia, workplace practices and control procedures consistent with provisions of State and Territory hazardous substances legislation should be in operation.

Disposal

- Where reuse or recycling are not appropriate, dispose of the notified chemical in an environmentally sound manner in accordance with relevant Commonwealth, state, territory and local government legislation.

Storage

- The handling and storage of the notified chemical should be in accordance with the Safe Work Australia Code of Practice for *Managing Risks of Hazardous Chemicals in the Workplace* (SWA, 2012) or relevant State or Territory Code of Practice.

Emergency procedures

- Spills or accidental release of the notified chemical should be handled by physical containment, collection and subsequent safe disposal.

Transport and Packaging

- Due to the serious eye damage properties of the notified chemical, the notifier should consider their obligations under the Australian Dangerous Goods Code (NTC 2017) when transporting the neat form of the notified chemical.

Regulatory Obligations

Secondary Notification
This risk assessment is based on the information available at the time of notification. The Director may call for the reassessment of the chemical under secondary notification provisions based on changes in certain circumstances. Under Section 64 of the *Industrial Chemicals (Notification and Assessment) Act (1989)* the notifier, as well as any other importer or manufacturer of the notified chemical, have post-assessment regulatory obligations to notify NICNAS when any of these circumstances change. These obligations apply even when the notified chemical is listed on the Australian Inventory of Chemical Substances (AICS).

Therefore, the Director of NICNAS must be notified in writing within 28 days by the notifier, other importer or manufacturer:

(1) Under Section 64(1) of the Act; if
- the use concentration of the chemical in coolant products exceeds 1%;
- further information has become available to the person as to reproduction/developmental toxicity effects of the chemical;

or

(1) Under Section 64(2) of the Act; if
- the function or use of the chemical has changed from a component of coolants and metalworking fluid, or is likely to change significantly;
- the amount of chemical being introduced has increased, or is likely to increase, significantly;
- the chemical has begun to be manufactured in Australia;
- additional information has become available to the person as to an adverse effect of the chemical on occupational health and safety, public health, or the environment.

The Director will then decide whether a reassessment (i.e. a secondary notification and assessment) is required.

Safety Data Sheet
The SDS of the products containing the notified chemical provided by the notifier was reviewed by NICNAS. The accuracy of the information on the SDS remains the responsibility of the applicant.

ASSESSMENT DETAILS

1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT
Clariant (Australia) Pty Ltd (ABN: 30 069 435 552)
Level 3, Olympus Building, 3 Acacia Place
296–324 Ferntree Gully Road
NOTTING HILL VIC 3168

NOTIFICATION CATEGORY
Standard: Chemical other than polymer (more than 1 tonne per year)

EXEMPT INFORMATION (SECTION 75 OF THE ACT)
Data items and details claimed exempt from publication: other names, spectral data, degree of purity, use details, and import volume.

VARIATION OF DATA REQUIREMENTS (SECTION 24 OF THE ACT)
Variation to the schedule of data requirements is claimed for vapour pressure, adsorption/desorption, dissociation constant, autoignition temperature and acute inhalation toxicity.

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT(S)
None

NOTIFICATION IN OTHER COUNTRIES
USA, China, Canada, EU, Japan, Philippines, South Korea

2. IDENTITY OF CHEMICAL

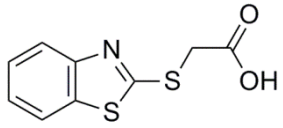
CHEMICAL NAME
Acetic acid, 2-(2-benzothiazolylthio)-

MARKETING NAME
PCA 100

CAS NUMBER
6295-57-4

MOLECULAR FORMULA
$C_9H_7NO_2S_2$

STRUCTURAL FORMULA



MOLECULAR WEIGHT
225.287 Da

3. COMPOSITION

DEGREE OF PURITY
> 97%

4. PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE AT 20 °C AND 101.3 kPa: Yellow powder

Property	Value	Data Source/Justification
Melting Point	156 – 158 °C	Measured
Boiling Point	Not determinable	Decomposed at ≥ 174 °C
Density	1,580 kg/m^3 at 27 °C	Measured
Vapour Pressure	< 1 × 10^{-6} kPa at 20 °C	Calculated
Water Solubility	133 mg/L at 20 °C	Measured
n-Octanol Solubility	12.6 g/L at 20 °C	Measured
Hydrolysis as a Function of pH	Hydrolytically stable at pH 4,7 and 9	Measured
Partition Coefficient (n-octanol/water)	log P_{ow} = 1.60 at 23 °C	Measured
Surface Tension	70.6 ± 0.9 mN/m at 20 °C	Measured
Adsorption/Desorption	log K_{oc} = 1.04	Estimated from log P_{ow} using KOCWIN v2.0
Dissociation Constant	pKa (acid) = 3.7 ± 0.4 pKa (base) = 1.8 ± 0.8	Calculated by the notifier using ACD/I-Lab Web service
Particle Size	Inhalable fraction (< 100 μm): 27.2% Respirable fraction (< 10 μm): 15.5% D10: 4.8 μm D50: 379.1 μm D90: 1075.7 μm	Measured
Flash Point	Not determined	Not expected to form flammable vapour
Flammability (Solids)	Not a highly flammable solid	Measured
Autoignition Temperature	Not determined	Not expected to undergo autoignition
Explosive Properties	Not determined	Contains no functional groups that would imply explosive properties
Oxidising Properties	Not determined	Contains no functional groups that would imply oxidative properties

DISCUSSION OF PROPERTIES
For full details of tests on physical and chemical properties, refer to Appendix A.

Reactivity
The notified chemical is expected to be stable under normal conditions of use.

Physical hazard classification
Based on the submitted physico-chemical data depicted in the above table, the notified chemical is not recommended for hazard classification according to the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia.

5. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will not be manufactured in Australia. It will be imported as a component in finished engine coolant at < 1% concentration. In the future, the neat notified chemical will be imported for local reformulation into coolants and metalworking products. The metalworking products will contain < 25% concentration of the notified chemical.

MAXIMUM INTRODUCTION VOLUME OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS

Year	*1*	*2*	*3*	*4*	*5*
Tonnes	3 – 6	3 – 6	3 – 6	3 – 6	3 – 6

PORT OF ENTRY
Melbourne and Sydney

IDENTITY OF RECIPIENT
Clariant (Australia) Pty Ltd

TRANSPORTATION AND PACKAGING
The finished coolants containing the notified chemical at < 1% concentration will be imported in 1 L, 5 L, 20 L, 200 L and 1,000 L containers, stored in the customer's warehouses and distributed to customer sites or retail outlets by railway or road.

In the future, the neat notified chemical will be imported in 20 kg plastic bags within drums or cardboard boxes, stored at the notifier's warehouse and distributed to reformulation sites by railway or road.

USE
The notified chemical will be used as a component of engine coolants at < 1% concentration and in metalworking products at < 25% concentration. The metalworking products will be diluted in metalworking baths to a final use concentration of < 5%.

OPERATION DESCRIPTION
The finished coolant products containing the notified chemical (at < 1% concentration) will be manually added into motor vehicle radiators. Spent coolant will be drained and replaced with fresh coolant. The finished coolant products will be mainly used by aftermarket vehicle service centres or by do-it-yourself (DIY) users.

