File No: LTD/1931

September 2016

NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

PUBLIC REPORT

2-Oxiraneacetic acid, 3-ethyl-, 1-(3,3-dimethylcyclohexyl)ethyl ester

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:

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Director
NICNAS

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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
LTD/1931	Firmenich Pty Ltd	2-Oxiraneacetic acid, 3-ethyl-, 1-(3,3-dimethylcyclohexyl)ethyl ester	Yes	< 1 tonne per annum	Fragrance ingredient

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
Sensitisation, Skin (Category 1)	H317 – May cause an allergic skin reaction

Based on the available information, the notified chemical is recommended for hazard classification according to the *Approved Criteria for Classifying Hazardous Substances* (NOHSC, 2004), with the following risk phrases:

R22: Harmful if swallowed
R43: May cause skin sensitisation by skin contact

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 2	H401 – Toxic to aquatic life
Chronic Category 2	H411 – Toxic 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.

Based on the available information, when used at ≤ 1% concentration in air fresheners and at ≤ 0.47% concentration in cosmetic and other household products, 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
  - Sensitisation, Skin (Category 1): H317 – May cause an allergic skin reaction

  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.

- The Delegate (and/or the Advisory Committee on Chemicals Scheduling) should consider the notified chemical for listing on the SUSMP.

Health Surveillance

- As the notified chemical is a skin sensitiser, employers should carry out health surveillance for any worker who has been identified in the workplace risk assessment as having a significant risk of skin sensitisation.

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, automated processes, where possible
  - Adequate 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 during reformulation:
  - Avoid contact with skin and eyes
  - Avoid inhalation

- 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 during reformulation:
  - Coveralls
  - Impervious gloves
  - Eye protection
  - Respiratory protection, if inhalation exposure may occur

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

- A copy of the (M)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.

Emergency procedures

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

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 importation volume exceeds one tonne per annum notified chemical;
- the concentration of the notified chemical exceeds or is intended to exceed 0.47% in cosmetic and household products and 1% in air fresheners;

or

(2) Under Section 64(2) of the Act; if
- the function or use of the chemical has changed from a fragrance ingredient, 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.

(Material) Safety Data Sheet
The (M)SDS of the notified chemical provided by the notifier was reviewed by NICNAS. The accuracy of the information on the (M)SDS remains the responsibility of the applicant.

ASSESSMENT DETAILS

1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT(S)
Firmenich Pty Ltd (ABN: 86 002 964 794)
73 Kenneth Road
BALGOWLAH NSW 2093

NOTIFICATION CATEGORY
Limited-small volume: Chemical other than polymer (1 tonne or less per year).

EXEMPT INFORMATION (SECTION 75 OF THE ACT)
Data items and details claimed exempt from publication: other names, analytical data, degree of purity, impurities, additives/adjuvants, use details, identity of manufacturer.

VARIATION OF DATA REQUIREMENTS (SECTION 24 OF THE ACT)
No variation to the schedule of data requirements is claimed.

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT(S)
None

NOTIFICATION IN OTHER COUNTRIES
None

2. IDENTITY OF CHEMICAL

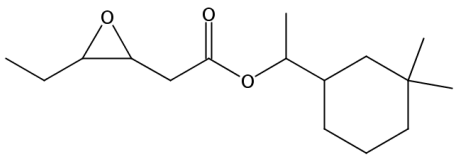
MARKETING NAME(S)
2-Oxiraneacetic acid, 3-ethyl-, 1-(3,3-dimethylcyclohexyl)ethyl ester

CAS NUMBER
1643921-90-7

CHEMICAL NAME
2-Oxiraneacetic acid, 3-ethyl-, 1-(3,3-dimethylcyclohexyl)ethyl ester

MOLECULAR FORMULA
$C_{16}H_{28}O_3$

STRUCTURAL FORMULA



MOLECULAR WEIGHT
268.39 Da

ANALYTICAL DATA
Reference NMR, FTIR, GC-MS and UV-Vis spectra were provided.

3. COMPOSITION

DEGREE OF PURITY
70-90%

4. PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE AT 20 °C AND 101.3 kPa: colourless liquid

Property	Value	Data Source/Justification
Melting Point/Freezing Point	-75.4 °C	Measured
Boiling Point	286-291 °C at 101.3 kPa	Measured
Density	969.2 kg/m^3 at 20 °C	Measured
Vapour Pressure	1.3×10^{-5} kg/m^3 at 20 °C 2.9×10^{-5} kg/m^3 at 25 °C 8.6×10^{-4} kg/m^3 at 50 °C	Measured
Water Solubility	0.0106 g/L at 20 °C	Measured
Hydrolysis as a Function of pH	Hydrolytically stable at pH 5 – 7	Measured
Partition Coefficient (n-octanol/water)	log Pow = 4.24	Measured
Surface Tension	50.48 mN/m at 20 °C	Measured
Adsorption/Desorption	log K_{oc} = 3.07 – 3.11	Calculated
Dissociation Constant	Not determined	No dissociable functions
Flash Point	148 °C at 101 kPa	Measured
Flammability	Not determined	Not expected to be flammable based on the measured flash point
Autoignition Temperature	255 °C	Measured
Explosive Properties	Predicted negative	Contains no functional groups that imply explosive properties
Oxidising Properties	Predicted negative	Contains no functional groups that 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 within Australia. The notified chemical will be imported into Australia as a component of fragrance formulations at ≤ 10% concentration.

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

Year	*1*	*2*	*3*	*4*	*5*
Tonnes	< 1	< 1	< 1	< 1	< 1

PORT OF ENTRY
Sydney (by air or sea)

IDENTITY OF RECIPIENTS
Firmenich Pty Ltd

TRANSPORTATION AND PACKAGING
The notified chemical will be imported as a component of fragrance formulations at ≤ 10% concentration in lacquered drums of sizes ranging 5-180 kg. Finished consumer products containing ≤ 1% notified chemical will be transported primarily by road to retail stores in packages suitable for retail sale.

USE
The notified chemical will be used as a fragrance ingredient in cosmetic and household products (including air fresheners, all-purpose cleaners, cleaning products and laundry products). The content in the final consumer products will vary, with the proposed usage concentrations of ≤ 1% for air fresheners and ≤ 0.47% for cosmetic products and other household products.

OPERATION DESCRIPTION
The notified chemical will be imported as a component of fragrance formulations at ≤ 10% concentration for reformulation into cosmetic and household products.

Reformulation
The procedures for reformulating the fragrance formula containing the notified chemical will likely vary depending on the nature of the cosmetic/household products, and may involve both automated and manual transfer steps. In general, it is expected that the reformulation processes will involve blending operations that will normally be automated and occur in an enclosed system, followed by automated filling of the finished products into consumer containers of various sizes.

End-use
The finished products containing the notified chemical at ≤ 1% concentration may be used by consumers and professionals such as hairdressers, workers in beauty salons or cleaners. Depending on the nature of the products, these could be applied in a number of ways, such as by hand, using an applicator or by spray.

