File No: STD/1450

March 2013

**NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)**

# PUBLIC REPORT

## Peracid in KX-6228

This Assessment has been compiled in accordance with the provisions of the *Industrial Chemicals (Notification and Assessment) Act 1989* (Cwlth) (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 Ageing, and conducts the risk assessment for public health and occupational health and safety. The assessment of environmental risk is conducted by the Department of Sustainability, Environment, Water, Population and Communities.

For the purposes of subsection 78(1) of the Act, this Public Report may be inspected at our NICNAS office by appointment only at Level 7, 260 Elizabeth Street, Surry Hills NSW 2010.

This Public Report is also 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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# TABLE OF CONTENTS


SUMMARY ..... 3
CONCLUSIONS AND REGULATORY OBLIGATIONS ..... 3
ASSESSMENT DETAILS ..... 5
1. APPLICANT AND NOTIFICATION DETAILS ..... 5
2. IDENTITY OF CHEMICAL ..... 6
3. COMPOSITION ..... 6
4. PHYSICAL AND CHEMICAL PROPERTIES ..... 6
5. INTRODUCTION AND USE INFORMATION ..... 7
6. HUMAN HEALTH IMPLICATIONS ..... 8
6.1. Exposure Assessment ..... 8
6.1.1. Occupational Exposure ..... 8
6.1.2. Public Exposure ..... 9
6.2. Human Health Effects Assessment ..... 9
6.3. Human Health Risk Characterisation ..... 10
6.3.1. Occupational Health and Safety ..... 10
6.3.2. Public Health ..... 11
7. ENVIRONMENTAL IMPLICATIONS ..... 11
7.1. Environmental Exposure & Fate Assessment ..... 11
7.1.1. Environmental Exposure ..... 11
7.1.2. Environmental Fate ..... 12
7.1.3. Predicted Environmental Concentration (PEC) ..... 12
7.2. Environmental Effects Assessment ..... 12
7.2.1. Predicted No-Effect Concentration ..... 13
7.3. Environmental Risk Assessment ..... 13
APPENDIX A: PHYSICAL AND CHEMICAL PROPERTIES ..... 14
APPENDIX B: TOXICOLOGICAL INVESTIGATIONS ..... 16
B.1. Acute toxicity – oral ..... 16
B.2. Irritation – skin (in vitro) ..... 16
B.3. Repeat dose toxicity ..... 17
B.4. Genotoxicity – bacteria ..... 18
B.5. Genotoxicity – in vitro ..... 19
B.6. Developmental toxicity ..... 20
APPENDIX C: ENVIRONMENTAL FATE AND ECOTOXICOLOGICAL INVESTIGATIONS ..... 21
C.1. Environmental Fate ..... 21
C.1.1. Ready biodegradability ..... 21
C.2. Ecotoxicological Investigations ..... 21
C.2.1. Acute toxicity to fish ..... 21
C.2.2. Acute toxicity to aquatic invertebrates ..... 22
C.2.3. Algal growth inhibition test ..... 22
C.2.4. Inhibition of microbial activity ..... 23
BIBLIOGRAPHY ..... 24

# 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/1450 | Ecolab Pty Ltd | Peracid in KX-6228 | Yes | < 10 tonnes per annum | Sanitiser for the food and beverage industry and bleaching agent for the laundry care industry |

# 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 for the Classification and Labelling of Chemicals (GHS),* as adopted for industrial chemicals in Australia. The recommended hazard classification is presented in the table below.

| *Hazard classification* | *Hazard statement* |
|---|---|
| Flammable Liquids (Category 4) | H227 - Combustible liquid |
| Oxidising liquids (Category 1) | H271 - May cause fire or explosion; strong oxidizer |
| Acute toxicity (Category 4) | H302- Harmful if swallowed |
| Skin irritation/corrosion (Category 1) | H314 - Causes severe skin burns and eye damage |

In Australia, additional non-GHS hazard statements apply (see *Guidance on the Classification of Hazardous Chemicals Under the WHS Regulations* for further information; SWA, 2012a). Based on the available information, the following additional (non-GHS) hazard statement is also recommended, if applicable:

AUH071 – Corrosive to the respiratory tract

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 phrase:

R22: Harmful if swallowed
R35: Causes severe burns

The environmental hazard classification according to the *Globally Harmonised System for the 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 Toxicity (Category 1) | H400 - Very toxic to aquatic life |

**Human health risk assessment**
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 assessed 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:
  - H302 - Harmful if swallowed
  - H314 - Causes severe skin burns and eye damage
  - AUH071 - Corrosive to the respiratory tract (if applicable)

- The following should be used for products/mixtures containing the notified chemical:
  - Conc. > 25%: H302, H314
  - ≥ 5% Conc. < 25%: H314
  - ≥ 3% Conc. < 5%: H315, H318
  - ≥ 1% Conc. < 3%: H315, H319

  H315 - Causes skin irritation
  H318 - Causes serious eye damage
  H319 - Causes serious eye irritation

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:
  - Enclosed, automated processes, where possible

- 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 in the formulated products:
  - Avoid contact with skin and eyes
  - Avoid inhalation of vapours and aerosols
  - A shower and eyewash station should be available

- 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 in the formulated products:
  - Impervious gloves
  - Goggles or face-shield
  - Coveralls
  - Respiratory protection (if inhalation exposure to the notified chemical is expected)

  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 for the 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

- The notified chemical should be disposed of to landfill.