In the future, the notified chemical may be imported for reformulation in Australia. At reformulation sites, process workers will receive the notified chemical in neat powder form. The notified chemical will be weighed within a fume extraction hood to a sealed transfer container. The notified chemical will then be manually added into a blending tank. The blended products, containing the notified chemical at < 25% concentration for metalworking products and < 1% concentration for finished coolant, will be assayed by quality assurance staff and then pumped into an automated and enclosed filling machine for packaging into various containers in the size ranging from 1 L to 1,000 L.

For metalworking, the products containing the notified chemical will be diluted in metalworking baths before final application. The final fluid, containing the notified chemical at < 5% concentration, will be recirculated during use and refilled by an automatic dosing pump when required. Used metalworking fluid in the baths will be replaced with new fluid every 12 months and the used fluid will be disposed of by waste disposal contractors.

6. HUMAN HEALTH IMPLICATIONS

6.1. Exposure Assessment

6.1.1. Occupational Exposure

CATEGORY OF WORKERS

Category of Worker	*Exposure Duration (hours/day)*	*Exposure Frequency (days/year)*
Transport and storage workers	1-2	24
Reformulation workers	8	50
Retail workers	1-2	250
Motor mechanics	8	250
Metalworking machine operators	8	250

EXPOSURE DETAILS
Transport and storage workers may come into contact with the notified chemical in neat form or as a component of finished products, only in the event of accidental rupture of containers.

During reformulation, dermal, ocular and inhalation exposure to the notified chemical (up to 100% neat form) may occur during the weighing and blending. The notified chemical will be weighed under a fume extractor and blending will be carried out in a close mixing tank with local exhaust ventilation. Personal protective equipment (PPE) such as coverall, respirators, gloves and eye protection will be worn during these activities. As the blending and packaging processes will occur in automated and enclosed systems, exposure to the notified chemical during the reformulation is expected to be limited.

Retail workers handling coolant products containing the notified chemical may come into contact with the notified chemical at < 1% concentration only in the unlikely event of an accident where the packaging is damaged and the product is spilt. During any necessary clean-up, workers are expected to wear appropriate PPE including gloves, overalls and eye protection.

Motor mechanics will handle the finished coolant products containing the notified chemical during refilling and draining of radiators. Exposure to the chemical at < 1% concentration via the dermal route may occur. Mechanics are expected to wear appropriate PPE including gloves, overalls and eye protection during handling and to be trained to handle the coolant products containing the notified chemical in a safe manner to minimise potential for exposure.

Metalworking machine operators may have the potential to be exposed to the notified chemical at < 25% concentration through dermal, ocular or inhalation routes during fluid preparation and at the time of metalworking operations. Exposure is expected to be limited due to the engineering controls in place and the use of PPE, such as gloves, overall, eye protection and, when necessary, respiratory protection.

6.1.2. Public Exposure

The neat form of the notified chemical and metalworking products containing the chemical will not be available to the general public. However, members of the general public may handle coolant products containing the notified chemical at 1% concentration for DIY use. Dermal and incidental ocular exposure to the chemical is possible during operations. DIY activities with the coolant products are expected to occur infrequently.

6.2. Human Health Effects Assessment

The results from toxicological investigations conducted on the notified chemical are summarised in the following table. For full details of the studies, refer to Appendix B.

Endpoint	*Result and Assessment Conclusion*
Rat, acute oral toxicity	LD50 = 1,580 mg/kg bw; harmful
Rat, acute dermal toxicity	LD50 > 2,000 mg/kg bw; low toxicity
Rabbit, skin irritation	non-irritating
Rabbit, eye irritation	severely irritating
Guinea pig, skin sensitisation – maximisation test	no evidence of sensitisation
Rat, repeat dose oral toxicity – 28-days	NOAEL > 100 mg/kg bw/day
Mutagenicity – bacterial reverse mutation	non mutagenic
Mutagenicity – *in vitro* mammalian cell gene mutation	non mutagenic
Genotoxicity – *in vitro* mammalian chromosome aberration	non genotoxic
Rat, reproductive and developmental toxicity	NOAEL (systemic) = 62.5 mg/kg bw/day (male); 125 mg/kg bw/day (female) NOAEL (reproductive) = 31.25 mg/kg bw/day (female)

Toxicokinetics, metabolism and distribution

No information on toxicokinetics of the notified chemical was provided. The notified chemical has a low molecular weight (< 500 Da). It has a water solubility of 133 mg/L at 20 °C and a log P_{ow} of 1.60. Therefore absorption of the notified chemical across biological membranes may occur.

Acute toxicity

The notified chemical was determined to be harmful to rats in an acute oral toxicity study (LD50 = 1,580 mg/kg bw) with dose levels from 1,000 mg/kg bw causing mortality. It was found to be of low acute toxicity to rats via the dermal route.

No acute inhalation toxicity data was provided.

Irritation and sensitisation

The notified chemical was determined to be severely irritating to the eyes when tested in rabbits, causing irreversible effects within 14 days of observation to conjunctiva, cornea and iris, including complete corneal opacity in one test animal. However, the notified chemical was found to be non-irritating to the skin of rabbits in a skin irritation study.

The notified chemical was not considered to be a skin sensitiser in a guinea pig skin sensitisation test.

Repeated dose toxicity
Based on a dose range finding study, the maximum tolerable dose (MTD) was considered by the study authors to be 100 mg/kg bw/day for repeated dose toxicity by the oral route.

In a subsequent 28-day repeated dose oral toxicity study, rats were treated with the notified chemical at 5, 15, 40 and 100 mg/kg bw/day. Toxicology effects, including clinical signs of toxicity, haematology, coagulation, clinical chemistry, urinalysis parameters, main organ effects and sperm parameters, were examined. There were no unscheduled deaths for animals in all dose groups and no adverse effects were reported by the study authors.

The No Observed Adverse Effect Level (NOAEL) was established for the notified chemical in the study as 100 mg/kg bw/day in rats based on the highest dose level tested. However, animals treated at 40 and 100 mg/kg bw/day showed statistically significant increase in kidney and adrenal weights compared to the vehicle control group.

Mutagenicity/Genotoxicity
The notified chemical was not mutagenic in a bacterial reverse mutation test and an *in vitro* mammalian cell gene mutation test. The notified chemical was not clastogenic in an *in vitro* mammalian chromosome aberration test.

Toxicity for reproduction and development
In a reproduction/developmental toxicity screening test, no treatment-related adverse effects for systemic toxicity were observed in females up to 125 mg/kg bw/day. The males treated at 125 mg/kg bw/day showed some changes in the haematology parameters and organ weights, including decreased reticulocyte count and globulin concentration, decreased body weight and thymus weight, and increased adrenal weight and kidney weight. These changes were considered as treatment-related changes.

No changes in the reproductive organs of males and females were observed in the study.

The NOAEL for female reproductive toxicity was reported by the study authors to be 62.5 mg/kg bw day. However, for females treated at 125 mg/kg bw day, increased pre-implantation loss was observed, which resulted in reduced implantation index. Increased post-implantation loss was observed at the 62.5 and 125 mg/kg bw/day dose groups, which resulted in a reduced mean litter size. Survival rate of the pups was also reduced due to cannibalism by the dam observed in all dosed groups.

These observations suggest that there is likely a potential for the notified chemical to cause certain adverse reproductive effects in females.