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 workers	unknown	unknown
Mixer	4	2
Drum handling	4	2
Drum cleaning	4	2
Maintenance	4	2
Quality control	0.5	1
Packaging	4	2
Professional end users	Not specified	Not specified

EXPOSURE DETAILS

Transport and storage
Transport and storage workers may come into contact with the notified chemical either at ≤ 10% concentration in fragrance formulations or at ≤ 1% concentration in consumer products only in the event of an unlikely accidental rupture of containers.

Reformulation
During reformulation into cosmetic products, dermal, ocular and inhalation exposure of workers to the notified chemical at ≤ 10% concentration may occur. Exposure is expected to be minimised through the use of exhaust ventilation and/or automated/enclosed systems as well as through the use of personal protective equipment (PPE) such as coveralls, eye protection, impervious gloves and respiratory protection (as appropriate).

End use
Exposure to the notified chemical in end-use products at ≤ 1% concentration may occur in professions where the services provided involve the application of cosmetic products to clients (e.g. hair dressers, workers in beauty salons), or the use of household products in the cleaning industry. The principal route of exposure will be dermal, while ocular and inhalation exposure is also possible. Such workers may use some PPE to minimise repeated exposure and good hygiene practices are expected to be in place. If PPE is used, exposure of such

workers is expected to be of a similar or lesser extent than that experienced by consumers using products containing the notified chemical.

6.1.2. Public Exposure

There will be widespread and repeated exposure of the public to the notified chemical through the use of cosmetic and household products (at ≤ 0.47% concentration) and air fresheners (at ≤ 1% concentration). The principal route of exposure will be dermal, while ocular and inhalation exposure is also possible, particularly if products are applied by spray.

A combined internal dose of 1.1285 mg/kg bw/day was estimated using data on typical use patterns of cosmetic product categories in which the notified chemical may be used (SCCS, 2012; Cadby *et al.*, 2002; Loretz *et. al.*, 2006; ACI, 2010; specific use details of the notified chemical are considered as exempt information). This estimation assumed a worst case scenario and is for a person who is a simultaneous user of a selection of cosmetic and household products that may contain the notified chemical.

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 = 300 - 2000 mg/kg bw; harmful
Rat, acute dermal toxicity	LD50 > 2000 mg/kg bw; low toxicity
Rat, acute inhalation toxicity	LC50 > 5.14 mg/L/4 hour; low toxicity
Skin irritation (*in vitro*)	non-irritating
Rabbit, skin irritation	non-irritating
Rabbit, eye irritation	slightly irritating
Mouse, skin sensitisation – Local lymph node assay	evidence of sensitisation (EC_3 = 22%)
Rat, repeat dose oral toxicity – 28 days	NOAEL = 1000 mg/kg bw/day
Mutagenicity – bacterial reverse mutation	non mutagenic
Genotoxicity – in vitro chromosome aberration	non genotoxic

Toxicokinetics

No toxicokinetic data on the notified chemical were submitted. Based on the low molecular weight (< 500 Da), water solubility (1.06×10^{-2} g/L) and partition coefficient (log Pow = 4.24) of the notified chemical, absorption across biological membranes may occur.

Acute toxicity

The notified chemical was found to be harmful via the oral route in an acute toxicity study conducted in rats. Two out of 6 animals treated at 2000 mg/kg bw/day died prematurely. Clinical signs prior to death included piloerection and elevated gait, hunched posture and loose faeces. Treatment-related clinical signs including salivation, chin rubbing, under activity, piloerection, elevated gait and loose faeces were also noted in 1 surviving animal treated at 2000 mg/kg bw/day.

The notified chemical was found to be of low acute dermal and inhalation toxicity in studies conducted in rats.

Irritation and sensitisation

The notified chemical was found to be non-irritating to the skin in a study conducted in rabbits and in an *in vitro* study conducted using the reconstructed human epidermis model.

The notified chemical was found to be slightly irritating to the eyes in a study conducted in rabbits.

The notified chemical was found to be sensitising in a Local Lymph Node Assay. The EC_3 value was calculated to be 22%.

Repeated dose toxicity

An repeated dose oral (gavage) toxicity study on the notified chemical was conducted in rats, in which the notified chemical was administered at 30, 300 and 1,000 mg/kg bw/day for 28 consecutive days, with a 14-day recovery period for high dose and control animals.

The No Observed Adverse Effect Level (NOAEL) was established as 1000 mg/kg bw/day (the highest dose tested) in the study, based on treatment-related effects were either adaptive changes (not associated with any histopathological changes and showed recovery in the recovery period), or not toxicologically significant.

Mutagenicity/Genotoxicity
The notified chemical was negative in a bacterial reverse mutation assay and in an *in vitro* chromosomal aberration study in human peripheral lymphocytes.

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
Sensitisation, Skin (Category 1)	H317 – May cause an allergic skin reaction

Based on the available information, the notified chemical is recommended for hazard classification according to the *Approved Criteria for Classifying Hazardous Substances* (NOHSC, 2004), with the following risk phrases:

R22: Harmful if swallowed
R43: May cause skin sensitisation by skin contact

6.3. Human Health Risk Characterisation

6.3.1. Occupational Health and Safety
Based on the available information the critical health effects of the notified chemical are acute oral toxicity and skin sensitisation.

Reformulation
During reformulation workers may be at risk of sensitisation when handling the notified chemical at ≤ 10% concentration. It is anticipated by the notifier that engineering controls such as enclosed and automated processes and local ventilation will be implemented where possible, and appropriate PPE (coveralls, imperious gloves, eye protection and respiratory protection) will be used to limit workers exposure.

Therefore, under the occupational settings described, the risk to the health of workers from use of the notified chemical is not considered to be unreasonable.

End-use
Cleaners, hair and beauty care professionals will handle the notified chemical at ≤ 1% concentration. Such professionals may use PPE to minimise repeated exposure, and good hygiene practices are expected to be in place. If PPE is used, exposure of such workers is expected to be of a similar or lesser extent than that experienced by consumers using products containing the notified chemical (for details of the public health risk assessment, see Section 6.3.2).

6.3.2. Public Health
Members of the public may be repeatedly exposed to the notified chemical during the use of cosmetic products (≤ 0.47% concentration), air fresheners (≤ 1% concentration) and other household products (≤ 0.47% concentration).

Sensitisation
Proposed methods for the quantitative risk assessment of dermal sensitisation have been the subject of significant discussion (see for example, Api *et al.*, 2008 and RIVM, 2010). Using fine fragrance as an example product that may contain the notified chemical (at 0.47% concentration), as a worst case scenario, the Consumer Exposure Level (CEL) for the notified chemical is estimated to be 17.63 μg/cm^2/day (Cadby *et al.,* 2002). When tested in an LLNA study, the notified chemical was a skin sensitiser with an EC_3 value of 22%. Consideration of each of the studies and application of appropriate safety factors, allowed the derivation of an Acceptable Exposure Level (AEL) of 17.77 μg/cm^2. In this instance, the factors employed included an interspecies factor

(3), intraspecies factor (10), a matrix factor (3.16), a use/time factor (3.16) and database factor (1), giving an overall safety factor of ~300.