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, 2012b) 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

- Transport and packaging of the notified chemical must be in accordance with the NTC (National Transport Commission) 2007 Australian Code for the Transport of Dangerous Goods by Road and Rail (ADG code), 7th Edition, Commonwealth of Australia

**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 concentration of the notified chemical in the formulated sanitiser and laundry care product exceeds 1.5%;
- the notified chemical is intended to be manufactured as a substance in its own right.

or

(2) Under Section 64(2) of the Act; if
- the function or use of the chemical has changed from a sanitizer for the food and beverage industry and as a bleaching agent for the laundry care industry, or is likely to change significantly;
- the amount of chemical being introduced has increased from 10 tonnes per annum, or is likely to increase, significantly;
- the method of manufacture of the chemical in Australia has changed, or is likely to change, in a way that may result in an increased risk of an adverse effect of the chemical on occupational health and safety, public health, or the environment;
- 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 and products containing the notified chemical were provided by the notifier and 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)
Ecolab Pty Ltd (ABN:59 000 449 990)
2 Drake Avenue
MACQUARIE PARK NSW 2113

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: chemical name, other names, CAS number, molecular and structural formulae, molecular weight, analytical data, degree of purity, impurities, use details and manufacture volume.

VARIATION OF DATA REQUIREMENTS (SECTION 24 OF THE ACT)
Variation to the schedule of data requirements is claimed as follows: vapour pressure, flammability, autoignition temperature, oxidising properties, acute dermal toxicity, eye irritation, skin sensitisation and bioaccumulation.

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT(S)
None

NOTIFICATION IN OTHER COUNTRIES
ECHA (2011)

## 2. IDENTITY OF CHEMICAL

MARKETING NAME(S)
KX-6228 (sanitising product containing the notified chemical at ≤ 1.5% concentration)

MOLECULAR WEIGHT
< 500 Da

ANALYTICAL DATA
Reference NMR, IR, LC-MS and UV spectra were provided.

## 3. COMPOSITION

DEGREE OF PURITY > 65% (for isolated test substance used in studies)

## 4. PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE AT 20 °C AND 101.3 kPa: white pasty liquid

| Property | Value | Data Source/Justification |
|---|---|---|
| Melting Point/Freezing Point | -48.1to -31.7 °C | Measured |
| Boiling Point | 100.2 °C (with decomposition) at 101.3 kPa | Measured |
| Density | 1048.4 kg/m$^3$ at 20°C | Measured |
| Vapour Pressure | 4.57 x $10^{-9}$ kPa at 25°C | Calculated |
| Water Solubility | 43.8 g/L at 20 °C | Measured |
| Hydrolysis as a Function of pH | $t_{½}$ < 365 days at pH 4, 7 and 9<br>$t_{½}$ < 1 day at pH 7 and 8 | Measured<br>Calculated (HYDROWIN v2.00) |
| Partition Coefficient (n-octanol/water) | log Pow ≤ 5.04 at 20 °C | Measured |
| Adsorption/Desorption | log Koc < 5.28 at 20 °C | Measured |
| Dissociation Constant | pKa = 8.8 | Analogue data |
| Flash Point | 62.5°C at 101.3 kPa | Measured |
| Flammability | Not determined | Combustible liquid based on flash point |
| Autoignition Temperature | Not determined | Not expected to autoignite under |

| | | |
|---|---|---|
| | | normal conditions |
| Explosive Properties | Not explosive | Measured |
| Oxidising Properties | Expected to be oxidising | Expected to be oxidising due to oxidising functional groups |
| Surface tension | 32.9 mN/m at 20 °C | Measured |

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 and is stable in water and air at ambient temperatures. The notified chemical contains hydro-peroxide functional groups which are expected to be highly reactive when in contact with organic or combustible materials.

***Physical hazard classification***
Based on the submitted physico-chemical data depicted in the above table, the notified chemical is recommended for hazard classification according to the *Globally Harmonised System for the 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*** |
|---|---|
| Flammable Liquids (Category 4) | H227 - Combustible liquid |
| Oxidising liquids (Category 1) | H271 - May cause fire or explosion; strong oxidizer |

## 5. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will be manufactured in situ as a component of formulated products at ≤ 1.5% concentration in Australia. The notified chemical will not be manufactured as a substance in its own right.