Health hazard classification
Based on the available information, the notified chemical is recommended for hazard classification according to the *Globally Harmonised System of Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia. The recommended hazard classification is presented in the following table.

Hazard classification	***Hazard statement***
Acute Toxicity (Oral) Category 4	H302 – Harmful if swallowed
Serious Eye Damage/Eye Irritation Category 1	H318 – Causes serious eye damage

Due to the serious eye damage properties of the notified chemical, the notifier should consider their obligations under the Australian Dangerous Goods Code (NTC 2017) when transporting the neat form of the notified chemical.

6.3. Human Health Risk Characterisation

6.3.1. Occupational Health and Safety
The notified chemical is expected to be harmful following oral exposure and to be a severe eye irritant causing irreversible serious eye damage. It may also have potential to cause adverse reproductive effects in females.

Adverse effects from inhalation cannot be ruled out, due to lack of data on the notified chemical. However, as the vapour pressure of the chemical is expected to be low, inhalation exposure may only occur if dusts or aerosols are formed.

Reformulation workers may be exposed to the imported notified chemical up to 100% concentration (neat form) during reformulation operations. Other workers may come into contact with the notified chemical at concentration ranging from < 25% to < 1%. The use of PPE and engineering controls during operations is expected to minimise potential for exposure and therefore reduce the risk of adverse health effects including eye irritation.

Provided that control measures are in place to minimise worker exposure to the notified chemical, the risk to the health of workers from use of the notified chemical is not considered to be unreasonable.

6.3.2. Public Health

The public may come into contact with the notified chemical at < 1% concentration when the coolant products containing the notified chemical are used. Eye irritation effects are not expected at this relatively low final use concentration. Consumer exposure is expected to be limited via dermal route. Repeated daily use of the products is unlikely.

When used in the proposed manner, the risk to public health associated with the use of the notified chemical in coolants at < 1% concentration is not considered to be unreasonable.

7. ENVIRONMENTAL IMPLICATIONS

7.1. Environmental Exposure & Fate Assessment

7.1.1. Environmental Exposure

RELEASE OF CHEMICAL AT SITE

The notified chemical will be imported as a component in finished engine coolants at < 1% concentration. In the future, the neat notified chemical will be imported for local reformulation into coolants and metalworking products. The reformulation process will involve blending operation that will occur in an enclosed system, followed by automated filling of the finished products into end-use containers. Blending equipment will be cleaned with solvents. The waste liquids containing the notified chemical are expected to be disposed of in accordance with local government regulations. Accidental spills of the notified chemical during reformulation, transport or storage are expected to be adsorbed onto a suitable material and disposal of in accordance with local regulations.

RELEASE OF CHEMICAL FROM USE

Engine Coolant

Engine coolant products containing < 1% of the notified chemical will primarily be used by commercial automotive servicing workshops, or by DIY consumers. DIY users are advised to collect the spent coolants and take it to automotive workshops for disposal. In the automotive servicing workshops, spent engine coolants, containing mostly ethylene glycol, will be collected and recycled through liquid waste treatment facilities using filtration, ion exchange or distillation process. The notifier indicates that the latter is the most prevalent and that the notified chemical is expected to remain in the bottoms/residues and is likely to decompose as a result of the high temperatures. Wastes containing residual amounts of the notified chemical from the above processes are expected to be disposed of to landfill.

Metalworking Fluids

For metalworking fluids, the products containing the notified chemical will be diluted in metalworking baths before final application. The final fluids, containing the notified chemical at < 5% concentration, will be recirculated during use and refilled by an automatic dosing pump when required. In large industrial facilities, the used metalworking fluids in the baths will be replaced with new fluids every 12 months, and the spent fluids will be collected and disposed of by waste disposal contractors. At the waste disposal facilities, the spent metalworking fluids containing the notified chemical will be subjected to oil/water separation procedures. Based on its moderate water solubility and predicted log P_{ow} = 1.6, the notified chemical is expected to present in both organic and aqueous phases. After being separated, the aqueous component will be treated at onsite wastewater treatment plants before being released to sewage systems, and the oily sludge will be disposed to landfill in

accordance with local regulations. It is estimated by the notifier that up to 25% of the import volume of the notified chemical may be released directly to sewers as used metalworking fluids from smaller factories.

Accidental spills of the notified chemical during uses are expected to be adsorbed onto a suitable material and disposal of in accordance with local regulations.

RELEASE OF CHEMICAL FROM DISPOSAL
The empty containers containing small amount of residue notified chemical will be disposed of to landfill in accordant with local regulations.

7.1.2. Environmental Fate
Following its uses, the main release of the notified chemical would be from direct discharge of used metalworking fluids to sewers from small factories, and possibly from discharge of treated effluent from onsite wastewater treatment plants at disposal facilities. A small fraction of the engine coolants used by DIY users may also be incorrectly disposed of to the sewers, drains, or the ground.

The biodegradability study conducted on the notified chemical shows that it is not readily biodegradable (0-3% degradation in 28 days). For details of the biodegradability study, please refer to Appendix C. The notified chemical released to sewers is expected to be degraded slowly through sewage treatment plants (STPs). The waste sludge from STPs is expected to be disposed of to landfill in accordance with local regulations. Because of its predicted low log P_{ow} (1.6), the notified chemical is not expected to bioaccumulate. The notified chemical in water, sludge and landfill is expected to eventually degrade through biotic and abiotic processes to form water and oxides of carbon, nitrogen and sulphur.

7.1.3. Predicted Environmental Concentration (PEC)
The predicted environmental concentration (PEC) has been calculated to assume the worst case scenario with 100% release of the notified chemical into sewer systems nationwide over 260 working days per annum. It is also assumed under the worst-case scenario that there is no removal of the notified chemical during sewage treatment processes.

Predicted Environmental Concentration (PEC) for the Aquatic Compartment		
Total Annual Import/Manufactured Volume	6,000	kg/year
Proportion expected to be released to sewer	100%	
Annual quantity of chemical released to sewer	6,000	kg/year
Days per year where release occurs	260	days/year
Daily chemical release:	23	kg/day
Water use	200	L/person/day
Population of Australia (Millions)	24.386	million
Removal within STP	0%	
Daily effluent production:	4,877	ML
Dilution Factor - River	1.0	
Dilution Factor - Ocean	10.0	
PEC - River:	4.73	µg/L
PEC - Ocean:	0.47	µg/L

STP effluent re-use for irrigation occurs throughout Australia. The agricultural irrigation application rate is assumed to be 1,000 L/m^2/year (10 ML/ha/year). The notified chemical in this volume is assumed to infiltrate and accumulate in the top 10 cm of soil (density 1,500 kg/m^3). Using these assumptions, irrigation with a concentration of 4.73 µg/L may potentially result in a soil concentration of approximately 31.5 µg/kg. Assuming accumulation of the notified chemical in soil for 5 and 10 years under repeated irrigation, the concentration of the notified chemical in the applied soil in 5 and 10 years may be approximately 157.7 µg/kg and 315 µg/kg, respectively.

7.2. Environmental Effects Assessment
The results from the ecotoxicological investigation conducted on the notified chemical are summarised in the table below. Details of these studies can be found in Appendix C.