As the AEL > CEL, the risk to the public of the induction of sensitisation that is associated with the use of fine fragrances (a worst case example of a leave-on cosmetic product) is not considered to be unreasonable. Based on the significantly lower expected exposure level from other leave-on cosmetic products, rinse-off products and household products, by inference, the risk of induction of sensitisation associated with the use of these products is also not considered to be unreasonable. It is acknowledged that consumers may be exposed to multiple products containing the notified chemical, and a quantitative assessment based on aggregate exposure has not been conducted.

Repeated-dose toxicity
The repeat dose toxicity potential was estimated by calculation of the margin of exposure (MoE) of the notified chemical using the worst case exposure scenario from use of multiple products of 1.1285 mg/kg bw/day (see Section 6.1.2). Using a NOAEL of 1000 mg/kg bw/day derived from a 28-day repeated dose oral toxicity study on the notified chemical, the margin of exposure (MOE) was estimated to be 886. A MOE value ≥ 100 is generally considered to be acceptable for taking into account intra- and inter-species differences.

Therefore, based on the information available, the risk to the public associated with use of the notified chemical at ≤ 1% concentration in air fresheners and at ≤ 0.47% concentration in cosmetic and other household products 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 not be manufactured in Australia, so there will be no environmental release associated with this activity. The notified chemical will be imported into Australia as a component of fragrance formulations that will be further reformulated into end-use cosmetic and household cleaning products. In the event of a spill, the notified chemical is expected to be contained and collected in an inert absorbent material and disposed of in accordance with local regulations.

A typical blending operation will be highly automated in a fully enclosed/contained environment. Potential sources of release include spills, equipment washing, and container residues. A total of 0.1% of waste may be generated as a result of spills. It is expected that equipment will be cleaned using water and the washings reused for subsequent operations. The average amount of residue in empty containers after removal by vacuum pump is estimated to be < 0.1%. Therefore, a total of < 0.2% (2 kg) of waste will be generated each year from reformulation processes.

RELEASE OF CHEMICAL FROM USE
The notified chemical will enter the aquatic compartment during use of the various products into which it will be incorporated. Cosmetic products are expected to be washed off the hair and skin and will enter the aquatic environment diluted in water. Cleaning products will also be diluted in water and will enter the aquatic environment. It is anticipated that the majority of the notified chemical released will enter into sewer systems. It is estimated that a maximum of 3% (30 kg) of the consumer products may remain in the consumer containers that will be sent for disposal.

RELEASE OF CHEMICAL FROM DISPOSAL
Empty containers containing the notified chemical at blending facilities will be recycled or disposed of through an approved waste management facility. Empty product containers are expected to be disposed of to landfill.

7.1.2. Environmental Fate
Following its use in Australia, the majority of the notified chemical is expected to enter the sewer system through its use as a component of cosmetics and household cleaning products before potential release to surface waters nationwide. The notified chemical is not considered to be readily biodegradable (41% in 28 days), but exhibited substantial biodegradation after 28 days. For details of the environmental fate studies, please refer to Appendix C. The calculated adsorption/desorption coefficient (log K_{oc} = 3.08 – 3.11) indicates that the notified

chemical may sorb to soil and sediment in the sludge fraction. In either landfill or water, the notified chemical will ultimately decompose to water and oxides of carbon. The notified chemical is expected to have potential for bioaccumulation in aquatic organisms given its low molecular weight and high log Pow.

The half-life of the notified chemical in air is calculated to be 8.04 hours based on reactions with hydroxyl radicals (AOPWIN v1.92; US EPA, 2011). Therefore, in the event of release to atmosphere, the notified chemical is not expected to persist in the atmospheric compartment.

The majority of the notified chemical will be released to sewer after use. A small proportion of the notified chemical may be applied to land when effluent is used for irrigation, or when sewage sludge is used for soil remediation, or disposed to landfill as collected spills and empty containers. The notified chemical has low water solubility and predicted to be hydrophobic. Therefore, in the waste water treatment processes in the sewage treatment plant (STP), most of the notified chemical is expected to partition to sludge or to suspended solids where it will be removed for disposal to landfill. In landfill the notified chemical is expected to slowly decompose by abiotic and biotic processes to form water and oxides of carbon. Therefore, the notified chemical is not expected to be bioavailable to aquatic organisms despite its potential for bioaccumulation.

7.1.3. Predicted Environmental Concentration (PEC)

The calculation for the predicted environmental concentration (PEC) is summarised in the table below. Based on the reported uses in cosmetic products and cleaning products, it is conservatively assumed that 100% of the notified chemical will be released to sewer on a nationwide basis over 365 days per year. It is also assumed that under a worst-case scenario there is no removal of the notified chemical during STP processes.

Predicted Environmental Concentration (PEC) for the Aquatic Compartment		
Total Annual Import/Manufactured Volume	1,000	kg/year
Proportion expected to be released to sewer	100%	
Annual quantity of chemical released to sewer	1,000	kg/year
Days per year where release occurs	365	days/year
Daily chemical release:	2.74	kg/day
Water use	200.0	L/person/day
Population of Australia (Millions)	22.613	million
Removal within STP	0%	
Daily effluent production:	4,523	ML
Dilution Factor - River	1.0	
Dilution Factor - Ocean	10.0	
PEC - River:	0.61	µg/L
PEC - Ocean:	0.06	µg/L

Partitioning to biosolids in STPs Australia-wide may result in an average biosolids concentration of 1.272 mg/kg (dry wt). Biosolids are applied to agricultural soils, with an assumed average rate of 10 t/ha/year. Assuming a soil bulk density of 1500 kg/m^3 and a soil-mixing zone of 10 cm, the concentration of the notified chemical may approximate 0.008 mg/kg in applied soil. This assumes that degradation of the notified chemical occurs in the soil within 1 year from application. Assuming accumulation of the notified chemical in soil for 5 and 10 years under repeated biosolids application, the concentration of notified chemical in the applied soil in 5 and 10 years may approximate 0.04 mg/kg and 0.08 mg/kg, respectively.

7.2. Environmental Effects Assessment

The results from ecotoxicological investigations conducted on the notified chemical are summarised in the table below. The provided studies include acute toxicity of the notified chemical to fish, aquatic invertebrates and algae, and inhibition of activated sludge. Details of these studies can be found in Appendix C.