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

| *Year* | *1* | *2* | *3* | *4* | *5* |
|---|---|---|---|---|---|
| *Tonnes* | < 10 | < 10 | < 10 | < 10 | < 10 |

PORT OF ENTRY
Not applicable (manufactured in Australia)

IDENTITY OF MANUFACTURER
Ecolab Cheltenham facility, 350 Reserve Road, Cheltenham, VIC 3192.

TRANSPORTATION AND PACKAGING
Following manufacture, the formulated sanitiser product KX-6228 containing the notified chemical at ≤ 1.5% concentration will be filled into 20 kg high-density polyethylene cubes, 200 kg high-density polyethylene drums, 1000 kg intermediate bulk containers (IBCs) and will be transported by road throughout Australia. Packaging details for the formulated laundry product containing the notified chemical at ≤ 1.5% concentration was not provided.

USE
The notified chemical will be used at ≤ 1.5% concentration, primarily as a sanitizer for the food and beverage industry; a minor secondary use is as a bleaching agent for the laundry care industry. The formulated laundry product will not be sold to the public and will be used solely for industrial laundry applications.

OPERATION DESCRIPTION

The notified chemical will be manufactured in situ in aqueous solution as a component of a formulated sanitiser and laundry care product in Australia.

Manufacture of products

The raw materials will be charged to a closed reaction vessel using mechanical pumps and dedicated lines. Following initiation of the chemical process that produces the notified chemical, the formulation will be pumped to a storage vessel and allowed to stand for seven days. A sample of the solution will be collected through dedicated valves for testing and quality control. The finished product containing the notified chemical at ≤ 1.5% concentration will then be packed off via a gravity fed mechanical operation.

End use

*As a sanitiser in the food and beverage industry:*

To sanitise food and beverage equipment, the finished product (KX-6228) containing the notified chemical at ≤ 1.5% concentration will predominantly (90% of use) be applied via a Clean in Place (CIP) system. A CIP system is a fully enclosed automatic process that delivers a number of wash and rinse cycles to sanitise tanks, pipe-work or other processing equipment. The finished product will be pumped to the CIP cycle directly from the packaging using a dedicated inlet line, where it will be further diluted to < 0.01% concentration notified chemical. After completion of the CIP sanitation cycle, the sanitiser will be collected to a waste tank and then pumped to a treatment plant. A rinse cycle will be performed and any residual sanitiser will be flushed to the waste tank.

The sanitiser may also be used (10% of use) in spray and immersion applications to clean external surfaces of equipment and machinery parts. In these cases the product (KX-6228) containing the notified chemical at ≤ 1.5% concentration will be diluted with water before use. The final use concentration of the notified chemical in the sanitising solution will be < 0.01%. After spray and immersion applications all exposed surfaces will be thoroughly rinsed with water and washings drained into the water treatment plant.

*As a bleaching agent for the laundry care industry:*

The laundry care product containing the notified chemical at ≤ 1.5% concentration will be delivered to the wash cycle automatically via a metered dose system. The system will be fully enclosed and programmed to deliver a specific volume of product at dedicated intervals throughout the wash program.

## 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)* |
|---|---|---|
| Manufacturing | 2 | 50 |
| Process operator/packing | 2 | 50 |
| Quality control | 1 | 25 |
| Warehouse | 4 | 30 |
| Transport | 4 | 10 |
| End user | 0.5 | 200 |

EXPOSURE DETAILS

Transport and storage

Transport and storage workers may come into contact with the notified chemical as a part of the finished product (≤ 1.5% concentration) only in the event of accidental rupture of containers.

Manufacture of products

Dermal, ocular and inhalation exposure to the notified chemical at ≤ 1.5% concentration may occur when performing quality control analysis and cleaning of manufacturing equipment. Exposure to the notified chemical at other times is not expected given manufacturing of the notified chemical will be conducted in enclosed reaction vessels and packaging of the finished products will be automated. All manufacturing workers are

expected to wear PPE including coveralls, goggles, full face respirator, gloves and safety boots to minimise any potential exposure.

End-use
*As a sanitiser in the food and beverage industry:*
Given the CIP system is a fully enclosed process and delivery of the sanitiser containing the notified chemical is automated, occupational exposure to the notified chemical from CIP end use is expected to be limited.

There is potential for dermal, ocular and inhalation exposure to the notified chemical when diluting the sanitiser product containing the notified chemical at ≤ 1.5% concentration and when applying the diluted sanitising solutions containing the notified chemical at < 0.01% during spray and immersion applications. The expected use of PPE by these workers should minimise exposure.