Endpoint	*Result*	*Assessment Conclusion*
Fish Toxicity	96 h EC50 = 14.5 mg/L	Harmful to fish
Daphnia Toxicity	48 h EC50 > 100 mg/L	Not harmful to aquatic invertebrates
Algal Toxicity	72 h EC50 = 34.3 mg/L	Harmful to alga
Inhibition of Bacterial Respiration	3 h EC50 > 1,000 mg/L	Not inhibit microbial activity

Under the *Globally Harmonised System of Classification and Labelling of Chemicals* (GHS), the notified chemical is expected to be harmful to fish and alga. Therefore, the notified chemical is formally classified as "Acute Category 3; Harmful to aquatic life" under the GHS. Based on the acute toxicity and lack of readily biodegradation, the notified chemical is formally classified as "Chronic Category 3; Harmful to aquatic life with long lasting effects" under the GHS (United Nations, 2009).

7.2.1. Predicted No-Effect Concentration

The predicted no-effects concentration (PNEC) has been calculated based on the most sensitive endpoint for fish as shown in the table below. An assessment factor of 100 was used given the acute endpoint for three trophic levels is available.

Predicted No-Effect Concentration (PNEC) for the Aquatic Compartment		
Fish 96 h EC50	14.5	mg/L
Assessment Factor	100	
Mitigation Factor	1	
PNEC	145	µg/L

7.3. Environmental Risk Assessment

Based on the above predicted PEC and PNEC, the following Risk Quotient (Q = PEC/PNEC) has been calculated.

Risk Assessment	PEC µg/L	PNEC µg/L	Q
Q - River	4.73	145	**0.033**
Q - Ocean	0.47	145	**0.003**

The conservative risk quotient for discharge of effluents containing the notified chemical to the aquatic environment indicates that the notified chemical is unlikely to reach ecotoxicologically significant concentrations based on its annual importation quantity. Based on the predicted log P_{ow}, the notified chemical is not expected to be bioaccumulative. Therefore, on the basis of the predicted PEC/PNEC ratio, the maximum annual importation volume, and the assessed use pattern as a component of engine coolants and metalworking fluids, the notified chemical is not expected to pose an unreasonable risk to the aquatic environment.

APPENDIX A: PHYSICAL AND CHEMICAL PROPERTIES

Melting Point 156 – 158 °C

Method OECD TG 102 Melting Point/Melting Range
Remarks Differential scanning calorimetry (DSC)
Test Facility Clariant (2014a)

Boiling Point Not determinable; decomposed at ≥ 174 °C

Method OECD TG 103 Boiling Point
Remarks Differential scanning calorimetry (DSC); decomposed before reaching a boiling point
Test Facility Clariant (2014b)

Density 1,580 kg/m^3 at 27 °C

Method OECD TG 109 Density of Liquids and Solids
Remarks Air comparison pycnometer method
Test Facility Clariant (2014c)

Vapour Pressure < 1 × 10^{-6} kPa at 20 °C

Method Calculated using MPBPWin v1.43, based on chemical structure
Remarks The calculation algorithm is derived from a correlation of chemical structures with experimental vapour pressure. The calculated values were 2.5 × 10^{-5} Pa for solid and 7.6 × 10^{-4} Pa for subcooled liquid. The final result was quoted as < 0.001 Pa.
Test Facility Clariant (2014d)

Water Solubility 133 mg/L at 20 °C

Method OECD TG 105 Water Solubility
Remarks Flask Method
Test Facility Clariant (2014e)

Fat (or n-octanol) Solubility 12.6 g/L at 20 °C

Method OECD TG 105 Water Solubility (adapted for n-octanol)
Remarks Flask Method
Test Facility Clariant (2014f)

Hydrolysis as a Function of pH

Method OECD TG 111 Hydrolysis as a Function of pH

pH	*T (°C)*	*$t_{½}$ (year)*
4	25	>1
7	25	>1
9	25	>1

Remarks The notified chemical is considered hydrolytically stable.
Test Facility Clariant (2015)

Partition Coefficient (n-octanol/water) log P_{ow} = 1.60 at 23 °C

Method OECD TG 107 Partition Coefficient (n-octanol/water)
Remarks Flask Method
Test Facility Clariant (2014g)

Surface Tension 70.6 ± 0.9 mN/m at 20 °C

Method OECD TG 115 Surface Tension of Aqueous Solutions
Remarks Concentration: 90% of saturation concentration
Test Facility Clariant (2014h)

Particle Size D10: 4.8 µm
D50: 379.1 µm
D90: 1075.7 µm

Method ISO 13320 (2009): Particle size analysis – Laser diffraction methods

Range (µm)	*Mass (%)*
< 1	< 0.1
< 4	8.1
< 10	15.5
< 100	27.2

Remarks Malvern Mastersizer 2000 with Scirocco 2000 dispersing unit (Software version 5.54)
Test Facility Clariant (2014i)

Flammability (Solids) Not a highly flammable solid

Method EC Council Regulation No 440/2008 A.10 Flammability (Solids).
Remarks Only preliminary test was conducted and the notified chemical could not be ignited. Main test was considered by the study authors to be unnecessary.
Test Facility Siemens AG (2014)

APPENDIX B: TOXICOLOGICAL INVESTIGATIONS

B.1. Acute toxicity – oral

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 401 Acute Oral Toxicity EC Council Regulation No 440/2008 B.1 Acute Toxicity (Oral)
Species/Strain	Rat/Sprague-Dawley
Vehicle	0.5% Methylcellulose (400 cPs)
Remarks - Method	No GLP Certificate No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw)*	*Mortality*
1	10 (5 M/5 F)	500	0/10
2	10 (5 M/5 F)	1,000	3/10
3	10 (5 M/5 F)	1,500	3/10
4	10 (5 M/5 F)	2,000	6/10
5	10 (5 M/5 F)	2,500	8/10
6	10 (5 M/5 F)	3,000	10/10

LD50	1,580 mg/kg bw
Signs of Toxicity	All animals became hypoactive within 24 hours after receiving the notified chemical. Mortality of some animals occurred between 2 hours to 6 days after administering the notified chemical at dose levels between 1,000 to 3,000 mg/kg bw. Several animals became cachectic prior to death. At the highest tested dose level of 3,000 mg/kg bw, all animals died within 2 days of treatment. No mortalities occurred at the lowest tested dose level of 500 mg/kg bw.
Effects in Organs	Necropsy of the animals that died during the study period revealed hyperaemia, erosion and/or haemorrhagic ulceration of the mucosa in the stomach.
Remarks - Results	No abnormalities were found in the surviving animals.
CONCLUSION	The notified chemical is harmful via the oral route.
TEST FACILITY	Clariant (1985a)

B.2. Acute toxicity – dermal

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 402 Acute Dermal Toxicity
Species/Strain	Rat/Wistar (Hsd Han)
Vehicle	None. The notified chemical was directly applied.
Type of dressing	Semi-occlusive
Remarks - Method	GLP Certificate No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw)*	*Mortality*
1	10 (5 M/5 F)	2,000	0/10

LD50 > 2,000 mg/kg bw

Signs of Toxicity - Local	No abnormalities were noted.
Signs of Toxicity - Systemic	No abnormalities were noted.
Effects in Organs	None
Remarks - Results	The animals showed expected body weight gain over the observation period.

CONCLUSION The notified chemical is of low acute toxicity via the dermal route.