Endpoint	*Result*	*Assessment Conclusion*
Fish Toxicity	LL50 = 12.2 mg/L	Harmful to fish
Daphnia Toxicity	EC50 = 3.18 mg/L	Toxic to aquatic invertebrates
Algal Toxicity	EC50 > 5.92 mg/L	Not harmful to algae up to the limit of its solubility
Inhibition of Bacterial Respiration	EC50 > 1000 mg/L	Not inhibitory to bacterial respiration

Based on the ecotoxicological endpoints for the notified chemical, it is expected to be harmful to fish, toxic to daphnids, and not harmful to algae on an acute basis. Therefore, under the Globally Harmonised System of Classification and Labelling of Chemicals (GHS) (United Nations, 2009), the notified chemical is formally classified as "Acute Category 2; Toxic to aquatic life". Based on the acute toxicity and biodegradability of the notified chemical, it is formally classified as "Chronic Category 2; Toxic to aquatic life with long lasting effects" under the GHS.

7.2.1. Predicted No-Effect Concentration

The predicted no-effects concentration (PNEC) has been calculated from the most sensitive endpoint for *Daphnia*. A safety factor of 100 was used given acute endpoints for three tropic levels are available.

Predicted No-Effect Concentration (PNEC) for the Aquatic Compartment		
EC50 (Invertebrates)	3.18	mg/L
Assessment Factor	100	
PNEC:	31.8	µg/L

7.3. Environmental Risk Assessment

Based on the above PEC and PNEC, the following Risk Quotient has been calculated:

Risk Assessment	PEC µg/L	PNEC µg/L	RQ
RQ - River	0.61	31.8	0.019
RQ - Ocean	0.06	31.8	0.002

The risk quotient for discharge of treated effluents containing the notified chemical to the aquatic environment (RQ < 1) indicates that the notified chemical is unlikely to reach ecotoxicologically significant concentrations in surface waters based on its maximum annual importation quantity. The notified chemical is not expected to be readily biodegradable in the environment but is expected to ultimately biodegrade. Therefore, the notified chemical is unlikely to result in ecotoxicologically significant concentrations in the aquatic environment on the basis of the PEC/PNEC ratio, maximum annual importation volume and assessed use pattern.

APPENDIX A: PHYSICAL AND CHEMICAL PROPERTIES

Melting Point/Freezing Point -75.4 °C at 101.3 kPa

Method	OECD TG 102 Melting Point/Melting Range.
Remarks	Determined by differential scanning calorimetry
Test Facility	Consilab (2014a)

Boiling Point 286-291 °C at 101.3 kPa

Method	OECD TG 103 Boiling Point.
Remarks	Determined by differential scanning calorimetry
Test Facility	Consilab (2014a)

Density 969.2 kg/m^3 at 20 °C

Method	OECD TG 109 Density of Liquids and Solids.
Remarks	Pycnometer method
Test Facility	Dr U Noack-Laboratorien (2014a)

Vapour Pressure 1.3×10^{-5} kg/m^3 at 20 °C
2.9×10^{-5} kg/m^3 at 25 °C
8.6×10^{-4} kg/m^3 at 50 °C

Method	OECD TG 104 Vapour Pressure.
Remarks	Determined using a vapour pressure balance
Test Facility	Consilab (2014b)

Water Solubility 0.0106 g/L at 20 °C

Method	OECD TG 105 Water Solubility
Remarks	Flask Method. Quantification was conducted by GC-MS based on the sum of four isomer peaks.
Test Facility	Dr U Noack-Laboratorien (2016)

Hydrolysis as a Function of pH Hydrolytically stable at pH 5 – 7

Method	Internal method
Remarks	The test media were standard aqueous buffers at pH 2, pH 5, pH 7, pH 8.5 and pH 12 containing 1% non-ionic surfactant. The tests were done in accelerated conditions at 40 °C over approximately one month. Analyses were conducted by GC-FID.
Test Facility	Firmenich (2012)

Partition Coefficient (n-octanol/water) log Pow = 4.24

Method	OECD TG 117 Partition Coefficient (n-octanol/water)
Remarks	HPLC Method.
Test Facility	Firmenich (2010)

Surface Tension 50.48 mN/m at 20 °C

Method	OECD TG 115 Surface Tension of Aqueous Solutions.
Remarks	Concentration: 90% saturated aqueous solution
Test Facility	Dr U Noack-Laboratorien (2014b)

Flash Point 148 °C at 101 kPa

Method	EC Council Regulation No 440/2008 A.9 Flash Point.
Remarks	Closed cup method

Test Facility Consilab (2014c)

Autoignition Temperature 255 °C

Method EC Council Regulation No 440/2008 A.15 Auto-Ignition Temperature (Liquids and Gases).
Test Facility Consilab (2014d)

APPENDIX B: TOXICOLOGICAL INVESTIGATIONS

B.1. Acute toxicity – oral

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 423 Acute Oral Toxicity – Acute Toxic Class Method.
Species/Strain	Rat/Crl:CD (SD)
Vehicle	Corn oil
Remarks - Method	No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	3F	300	0/3
2	3F	300	0/3
3	3F	2000	0/3
4	3F	2000	2/3

LD50	300-2000 mg/kg bw
Signs of Toxicity	Two animals treated at 2000 mg/kg died on Days 2 and 7, respectively. Clinical signs prior to death included piloerection and elevated gait, hunched posture and loose faeces. Treatment-related clinical signs including salivation, chin rubbing, under activity, piloerection, elevated gait and loose faeces were noted in 1 surviving animal at 2000 mg/kg. No clinical signs were noted in animals at 300 mg/kg or in the remaining animals at 2000 mg/kg.
Effects in Organs	Macroscopic examination of the animals died prematurely showed congestion (characterised by darkened tissues/organs) of the subcutaneous tissue, lungs, liver, spleen and kidneys, clear fluid content in the thoracic cavity, pallor of the stomach, small caecum, yellow fluid content in the small intestine and duodenum, enlarged spleen, red fluid content in the duodenum and gaseous distension in the small and large intestine, yellow fluid content in the stomach, gaseous distension in the duodenum and yellow fluid content in the large intestine. Macroscopic examination at study termination on Day 15 showed pallor of the liver and kidneys in one female treated at 2000 mg/kg. No abnormalities were noted in other animals.
Remarks - Results	A slight bodyweight loss or low body weight gain was noted between Days 8 and 15 for most animals at 300 mg/kg, which was not considered by the study authors to be treatment-related as no such effects were seen at 2000 mg/kg.
CONCLUSION	The notified chemical is harmful via the oral route.
TEST FACILITY	HLS (2014)

B.2. Acute toxicity – dermal

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 402 Acute Dermal Toxicity – Limit Test.
Species/Strain	Rat/Wistar
Vehicle	None
Type of dressing	Semi-occlusive
Remarks - Method	No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	5M, 5F	2000	0/10

LD50	> 2000 mg/kg bw
Signs of Toxicity - Local	No signs of dermal irritation were noted.
Signs of Toxicity - Systemic	No signs of systemic toxicity were noted.
Effects in Organs	No abnormalities were noted at necropsy.
Remarks - Results	All animals showed expected body weight gains.

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

TEST FACILITY Harlan (2014a)

B.3. Acute toxicity – inhalation

TEST SUBSTANCE Notified chemical

METHOD OECD TG 403 Acute Inhalation Toxicity.