TEST FACILITY Advinus (2014)

B.3. Irritation – skin

TEST SUBSTANCE Notified chemical

METHOD OECD TG 404 Acute Dermal Irritation/Corrosion

Species/Strain	Rabbit/New Zealand White
Number of Animals	6
Vehicle	None. The notified chemical was directly applied.
Observation Period	72 hours
Type of Dressing	Occlusive
Remarks - Method	No GLP Certificate No significant protocol deviations

RESULTS No effects on the skin were noted.

Remarks - Results No signs of irritation were observed.

CONCLUSION The notified chemical is non-irritating to the skin.

TEST FACILITY Clariant (1985b)

B.4. Irritation – eye

TEST SUBSTANCE Notified chemical

METHOD OECD TG 405 Acute Eye Irritation/Corrosion.

Species/Strain	Rabbit/New Zealand White
Number of Animals	6
Observation Period	14 days
Remarks - Method	No GLP Certificate. Observation period was reduced to 14 days.

RESULTS

Lesion	*Mean Score**						*Maximum Value*	*Maximum Duration of Any Effect*	*Maximum Value at End of Observation Period*
	1	2	3	4	5	6			
Conjunctiva: redness	1.7	1.7	2	2	2	1.3	3	Not reversible within 14 days	2
Conjunctiva: chemosis	0.3	0.3	0.3	0.3	1	0.3	1	< 96 hours	0
Corneal opacity	0.3	0.3	0.3	0.3	1	0.7	4	Not reversible within 14 days	4
Iridial inflammation	0.7	0.7	1.3	1	1	0.3	2	Not reversible within 14 days	1

* Calculated on the basis of the scores at 24, 48, and 72 hours for EACH animals.

Remarks - Results No mortality or signs of systemic toxicity was observed.

Hyperaemia of the conjunctiva was observed in all animals. This effect was not reversible within 14 days for 2 animals.

Chemosis was observed in all animals within 24 hours after application, and this effect was reversible within 96 hours.

Diffuse corneal opacity areas were observed in all animals within 72 hours after application. This effect was reversible in 4 animals within 14 days, but progressed in the remaining 2 animals up to complete opacity in one case.

Hyperaemia of the iris was observed in all animals within 72 hours after application. This effect was reversible in 3 animals and not reversible in 2 animals within 14 days. The iris could not be evaluated for the one remaining animal due to the complete opacity of the cornea.

CONCLUSION The notified chemical is severely irritating to the eye.

TEST FACILITY Clariant (1985c)

B.5. Skin sensitisation

TEST SUBSTANCE Notified chemical

METHOD OECD TG 406 Skin Sensitisation – Guinea-Pig Maximization Test
EC Directive 440/2008 B.6 Skin Sensitisation – Guinea-Pig Maximization Test

Species/Strain Guinea pig/Dunkin Hartley

PRELIMINARY STUDY Maximum Non-irritating Concentration:
intradermal: 50% (highest concentration evaluated)
topical: 50% (highest concentration evaluated)

MAIN STUDY

Number of Animals Test Group: 10 Control Group: 5

Vehicle Polyethylene glycol 400 (PEG 400)

Positive control Not conducted in parallel with the test substance

INDUCTION PHASE Induction Concentration:
intradermal: 2%
topical: 50%

Signs of Irritation Tested in a preliminary irritation test

CHALLENGE PHASE topical: 50%

Remarks - Method No GLP Certificate
No significant protocol deviations

The test sites were pre-treated with 10% sodium lauryl sulphate before topical induction.

RESULTS

Animal	*Challenge Concentration*	*Number of Animals Showing Skin Reactions after Challenge*	
		24 h	*48 h*
Test Group	50%	0/10	0/10
Control Group	50%	0/5	0/5

Remarks - Results In the preliminary irritation test, signs of necrosis were observed after intradermal injection. These effects were considered to be caused by PEG and not by the notified chemical. This was confirmed by comparison with control animals which only received the vehicle.

No signs of irritation were observed after topical induction. No mortality or signs of systemic toxicity were observed in the animals used in the main

	study. The animals showed expected body weight gain over the observation period.
CONCLUSION	There was no evidence of reactions indicative of skin sensitisation to the notified chemical under the conditions of the test.
TEST FACILITY	WIL (2014)

B.6. Repeat dose oral toxicity – 14-day dose range finding study

TEST SUBSTANCE	Notified chemical
METHOD	Dose range finding study for subsequent OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents
Species/Strain	Rat/Wistar (Hsd Han)
Route of Administration	Oral – gavage
Exposure Information	Total exposure days: 14 days Dose regimen: 7 days per week
Vehicle	Water with 0.5% (w/v) carboxymethyl cellulose sodium salt (NaCMC)
Remarks - Method	GLP Certificate
	The study was conducted to determine the maximum tolerable dose (MTD) of the notified chemical and to define the dose levels for subsequent 28-day repeated dose oral toxicity study (see Appendix B.7.)

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
control	12 (6 M/6 F)	0	0/12
low dose	12 (6 M/6 F)	100	0/12
mid dose	12 (6 M/6 F)	300	4/12
high dose	12 (6 M/6 F)	1,000	12/12

Mortality and Time to Death

There were no unscheduled deaths for animals in the low dose groups.

Mortalities (4 of 12 animals) were observed in the mid dose group by Day 7. Due to their declining conditions, the remaining animals in the group were euthanized on Day 8.

All animals in the high dose group had died between Day 4 to 7.

Clinical Observations

No clinical signs were observed for animals in the low dose group.

In the mid dose group, animals exhibited clinical signs on Day 6 including hypoactivity, dehydration and piloerection. Animals in the high dose group exhibited clinical signs of hypoactivity, piloerection, dehydration, and ventral recumbent and tremors from Day 4.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

Changes in haematology and clinical chemistry parameters were noted in all groups, but these changes fell within historical control data.

Effects in Organs

In all animals in mid and high dose groups, dark red fluids were found in intestinal segments of the duodenum, jejunum, ileum and cecum, and multiple brown foci were found in glandular mucosa. Thinning of non-glandular mucosa and distended stomach were also observed.

Small and non-prominent thymus was observed in males in the mid and high dose groups and females in the high dose group. Distended urinary bladders were observed in all animals in the high dose group.

Remarks – Results
The clinical observations and effects in the organs of both male and female rats treated at the mid and high doses were considered by the study authors to be treatment related adverse effects.

The study authors observed statistically significant changes in the kidney and adrenal weights in the low-dose animals, but no toxicologically relevant findings can be established.

CONCLUSION
The MTD was considered to be 100 mg/kg bw/day in this study.

TEST FACILITY Advinus (2015a)

B.7. Repeat dose oral toxicity – 28-day (with recovery)

TEST SUBSTANCE Notified chemical

METHOD OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents
Species/Strain Rat/Wistar (Hsd Han)
Route of Administration Oral – gavage
Exposure Information Total exposure days: 28 days
Dose regimen: 7 days per week
Post-exposure observation period: 14 days
Vehicle Water with 0.5% (w/v) NaCMC
Remarks - Method GLP Certificate
No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
control	12 (6 M/6 F)	0	0/12
low dose	12 (6 M/6 F)	5	0/12
mid-intermediate dose	12 (6 M/6 F)	15	0/12
mid dose	12 (6 M/6 F)	40	0/12
high dose	12 (6 M/6 F)	100	0/12
control recovery	10 (5 M/5 F)	0	0/10
mid dose recovery	10 (5 M/5 F)	40	0/10
high dose recovery	10 (5 M/5 F)	100	0/10

Mortality and Time to Death
There were no unscheduled deaths for animals in all dose groups.

Clinical Observations
No clinical signs of toxicity were observed for animals in all dose groups. No test substance-related variations were observed in the mean body weights and net body weight gains in all dose groups in both sexes.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis
No toxicologically significant changes in haematology and urinalysis parameters were found for animals in all dose groups.

In high dose group, decreased cholesterol levels were observed in males and increased triglycerides were observed in females. These changes were statistically significant, but were reversible after the 14 days recovery period and were not considered by the study authors to have toxicological significance.

Effects in Organs
The kidney weight of males in the high dose group and females in the mid and high dose groups were statistically significantly higher than the vehicle control, but were not associated with any microscopic change and were reversible after the recovery period.

In the high dose group, an increased relative liver weight of all animals and a decreased absolute thymus weight of females were observed. These changes were not associated with any gross or microscopic changes. As these

weight changes were reversible after the 14 days recovery period, the changes were not considered by the study authors to have toxicological relevance.

Sperm Evaluation
There were no test substance-related changes in sperm parameters, including vas deferens sperm motility, cauda epididymal sperm counts and sperm morphology.

Remarks – Results
The authors stated that there were no toxicologically relevant adverse effects observed in animals treated up to 100 mg/kg bw/day. However, animals in the 40 and 100 mg/kg bw/day dose group showed a statistically significant increase in kidney and adrenal weights compared to the control group.

CONCLUSION
The No Observed Adverse Effect Level (NOAEL) was established as 100 mg/kg bw/day in this study, based on the highest dose level tested with no adverse effects observed.

TEST FACILITY Advinus (2015b)

B.8. Mutagenicity – bacterial reverse mutation

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 471 Bacterial Reverse Mutation Test EC Directive 2000/32/EC B.13/14 Mutagenicity – Reverse Mutation Test using Bacteria Pre incubation procedure
Species/Strain	*Salmonella typhimurium*: TA1535, TA1537, TA98, TA100 *Escherichia coli*: WP2uvrA
Metabolic Activation System	S9-Mix from phenobarbital (PB)/β-naphthoflavone (NF) induced rat liver
Concentration Range in Main Test	a) With metabolic activation: 313 – 5,000 µg/plate b) Without metabolic activation: 313 – 5,000 µg/plate
Vehicle	DMSO
Remarks - Method	No GLP Certificate No significant protocol deviations Preliminary test was designed for dose finding. Confirmation test was conducted in TA1537 only.

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/plate) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Preliminary Test	> 5,000	-	> 5,000	Negative
Main Test	-	> 5,000	> 5,000	Negative
Confirmation Test*	-	> 5,000	> 5,000	Negative
Present				
Preliminary Test	> 5,000	-	> 5,000	Negative
Main Test	-	> 5,000	> 5,000	Negative
Confirmation Test*	-	> 5,000	> 5,000	Negative

* Tested in TA1537 only

Remarks - Results	No test material precipitate was observed at any doses tested. No significant increases in the frequency of revertant colonies were recorded for any strains. The concurrent controls gave satisfactory responses confirming the validity of the test system.
CONCLUSION	The notified chemical was not mutagenic to bacteria under the conditions of the test.

TEST FACILITY CERI (1999)

B.9. Genotoxicity – *in vitro* mammalian chromosome aberration

TEST SUBSTANCE Notified chemical

METHOD OECD TG 473 *In vitro* Mammalian Chromosome Aberration Test
Species/Strain Chinese Hamster Cells
Cell Type/Cell Line Ovary cell line/CHO-K1
Metabolic Activation System S9 mix from Aroclor 1254-induced rat liver
Vehicle DMSO
Remarks - Method GLP certificate.

A dose range-finding study was carried out at 20 – 2,300 µg/mL. The dose selection for the main experiments was based on toxicity observed in the range-finding study and solubility test.

Vehicle and positive controls (ethyl methanesulfonate and cyclophosphamide) were run concurrently with the notified chemical.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	40, 126, 400	3 h	21 h
Test 2*	40, 126, 400	21 h	21 h
Present			
Test 1	50, 158, 500	3 h	21 h

* Confirmatory assay

All cultures and positive controls were selected for metaphase analysis.

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/mL) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	≥ 640	≥ 400	≥ 1,280*	Negative
Test 2	-	≥ 400	-	Negative
Present				
Test 1	≥ 640	≥ 500	≥ 1,280*	Negative

* Observed in preliminary test.

Remarks - Results The notified chemical did not cause any increase in the number of structurally aberrant metaphases including and excluding gaps at both sampling times either in the absence or presence of S9 mix. No increase in the frequency of cells containing numerical chromosome aberrations was noted.

The positive and vehicle controls gave satisfactory responses confirming the validity of the test system.

CONCLUSION The notified chemical was not clastogenic to Chinese hamster ovary cells treated *in vitro* under the conditions of the test.

TEST FACILITY Advinus (2015c)

B.10. Mutagenicity – *in vitro* mammalian cell gene mutation

TEST SUBSTANCE Notified chemical

METHOD OECD TG 476 *In vitro* Mammalian Cell Gene Mutation Test
Species/Strain Chinese Hamster Cells
Cell Type/Cell Line Ovary cell line/CHO-K1
Metabolic Activation System S9 mix from Aroclor 1254-induced rat liver
Vehicle DMSO
Remarks - Method GLP certificate.

A dose range-finding study was carried out at 20 – 2,300 µg/mL. The dose selection for the main experiments was based on toxicity observed in the range-finding study and solubility test.

Vehicle and positive controls (ethyl methanesulfonate and 3-methylcholanthrene) were run concurrently with the notified chemical.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Expression Time*	*Selection Time*
Absent				
Test 1	56, 167, 500, 1,500	3 h	9 days	10 days
Test 2	48, 150, 475, 1,500	3 h	9 days	10 days
Present				
Test 1	57, 180, 570, 1,800	3 h	9 days	10 days
Test 2	48, 150, 475, 1,500	3 h	9 days	10 days

All cultures were analysed for 6-thioguanine (6TG) resistant phenotype.

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/mL) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	≥ 2,300	> 1,500	≥ 1,280*	Negative
Test 2	-	> 1,500	-	Negative
Present				
Test 1	≥ 2,300	> 1,800	≥ 1,280*	Negative
Test 2	-	> 1,500	-	Negative

* Observed in preliminary test.

Remarks - Results The notified chemical did not lead to a statistically significant increase in the number of mutant colonies either in the presence or absence of S9 mix. The mutant frequencies at any concentration were within the range of the concurrent vehicle control and the historical negative control data.

The increase in the frequencies of mutant colonies induced by the positive control demonstrated the sensitivity of the test method and the metabolic activity of the S9 mix

CONCLUSION The notified chemical was not mutagenic to Chinese hamster ovary cells treated *in vitro* under the conditions of the test.