Species/Strain	Rat/RccHam:WIST
Vehicle	None
Method of Exposure	Nose-only
Exposure Period	4 hours
Physical Form	Liquid aerosol
Particle Size	1.85 µm (mean mass median aerodynamic diameter); % < 4 µm: 77.7%
Remarks - Method	No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Concentration <mg/L>*		*Mortality*
		Nominal	*Actual*	
1	5M, 5F	14.1	5.14	0/10

LC50	> 5.14 mg/L/4 hours
Signs of Toxicity	All animals showed increased respiratory rate, hunched posture, pilo-erection and wet fur. All animals appeared normal on Day 5.
Effects in Organs	No abnormalities were noted at necropsy.
Remarks - Results	Seven animals showed body weight losses or no body weight gain on Day 1. Three animals showed no body weight gains from Days 1 to 3 and 2 animals showed slight body weight losses from Days 3 to 7. Body weight gains were noted in all animals during the final week of recovery, with the exception of 1 animal which had a slight body weight loss.

CONCLUSION The notified chemical is of low toxicity via inhalation.

TEST FACILITY Harlan (2014b)

B.4. Irritation – skin (in vitro)

TEST SUBSTANCE Notified chemical

METHOD OECD TG 439 In vitro Skin Irritation: Reconstructed Human *Epidermis* Test Method
EPISKIN™ Reconstructed Human Epidermis Model

Vehicle	None
Remarks - Method	No significant protocol deviations. In a preliminary test the test substance was shown not to directly reduce MTT.

The test substance (10 µL) was applied to the tissues in triplicate. Following exposure period of 15 minutes (room temperature), the tissues were rinsed, treated with MTT and then incubated at 37 °C for 42 hours.

Negative and positive controls were run in parallel with the test substance:
- Negative control: Dulbecco's phosphate buffered saline
- Positive control: 5% aqueous sodium dodecyl sulphate

RESULTS

Test material	*Mean OD_{562} of triplicate tissues*	*Relative mean Viability (%)*	*SD of relative mean viability*
Negative control	0.980	100	4.7
Test substance	1.023	104.4	16.7
Positive control	0.054	5.5	0.4

OD = optical density; SD = standard deviation

Remarks - Results: The relative mean viability of the tissues treated with the test substance was > 50% (predicted as non-irritant according to the criteria).

The positive and negative controls gave satisfactory results, confirming the validities of the test systems.

CONCLUSION: The notified chemical was non-irritating to the skin under the conditions of the test.

TEST FACILITY: Harlan (2014c)

B.5. Irritation – skin

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 404 Acute Dermal Irritation/Corrosion.

Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Vehicle	None
Observation Period	72 hours
Type of Dressing	Semi-occlusive
Remarks - Method	No significant protocol deviations

RESULTS

Lesion	*Mean Score* Animal No.*			*Maximum Value*	*Maximum Duration of Any Effect*	*Maximum Value at End of Observation Period*
	1	2	3			
Erythema/Eschar	0	0	0	1	< 1 hour	0
Oedema	0	0	0	1	< 1 hour	0

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

Remarks - Results: Very slight erythema and oedema were noted at all treated skin sites immediately after patch removal which disappeared within 1 hour.

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

TEST FACILITY: Harlan (2014d)

B.6. Irritation – eye

TEST SUBSTANCE Notified chemical

METHOD OECD TG 405 Acute Eye Irritation/Corrosion.
Species/Strain Rabbit/New Zealand White
Number of Animals 3
Observation Period 72 hours
Remarks - Method No significant protocol deviations

RESULTS

Lesion	*Mean Score* Animal No.*			*Maximum Value*	*Maximum Duration of Any Effect*	*Maximum Value at End of Observation Period*
	1	2	3			
Conjunctiva: redness	0.3	0.7	0.7	2	< 72 hours	0
Conjunctiva: chemosis	0	0.7	0.7	2	< 72 hours	0
Conjunctiva: discharge	0	0.3	0.3	2	< 48 hours	0
Corneal opacity	0	0	0	0	-	0
Iridial inflammation	0	0	0	1	< 24 hours	0

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

Remarks - Results No corneal effects were noted. Iridial inflammation was noted in two treated eyes one hour after treatment.

Moderate conjunctival irritation was noted in all treated eyes one hour after treatment. Minimal conjunctival irritation was noted in all treated eyes at the 24-hour observation and in two treated eyes at the 48-hour observation. All signs of irritation were resolved at the 72-hour observation.

All animals showed expected body weight gains.

CONCLUSION The notified chemical is slightly irritating to the eye.

TEST FACILITY Harlan (2015a)

B.7. Skin sensitisation – mouse local lymph node assay (LLNA)

TEST SUBSTANCE Notified chemical

METHOD OECD TG 429 Skin Sensitisation: Local Lymph Node Assay
Species/Strain Mouse/CBA/Ca
Vehicle Acetone/olive oil (4:1)
Preliminary study Yes
Positive control α-Hexyl cinnamaldehyde
Remarks - Method No significant protocol deviations. A preliminary test was conducted on 1 mouse. Initial main tests were conducted at 25%, 50% and 100% concentrations. In order to determine the concentration of the test substance expected to cause a 3 fold increase in ^{3}HTdR incorporation (EC3 value), additional tests were conducted at 1%, 10% and 25% concentrations.

RESULTS

Main Test - Initial Test

Concentration (% w/w)	*Number and sex of animals*	*Proliferative response (DPM/lymph node)*	*Stimulation Index (Test/Control Ratio)*
Test Substance			
0 (vehicle control)	5F	1296.34 ± 177.60	1.00

25	5F	4979.59 ± 1754.69	3.84
50	5F	4218.96 ± 1623.65	3.25
100	5F	13050.28 ± 1020.38	10.07

Main Test - Additional Test

Concentration (% w/w)	*Number and sex of animals*	*Proliferative response (DPM/lymph node)*	*Stimulation Index (Test/Control Ratio)*
Test Substance			
0 (vehicle control)	5F	1950.77 ± 851.53	1.00
1	5F	3025.68 ± 907.54	1.55
10	5F	3012.33 ± 1705.57	1.54
25	5F	6561.92 ± 2717.95	3.36
Positive Control			
25	5F	14024.29 ± 2103.42	7.19

EC3 22%

Remarks - Results There were no premature deaths, signs of systemic toxicity, local skin irritation or marked increase in ear thickness noted in the test or control animals.

CONCLUSION There was evidence of induction of a lymphocyte proliferative response indicative of skin sensitisation to the notified chemical.

TEST FACILITY Harlan (2015b)

B.8. Repeat dose toxicity

TEST SUBSTANCE Notified chemical

METHOD OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents.

Species/Strain Rat/Crl:CD(SD)

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 Corn oil

Remarks - Method No significant protocol deviations

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw/day*	*Mortality*
control	5 per sex	0	0/10
low dose	5 per sex	30	0/10
mid dose	5 per sex	300	0/10
high dose	5 per sex	1000	0/10
control recovery	5 per sex	0	0/10
high dose recovery	5 per sex	1000	0/10

Mortality and Time to Death

There were no unscheduled deaths.

Clinical Observations

No toxicologically significant clinical signs of systemic toxicity were noted. There were no treatment-related changes in grip strength, motor activity and body weight gains. Higher than control high beam motor activity scores for male animals at 1000 mg/kg/day indicated an increase in rearing activity in Week 4. The values were within the testing facility's background range from previous studies and the difference was considered by the study authors to be unlikely to be treatment-related.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

There were increased cholesterol and triglyceride levels, disturbances in liver enzyme levels and increased liver weights, principally in female animals but also in male animals at 1000 mg/kg/day and to a lesser extent at 300 mg/kg/day. All changes showed full recovery and in the absence of any corresponding pathological findings in the liver these changes were considered by the study authors to be adaptive and not adverse.

Reduced urine output with high specific gravity, protein and sodium levels and reduced pH were noted in both sexes at Week 4 and appeared similar to control at the end of the recovery. There was no associated change in water consumption. The aetiology of these findings was unknown and in the absence of any pathological findings in the kidney these changes were not considered by the study authors to be of toxicological significance.

Higher than control group mean monocyte, large unstained cell and platelet counts were noted for animals at 1000 mg/kg/day and were inconsistent between the sexes. The changes were not considered by the study authors to represent adverse toxicity as they did not correspond with changes in any other parameters.

Effects in Organs

Organ weights

Group mean body weight adjusted kidney weights were statistically significant higher than control for all treated male groups and appeared comparable to control following the recovery period. Group mean body weight adjusted liver weights were statistically significantly higher than control for female animals at 300 mg/kg/day or 1000 mg/kg/day and male animals at 1000 mg/kg/day and remained statistically significantly higher than control for female animals; however, there was evidence of recovery in the male animals.

Necropsy

No treatment-related lesions were noted at necropsy following treatment or recovery.

Histopathology

Hyaline droplets were noted in the kidneys of male animals at 1000 mg/kg/day after treatment. The study authors stated that the presence of hyaline droplets in the cortical tubules of male kidneys is the early indicator of hydrocarbon neuropathy, which is commonly seen in mature male rats after administration of volatile hydrocarbons. The droplets are considered to be the result of reversible binding of the notified chemical to α2-microglobulin, and this complex being resistant to proteolytic hydrolysis leading to accumulation within the tubular cell lysoma in the kidney. The study authors stated that the finding did not represent adverse toxicity as hyaline droplet production is not relevant to humans and consequently histopathological investigations were not extended to include rats from the low or intermediate dose groups or for recovery groups.

CONCLUSION

The No Observed Adverse Effect Level (NOAEL) was established as 1000 mg/kg bw/day in this study, based on treatment-related effects were either adaptive changes (not associated with any histopathological changes and showed recovery in the recovery period), or not toxicologically significant.

TEST FACILITY	Envigo (2016)

B.9. Genotoxicity – bacteria

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 471 Bacterial Reverse Mutation Test. Pre incubation procedure
Species/Strain	*S. typhimurium*: TA1535, TA1537, TA98, TA100 *E. coli*: WP2uvrA
Metabolic Activation System	S9 mix from phenobarbital/β-naphthoflavone induced rat liver
Concentration Range in Main Test	a) With metabolic activation: 0.5-500 µg/plate (TA100, TA1537), 0.15-150 µg/plate (TA98, TA1535); 1.5-1500 µg/plate (WP2uvrA) b) Without metabolic activation: 0.05-50 µg/plate (TA1535, TA1537, TA100, TA98); 0.5-500 µg/plate (WP2uvrA)
Vehicle	Dimethyl sulphoxide
Remarks - Method	No significant protocol deviations. The dose selection for the main test was based on the toxicity results in the preliminary test.

Positive controls:
With metabolic activation: 2-aminoanthracene (TA1535, TA1537, TA100, WP2uvrA); benzo(a)pyrene (TA98)
Without metabolic activation: N-ethyl-N'-nitro-N-nitrosoguanidine [TA1535, TA100, WP2uvrA]; 9-aminoacridine (TA1537); 4-nitroquinoline-1-oxide (TA98)

RESULTS

Metabolic Activation	*Test Substance Concentration (μg/plate) Resulting in:* *Cytotoxicity in Preliminary Test*	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	> 5	> 5	> 50 (TA1535, TA1537, TA100, TA98); > 500 (WP2uvrA)	Negative
Present				
Test 1	> 50	> 50	> 500 (TA100, TA1537), > 150 (TA98, TA1535); > 1500 (WP2uvrA)	Negative

Remarks - Results
No significant increases in the frequency of revertant colonies were noted for any of the bacterial strains, with any dose of the test substance, either with or without metabolic activation.

The positive and negative controls gave a satisfactory response confirming the validity of the test system.

CONCLUSION
The notified chemical was not mutagenic to bacteria under the conditions of the test.

TEST FACILITY
Harlan (2014e)

B.10. Genotoxicity – in vitro

TEST SUBSTANCE
Notified chemical

METHOD
OECD TG 473 In vitro Mammalian Chromosome Aberration Test.
Species/Strain
Human
Cell Type/Cell Line
Peripheral lymphocytes
Metabolic Activation System
S9 mix from phenobarbitone/β-naphthoflavone induced rat livers
Vehicle
Dimethyl sulphoxide
Remarks - Method
No significant protocol deviations. Whole blood cultures were used. A dose range-finding study was carried out at 10.48-2684 μg/mL. The dose selection for the main experiments (both short-term exposure groups and continuous exposure group) was based on the toxicity results in the range-finding study.

Vehicle and positive controls (mitomycin C and cyclophosphamide) were run concurrently with the notified chemical.

Metabolic Activation	*Test Substance Concentration (μg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	10, 20, 30*, 40*, 60*, 80*	4h	24h
Test 2	10, 20*, 30*, 40*, 60*, 80	24h	24h
Present			
Test 1	10, 20, 40, 50*, 100*, 120*, 180*	4h	24h
Test 2	10, 20, 40, 50*, 100*, 120*, 180*	4h	24h

*Cultures 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	> 20.97	> 60	> 80	Negative
Test 2	> 20.97	> 40	> 80	Negative
Present				
Test 1	> 83.88	> 120	> 180	Negative
Test 2		> 120	> 180	Negative