TEST FACILITY Advinus (2015d)

B.11. Toxicity to reproduction/development – screening test

TEST SUBSTANCE Notified chemical

METHOD OECD TG 421 Reproduction/Developmental Toxicity Screening Test
Species/Strain Rat/Wistar (Hsd Han)
Route of Administration Oral – gavage

Exposure Information	Exposure days: Males: Days 1-14 pre-mating, during mating, and Days 1-14 post-mating Females: Days 1-14 pre-mating, during mating, during pregnancy and lactation Days 1-13
Remarks – Method	GLP certificate No significant protocol variation

RESULTS

Group	*Number and Sex of Animals*	*Dose (mg/kg bw/day)*	*Mortality*
1 (vehicle control)	20 (10 M/10 F)	0	0/20
2	20 (10 M/10 F)	31.25	0/20
3	20 (10 M/10 F)	62.5	0/20
4	20 (10 M/10 F)	125	0/20

Mortality and Time to Death

No mortality was observed at all dose levels tested.

Effects on Parental (P) animals:

Systemic Toxicity

No treatment-related adverse effects were observed in females up to the highest dose tested.

At the highest dose level (125 mg/kg bw/day), decreased absolute and relative reticulocyte counts were observed in males. Decreased globulin concentration, body weight and thymus weight, and increased adrenal and kidney weights were observed. However, there were no microscopic changes observed in the kidney.

Reproductive Toxicity

No significant changes in the reproductive organs of males and females were observed.

For females treated at 125 mg/kg bw day, increased pre-implantation loss was observed, which resulted in reduced implantation index. Increased post-implantation loss was observed at the 62.5 and 125 mg/kw bw day dose groups, which resulted in a reduced mean litter size.

Effects on 1st Filial Generation (F1)

A reduction in the ano-genital distance was observed in female pups from the 31.25 mg/kg bw/day dose group and male pups from the 31.25 and 62.5 mg/kg bw/day dose groups. These changes were considered incidental by the study authors because of the lack of any dose relationship.

Survival rate of the pups was lower for all doses tested, due to cannibalism of pups by the dam from Days 1-4. No other developmental effects were noted in the surviving pups.

Remarks - Results

Increased consumption of water was observed in some animals. No significant changes were observed in mean body weight of the pups.

CONCLUSION

The No Observed Adverse Effect Levels (NOAELs) were established by the study authors as shown in the table.

Toxicity	*NOAEL (mg/kg bw/day)*	*Effect Observed at Higher Doses*
Male Systemic	62.5	Decreased reticulocyte count, decreased globulin concentration, decreased body weight, decreased thymus weight, increased adrenal weight and increased kidney weight
Female Systemic	125	Highest dose tested, no treatment-related adverse effects
Female Reproductive	31.25	Increased pre-implantation losses in the high dose group; increased post-implantation losses in the mid and high dose groups; reduced mean litter size in the mid and high dose groups

TEST FACILITY Advinus (2016)

APPENDIX C: ENVIRONMENTAL FATE AND ECOTOXICOLOGICAL INVESTIGATIONS

C.1. Environmental Fate

C.1.1. Ready biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 301 B Ready Biodegradability: CO2 Evolution Test.
Inoculum	Activated sludge from a local STP
Exposure Period	28 days
Auxiliary Solvent	None
Analytical Monitoring	CO_2
Remarks - Method	No significant deviations from the test guidelines were reported.

RESULTS

Test substance		*Sodium acetate*	
Day	*% Degradation*	*Day*	*% Degradation*
6	2	6	56
14	3	14	72
20	3	20	89
28	0	28	83

Remarks - Results	All validity criteria for the test were satisfied. The percentage degradation of the reference compound, sodium acetate surpassed the threshold level of 60% within 14 days indicating the suitability of the inoculums. The toxicity control exceeded 25% biodegradation after 14 days showing that toxicity was not a factor inhibiting the biodegradability of the test substance. The degree of degradation of the notified chemical after 28 days was 0-3%.
CONCLUSION	The notified chemical is not readily biodegradable.
TEST FACILITY	Dr U Noack Laboratorium (2001a)

C.2. Ecotoxicological Investigations

C.2.1. Acute toxicity to fish

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 203 Fish, Acute Toxicity Test - Static EC Council Regulation No 440/2008 C.1 Acute Toxicity for Fish - Static
Species	*Brachydanio rerio*
Exposure Period	96 hours
Auxiliary Solvent	None
Water Hardness	59 mg $CaCO_3$/L
Analytical Monitoring	Dissolved Organic Carbon (DOC) analyser
Remarks – Method	No significant deviations from the test guidelines were reported. The test substance was weighed and directly added into the 15 L glass aquaria with 10 L of dilution water before the start of the experiment.

RESULTS

Concentration mg/L		*Number of Fish*	*Mortality (%)*				
Nominal	*Actual*		*1 h*	*24 h*	*48 h*	*72 h*	*96 h*
Control	Control	7	0	0	0	0	0
6.25	Not determined	7	0	0	0	0	0
12.5	Not determined	7	0	0	0	0	0

Concentration mg/L		*Number of Fish*	*Mortality (%)*				
25	Not determined	7	0	0	0	0	14
50	Not determined	7	0	86	100	100	100
100	Not determined	7	100	100	100	100	100

LC50 14.5 mg/L at 96 hours (using Probit Analysis)

Remarks – Results All validity criteria for the test were satisfied.

CONCLUSION The notified chemical is harmful to fish.

TEST FACILITY Dr U Noack Laboratorium (2001b)

C.2.2. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE Notified chemical

METHOD OECD TG 202 Daphnia sp. Acute Immobilisation Test and Reproduction Test - Static

Species *Daphnia magna*
Exposure Period 48 hours
Auxiliary Solvent None
Water Hardness 269 mg $CaCO_3$/L
Analytical Monitoring High Performance Liquid Chromatography – Diode Array Detector (HPLC-DAD)
Remarks - Method No significant deviations from the test guidelines were reported. The stock solution of 100 mg/L was freshly prepared and mixed well before the start of the exposure.

RESULTS

Concentration mg/L		*Number of D. magna*	*Number Immobilised*	
Nominal	*Actual*		*24 h*	*48 h*
Control	< LOQ*	5	0	0
100	101	5	0	0

**Limit of Quantitation (LOQ) of 0.2 mg/L.*

LC50 > 100 mg/L at 48 hours

Remarks - Results All validity criteria for the test were satisfied.

CONCLUSION The notified chemical is not harmful to aquatic invertebrates.

TEST FACILITY Dr U Noack Laboratorium (2015a)

C.2.3. Algal growth inhibition test

TEST SUBSTANCE Notified chemical

METHOD OECD TG 201 Alga, Growth Inhibition Test
EC Council Regulation No 440/2008 C.3 Algal Inhibition Test

Species *Desmodesmus subspicatus*
Exposure Period 72 hours
Concentration Range Nominal: 0.12, 0.38, 1.20, 3.20, 12.0, 38.0, 120 mg/L
Actual: 0.129, 0.388, 1.20, 3.89, 12.2, 38.9, 121 mg/L
Auxiliary Solvent None
Water Hardness 0.24 mmol Ca+Mg/L (nominal)
Analytical Monitoring HPLC-DAD
Remarks - Method No significant deviations from the test guidelines were reported. The stock solution of 120 mg/L was freshly prepared with dilution water before the

start of the experiment.

RESULTS

Biomass		*Growth*	
EC50	*NOEC*	*EC50*	*NOEC*
mg/L at 72 h	*mg/L*	*mg/L at 72 h*	*mg/L*
34.3	12.0	16.9	3.80

Remarks - Results All validity criteria for the test were satisfied.

CONCLUSION The notified chemical is harmful to alga.