* Indicated by > 50% reduction in mitotic index

Remarks - Results	In Test 1, haemolysis was observed at the end of exposure at ≥ 10 µg/mL and ≥ 40 µg/mL in the absence and presence of metabolic activation, respectively. In Test 2, haemolysis was observed at the end of exposure at ≥ 60 µg/mL and ≥ 100 µg/mL in the absence and presence of metabolic activation, respectively. It was stated by the study authors that haemolysis was an indication of a toxic response to erythrocytes and not indicative of any genotoxic response to the lymphocytes. In both main tests, no statistically significant increases in the frequency of cells with structural or numerical chromosome aberrations were noted in the presence or absence of metabolic activation. The positive and negative controls gave a satisfactory response confirming the validity of the test system.
CONCLUSION	The notified chemical was not clastogenic to human peripheral lymphocytes treated *in vitro* under the conditions of the test.
TEST FACILITY	Harlan (2015c)

APPENDIX C: ENVIRONMENTAL FATE AND ECOTOXICOLOGICAL INVESTIGATIONS

C.1. Environmental Fate

C.1.1. Ready biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 301 C Ready Biodegradability: Modified MITI Test (I).
Inoculum	Activated sludge
Exposure Period	28 days
Auxiliary Solvent	Ethyl acetate
Analytical Monitoring	TOC, GC, GC-MS, LC-MS
Remarks - Method	The test was conducted in accordance with the test guidelines specified above with no significant deviations reported. A reference test was conducted with aniline to confirm the sludge was sufficiently active.

RESULTS

Test substance		*Aniline*	
Day	*% Degradation*	*Day*	*% Degradation*
-	-	7	83
28	41*	14	99

*Calculated by BOD

Remarks - Results	All validity criteria of the test guideline were satisfied. None of the test substance remained at the end of the test, however several degradants were detected. The pass level of 60% biodegradation was not reached by the end of the test and therefore the notified chemical is not considered to be ready biodegradable.
CONCLUSION	The notified chemical is not ready biodegradable
TEST FACILITY	CERI (2015)

C.2. Ecotoxicological Investigations

C.2.1. Acute toxicity to fish

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 203 Fish, Acute Toxicity Test – Semi static
Species	*Danio rerio* (Zebrafish)
Exposure Period	96 hours
Auxiliary Solvent	None reported
Water Hardness	118 mg $CaCO_3$/L
Analytical Monitoring	GC
Remarks – Method	The study was carried out according to the test guideline above with no significant deviations reported. For each test concentration, test substance was added to test water to give the desired loading rate. The solution was then stirred for 24 hours and allowed to stand for 2 hours. The aqueous phase (water accommodated fraction, WAF) was removed by mid-depth siphoning. Microscopic inspection of the WAF showed no micro-dispersions or undissolved test item to be present. A reference test was conducted with potassium dichromate.

RESULTS

Concentration mg/L		*Number of Fish*	*Cumulative Mortality (%)*				
Nominal	*Actual**		*3 h*	*24 h*	*48 h*	*72 h*	*96 h*
Blank Control	-	10	0	0	0	0	0
7.80	4.28	10	0	0	0	0	0
11.0	5.86	10	0	20	20	20	20
15.4	7.16	10	0	100	100	100	100
21.4	8.34	10	0	100	100	100	100
30.0	9.16	10	0	100	100	100	100

*Geometric mean, measured in the period 0-24h

LL50	12.2 mg/L at 96 hours (WAF)
LL0	7.80 mg/L at 96 hours (WAF)
Remarks – Results	The 24 h LC50 value of the reference test was 252 mg/L which was within the prescribed concentration range 200 – 400 mg/L. All validity criteria of the test guideline were satisfied. The study results were based on nominal loading rates.
CONCLUSION	The notified chemical is harmful to fish
TEST FACILITY	Guangdong Detection Center of Microbiology (2015)

C.2.2. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 202 Daphnia sp. Acute Immobilisation Test and Reproduction Test – Semi Static
Species	*Daphnia magna*
Exposure Period	48 hours
Auxiliary Solvent	None reported
Water Hardness	160 – 180 mg $CaCO_3$/L
Analytical Monitoring	GC-MS
Remarks - Method	The study was carried out according to the test guideline above with no significant deviations reported. A reference test was conducted with potassium dichromate.

RESULTS

Concentration mg/L		*Number of D. magna*	*Percent Immobilised (mean value)*	
Nominal	*Actual**		*24 h*	*48 h*
Control	< LOQ**	4 × 5	0	0
0.5	0.392	4 × 5	0	0
1	0.860	4 × 5	0	0
2	1.65	4 × 5	0	0
4	2.79	4 × 5	25	35
8	6.56	4 × 5	75	95

*Geometric mean
**LOQ = limit of quantification (0.1 mg/L test substance)

EC50	3.18 mg/L at 48 hours
Remarks - Results	The EC50 value of the reference test was 2.01 mg/L which was within the prescribed concentration range 0.6 – 2.4 mg/L. All validity criteria of the test guideline were satisfied.
CONCLUSION	The notified chemical is toxic to aquatic invertebrates
TEST FACILITY	Dr U Noack-Laboratorien (2015a)

C.2.3. Algal growth inhibition test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 201 Alga, Growth Inhibition Test – Static
Species	*Pseudokirchneriella subcapitata*
Exposure Period	72 hours
Concentration Range	Nominal: 0.5 – 8.0 mg/L Actual: 0.319 – 5.92 mg/L
Auxiliary Solvent	None reported
Water Hardness	0.24 mmol Ca + Mg/L
Analytical Monitoring	GC-MS/MS
Remarks - Method	The study was carried out according to the test guideline above with no significant deviations reported. The study was carried out in closed bottles without headspace to avoid losses of the test substance. A reference test was conducted with potassium dichromate.

RESULTS

Biomass		*Growth*	
NOEC *mg/L at 72 h*	*EC50* *mg/L*	*NOEC* *mg/L at 72 h*	*EC50* *mg/L*
2.60	> 5.92	1.35	> 5.92

Remarks - Results	The study satisfied all the validity criteria of the guideline. The E_rC50 values of the reference test were 0.749 and 0.774 mg/L with and without headspace, respectively. These values were within the prescribed concentration range of 0.821± 0.388 mg/L. All effect values are based on the geometric mean of measured test substance concentrations.
CONCLUSION	The notified chemical is not harmful to algae up to the limit of its solubility in water.
TEST FACILITY	Dr U Noack-Laboratorien (2015b)

C.2.4. Inhibition of microbial activity

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test – Static
Inoculum	Activated sludge
Exposure Period	3 hours
Concentration Range	Nominal: 10 – 1000 mg/L Actual: Not reported
Remarks – Method	The test was conducted in accordance with the test guideline above, with no significant deviation to the test protocol reported. A reference test was carried out with copper (II) sulfate pentahydrate.
RESULTS	
IC50	> 1000 mg/L
NOEC	32 mg/L
Remarks – Results	The study satisfied all the validity criteria of the guideline except the mean specific oxygen uptake rate of control replicates (18 mg O_2 / g h) was slightly below the validity threshold (20 mg O_2 / g h). This deviation was not considered to affect the quality or integrity of the study. In the reference test an EC50 of 96.2 mg/L was obtained, which is in the recommended validity range of 53 – 155 mg/L.
CONCLUSION	The notified chemical is not inhibitory to microbial respiration
TEST FACILITY	Dr U Noack-Laboratorien (2014c)

BIBLIOGRAPHY

Api AM, Basketter DA, Cadby PA, Cano MF, Ellis G, Gerberick GF, Griem P, McNamee PM, Ryan CA and Safford R (2008) Dermal Sensitisation Quantitative Risk Assessment (QRA) for Fragrance Ingredients, Regulatory Toxicology and Pharmacology, 52:3-23.