TEST FACILITY Dr U Noack Laboratorium (2015b)

C.2.4. Inhibition of microbial activity

TEST SUBSTANCE Notified chemical

METHOD OECD TG 209 Activated Sludge, Respiration Inhibition Test
Inoculum Activated sludge from a local STP
Exposure Period 3 hours
Concentration Range Nominal: 10, 32, 100, 320, 1000 mg/L
Remarks – Method No significant deviations from the test guidelines were reported. The test substance was weighed out directly in Erlenmeyer flasks before the start of the experiment.

RESULTS
IC50 > 1,000 mg/L

Remarks – Results All validity criteria for the test were satisfied.

CONCLUSION The notified chemical does not inhibit microbial activity.

TEST FACILITY Dr U Noack Laboratorium (2015c)

BIBLIOGRAPHY

ACD/Labs (2017) I-Lab 2.0 - ilab.acdlabs.com, Algorithm version: v5.0.0.184 (accessed on 30th March 2017). ACD/Labs.

Advinus (2014) [Notified chemical]: Acute dermal toxicity study in Wistar Rats (Study No. G9833, December 2014). Bangalore, India, Advinus Therapeutics Ltd (Unpublished report submitted by the notifier).

Advinus (2015a) [Notified chemical]: 14-Day repeated dose toxicity study by oral gavage in Wistar Rats (Study No. G9836, February 2015). Bangalore, India, Advinus Therapeutics Ltd (Unpublished report submitted by the notifier).

Advinus (2015b) [Notified chemical]: 28-Day oral toxicity study in Wistar Rats followed by 14-day recovery period (Study No. G9837, October 2015). Bangalore, India, Advinus Therapeutics Ltd (Unpublished report submitted by the notifier).

Advinus (2015c) [Notified chemical]: *in vitro* mammalian chromosome aberration test in CHO cells (Study No. G9834, January 2015). Bangalore, India, Advinus Therapeutics Ltd (Unpublished report submitted by the notifier).

Advinus (2015d) [Notified chemical]: *in vitro* mammalian gene mutation test in CHO cells (Study No. G9835, January 2015). Bangalore, India, Advinus Therapeutics Ltd (Unpublished report submitted by the notifier).

Advinus (2016) Reproduction/developmental toxicity screening test of [notified chemical] in Wistar rats by oral gavage (Study No. G9838, December 2016). Bangalore, India, Advinus Therapeutics Ltd (Unpublished report submitted by the notifier).

CERI (2007) Mutagenicity test of [notified chemical] using microorganisms (Report No. T-5609). Hita, Japan, Chemicals Evaluation and Research Institute (Unpublished report submitted by the notifier).

Clariant (1985a) Untersuchung der akuten oralen Toxizitat unterschiedlich hoher Einmaldosen des [notified chemical] bei der Ratte (RBM-Gutachten NR. 2165, July 1985). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier – in German with English translation in IUCLID).

Clariant (1985b) Prufung der pimaren Hautreizung nach einmaliger epikutaner Applikation des [notified chemical] beim New Zealand White-Kaninchen (RBM-Gutachten NR. 2164, July 1985). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier – in German with English translation in IUCLID).

Clariant (1985c) Prufung der Augenreizung nach intraokularer Applikation einer Einzeldosis des [notified chemical] beim New Zealand White-Kaninchen (RBM-Gutachten NR. 2163, July 1985). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier – in German with English translation in IUCLID).

Clariant (2014a) [Notified chemical] – Melting Point (Report No. 14-163043-2, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014b) [Notified chemical] – Boiling Point State of a Substance (Report No. 14-163043-1, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014c) [Notified chemical] – Density (Report No. 14-163043-3, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014d) [Notified chemical] – Calculated property: Vapour pressure (Report No. 14-163043-6, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014e) [Notified chemical] – Water Solubility (Report No. 14-174153-1, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014f) [Notified chemical] – Solubility in n-Octonal (Report No. 14-174153-4, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014g) [Notified chemical] – Partition Coefficient (n-octanol/water): Shake Flask Method (Report No. 14-174153-2, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014h) [Notified chemical] – Surface Tension (Report No. 14-163043.4, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2014i) [Notified chemical] – Granulometry (Report No. 14-163043-5, September 2014). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Clariant (2015) [Notified chemical] – Hydrolysis as a Function of pH (Report No. 15-190587, July 2015). Frankfurt am main, Germany, Clariant Produkte (Deutschland) GmbH (Unpublished report submitted by the notifier).

Dr U Noack Laboratorium (2001a) [Notified chemical] – Ready Biodegradability Modified Sturm Test (Study No. AST77551, March, 2001). Sarstedt, Germany, Dr U Noack Laboratorium Für Angewandte Biologie, (Unpublished report submitted by the notifier).

Dr U Noack Laboratorium (2001b) [Notified chemical] – Zebra Fish Acute Toxicity Test, Static, 96 Hours (Study No. FAZ77551, January, 2001). Sarstedt, Germany, Dr U Noack Laboratorium Für Angewandte Biologie, (Unpublished report submitted by the notifier).

Dr U Noack Laboratorium (2015a) [Notified chemical] – Acute Immobilisation Test to Daphnia magna, Static, 48 Hours, Limit-Test (Study ID 141022CB/DAI16284, September, 2015). Sarstedt, Germany, Dr U Noack Laboratorium Für Angewandte Biologie, (Unpublished report submitted by the notifier).

Dr U Noack Laboratorium (2015b) [Notified chemical] – Alga, Growth Inhibition Test with *Desmodesmus subspicatus*, 72 Hours (Study ID 141022CB/SSO16284, September, 2015). Sarstedt, Germany, Dr U Noack Laboratorium Für Angewandte Biologie, (Unpublished report submitted by the notifier).

Dr U Noack Laboratorium (2015c) [Notified chemical] – Respiration Inhibition Test with Activated Sludge (Study ID 141022CB/BBR16284, March, 2015). Sarstedt, Germany, Dr U Noack Laboratorium Für Angewandte Biologie, (Unpublished report submitted by the notifier).

NTC (2017) Australian Code for the Transport of Dangerous Goods by Road and Rail (ADG code), Edition 7.5, National Transport Commission, Commonwealth of Australia.

Siemens AG (2014) [Notified chemical] – Flammability (Solids) N.1. & A.10 (Report No. 20140212.01, July 2014). Frankfurt am main, Germany, Siemens AG (Unpublished report submitted by the notifier).

SWA (2012) Code of Practice: Managing Risks of Hazardous Chemicals in the Workplace, Safe Work Australia, https://www.safeworkaustralia.gov.au/system/files/documents/1702/managing_risks_of_hazardous_chemicals2.pdf

United Nations (2009) Globally Harmonised System of Classification and Labelling of Chemicals (GHS), 3rd revised edition. United Nations Economic Commission for Europe (UN/ECE), http://www.unece.org/trans/danger/publi/ghs/ghs_rev03/03files_e.html.

US EPA (2012) Estimation Programs Interface (EPI) SuiteTM for Microsoft® Windows, v 4.10. United States Environmental Protection Agency. Washington DC, USA.

WIL (2014) Assessment of contact hypersensitivity to [notified chemical] in the Albino Guinea Pig (Maximisation-Test) (Project 506717, November 2014). 's-Hertogenbosch, The Netherlands, WIL Research Europe B.V. (Unpublished report submitted by the notifier).