Cadby, PA., Troy, WR., Vey, MGH. (2002) Consumer Exposure to Fragrance Ingredients: Providing Estimates for Safety Evaluation. Regulatory Toxicology and Pharmacology, 36:246-52.

CERI (2015) Biodegradation Study of [Notified Chemical] (Study No. 16170, June, 2015). Kurume, Japan, Chemicals Evaluation and Research Institute (Unpublished report submitted by the notifier).

Consilab (2014a) [Notified Chemical]: Determination of Physico-Chemical Properties – Melting Point (EC A.1. and OECD 102) and Boiling Point (EC A.2. and OECD 103) (Study No. CSL-14-0218.01, November, 2014). Frankfurt am Main, Germany, consilab Gesellschaft für Anlagensicherheit, (unpublished report submitted by the notifier).

Consilab (2014b) [Notified Chemical]: Determination of Physico-Chemical Properties – Vapour Pressure (EC A.4. and OECD 104) (Study No. CSL-14-0218.02, November, 2014). Frankfurt am Main, Germany, consilab Gesellschaft für Anlagensicherheit, (unpublished report submitted by the notifier).

Consilab (2014c) [Notified Chemical]: Determination of Physico-Chemical Properties – Flash point (EC A.9.) (Study No. CSL-14-0218.03, November, 2014). Frankfurt am Main, Germany, consilab Gesellschaft für Anlagensicherheit, (unpublished report submitted by the notifier).

Consilab (2014d) [Notified Chemical]: Determination of Physico-Chemical Properties – Auto-Ignition Temperature (liquids and Gases) (EC A.15.) (Study No. CSL-14-0218.04, November, 2014). Frankfurt am Main, Germany, consilab Gesellschaft für Anlagensicherheit, (unpublished report submitted by the notifier).

Dr U Noack-Laboratorien (2014a) [Notified Chemical]: Determination of the Density / Relative Density (Study No. CPD15827, May, 2014). Sarstedt, Germany, Dr U Noack-Laboratorien (Unpublished report submitted by the notifier).

Dr U Noack-Laboratorien (2014b) [Notified Chemical]: Determination of Surface Tension (Study No. CPT15827, August, 2014). Sarstedt, Germany, Dr U Noack-Laboratorien (Unpublished report submitted by the notifier).

Dr U Noack-Laboratorien (2014c) [Notified Chemical]: Respiration Inhibition Test with Activated Sludge (Study No. BBR15827, September, 2015). Sarstedt, Germany, Dr U Noack-Laboratorien (Unpublished report submitted by the notifier).

Dr U Noack-Laboratorien (2015a) [Notified Chemical]: Acute Immobilisation Test to Daphnia magna, Semi-static, 48 h. (Study No. DAI15827, June, 2015). Sarstedt, Germany, Dr U Noack-Laboratorien (Unpublished report submitted by the notifier).

Dr U Noack-Laboratorien (2015b) [Notified Chemical]: Alga, Growth Inhibition Test with *Pseudokirchneriella subcapitata*, 72 hours (Study No. SPO15827, September, 2015). Sarstedt, Germany, Dr U Noack-Laboratorien (Unpublished report submitted by the notifier).

Dr U Noack-Laboratorien (2016) [Notified Chemical]: Water Solubility (Flask Method) (Study No. 131022FG/CWF15827, 31 May 2016). Sarstedt, Germany, Dr U Noack-Laboratorien (Unpublished report submitted by the notifier).

Envigo (2016) [Notified Chemical]: Toxicity Study by Oral Administration to CD Rats for 4 Weeks Followed by a 2 Week Recovery Period (Study No. HIK0036, January, 2016). Huntingdon, Cambridgeshire, UK, Envigo CRS limited (Unpublished report submitted by the notifier).

Firmenich (2010) Partition Coefficient “Log Pow” (Study No. 105907-DMR, 13 April 2010) Geneva, Switzerland, Firmenich SA Geneva (Unpublished report submitted by the notifier).

Firmenich (2012) Stability Test of Perfumery Raw Materials (No Study No., April 2012) Geneva, Switzerland, Firmenich SA Geneva (Unpublished report submitted by the notifier).

Guangdong Detection Center of Microbiology (2015) Acute Toxicity Test of [Notified Chemical] with Zebra fish (*Danio rerio*) (Study No. 2014ESG0111, July, 2015) Guangzhou, China (Unpublished report submitted by the notifier).

HLS (2014) [Notified Chemical]: Acute Oral Toxicity to the Rat (Acute Toxic Class Method) (Study No. HIK0034, June, 2014). Huntingdon, Cambridgeshire, UK, Huntingdon Life Sciences (Unpublished report submitted by the notifier).

Harlan (2014a) [Notified Chemical]: Acute Dermal Toxicity (Limit Test) in the Rat (Study No. 41304114, October, 2014). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2014b) [Notified Chemical]: Acute Inhalation Toxicity (Nose Only) Study in the Rat (Study No. 41304113, November, 2014). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2014c) [Notified Chemical]: Determination of Skin Irritation Potential Using the EPISKIN Reconstructed Epidermis Model (Study No. 41304108, July, 2014). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2014d) [Notified Chemical]: Acute Dermal Irritation in the Rabbit (Study No. 41304110, October, 2014). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2014e) [Notified Chemical]: Reverse Mutation Assay 'Ames Test' Using *Salmonella typhimurium* and *Escherichia coli* (Study No. 41303929, May, 2014). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2015a) [Notified Chemical]: Acute Eye Irritation in the Rabbit (Study No. 41304111, January, 2015). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2015b) [Notified Chemical]: Local Lymph Node Assay in the Mouse – Individual Method (Study No. 41304112, March, 2015). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

Harlan (2015c) [Notified Chemical]: Chromosome Aberration Test in Human Lymphocytes *in vitro* (Study No. 41304115, February, 2015). Shardlow, Derbyshire, UK, Harlan Laboratories Ltd (Unpublished report submitted by the notifier).

NOHSC (2004) Approved Criteria for Classifying Hazardous Substances, 3rd edition [NOHSC:1008(2004)]. National Occupational Health and Safety Commission, Canberra, AusInfo.

RIVM (2010) Observations on the Methodology for Quantitative Risk Assessment of Dermal Allergens, Report 320015003/2010, National Institute for Public Health and the Environment.

SCCS (2012) Notes of Guidance for testing of Cosmetic Ingredients and Their Safety Evaluation (7th revision) European Commission - Scientific Committee on Consumer Safety.

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>.