*As a bleaching agent for the laundry care industry:*
The laundry care product containing the notified chemical at ≤ 1.5% concentration will be delivered to the wash cycle via a metered dose system. As the system is fully enclosed and transfer of the product to the wash program is fully automated exposure to the notified chemical is expected to be negligible.

There is potential for dermal exposure to the notified chemical from wearing clothes washed in the laundry product containing the notified chemical. However, given the expected low concentration of the notified chemical in the wash cycle, which is further reduced during rinsing, significant quantities of the notified chemical are not expected to remain on the dried clothing articles.

### 6.1.2. Public Exposure
The finished product containing the notified chemical will only be used by professionals in an industrial setting and will not be sold to the public. Therefore the public will not be exposed to the notified chemical at concentrations up to 1.5% unless an accidental spillage occurred during transport.

It is possible that the public could be exposed indirectly to very low levels of the notified chemical through foodstuffs that have come into contact with surfaces sanitised with products containing the notified chemical. However, it is unlikely that there will be any significant oral/dietary exposure to the notified chemical as the concentration of the notified chemical in the sanitising solutions will be very low (< 0.01%) and the processing equipment is expected to be thoroughly rinsed before use.

The potential for indirect dermal exposure to members of the public from wearing clothes washed in laundry product containing the notified chemical is deemed negligible, given its low concentration (≤ 1.5%), which is further reduced during the rinsing cycle.

Therefore, members of the public are unlikely to be exposed to any significant quantities of the notified chemical from the proposed uses.

## 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 mg/kg bw |
| Skin irritation (*in vitro*) | corrosive |
| Rat, repeat dose oral toxicity – 28 days. | NOEL 50 mg/kg bw/day |
| Mutagenicity – bacterial reverse mutation | non mutagenic |
| Genotoxicity – *in vitro* mammalian cell gene mutation test | non genotoxic |
| Rat, reproductive and developmental toxicity | NOEL 50 mg/kg bw/day foetus;<br>NO(A)EL 50 mg/kg bw/day maternal |

*Toxicokinetic.*
The notified chemical is of low molecular weight (< 500 Da), surface active and corrosive hence dermal absorption may occur. Based on the low molecular weight there is also potential for absorption across the gastrointestinal and respiratory tract.

*Acute toxicity.*
The notified chemical was tested in an acute oral toxicity study in rats. For humane reasons, the test was limited to a top dose of 300 mg/kg bw as the notified chemical was expected to cause corrosion at higher concentrations. There were no mortalities in the study and the only clinical sign of toxicity was one animal displaying a hunched posture in the first 2 hours after administration. The LD50 was therefore determined in the study to be > 300 mg/kg bw. The results of the study are therefore inconclusive for classification purposes. However, the notifier has classified the notified chemical for precautionary reasons as R22 'Harmful by the oral route'.

For humane reasons, no acute dermal and inhalation toxicity studies have been performed for the notified chemical. The calculated vapour pressure ($4.57 \times 10^{-9}$ kPa at 25 °C) of the notified chemical is low and therefore inhalation of the vapour is not expected to occur under normal environmental conditions unless aerosols are formed.

*Irritation and sensitisation.*
The notified chemical was found to be corrosive to the skin in an *in vitro* skin irritation study in a human skin model (Human Skin Model EST-1000$^{TM}$). No skin sensitization data was submitted for the notified chemical. However the notified chemical does not contain a structural alert for sensitisation. Furthermore, close structural analogues of the notified chemical have been shown to be non-sensitising. Thus, the notified chemical is unlikely to be a skin sensitizer.

*Repeated Dose Toxicity.*
In a 28-day repeated dose oral toxicity study in rats dosed with the notified chemical at 5, 15 and 50 mg/kg bw/day by gavage, no treatment related effects were observed. The NOEL for the notified chemical was therefore determined to be 50 mg/kg bw/day.

*Mutagenicity/Genotoxicity.*
The notified chemical was found to be non-mutagenic in a bacterial reverse mutation assay and non-clastogenic in an *in vitro* mammalian cell mutation test.

*Toxicity for reproduction.*
In a developmental toxicity study the notified chemical was administered by oral gavage at doses of 5, 15 and 50 mg/kg bw/day from Day 6-19 of gestation. Decreased food consumption and weight loss was observed in the dams of the high dose treatment group; however, this effect was deemed non-adverse by the study authors as this change did not reach statistical significance during the course of the study. No treatment related effects were observed in the foetuses. The foetal No Observed Effect Level (NOEL) and maternal No Observed (Adverse) Effect Level (NO(A)EL) was therefore established as 50 mg/kg bw/day in this study.

***Health hazard classification***
Based on the available information, the notified chemical is recommended for hazard classification according to the *Globally Harmonised System for the 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 (Category 4) | H302- Harmful if swallowed |
| Skin Corrosion (Category 1) | H314- Causes severe skin burns and eye damage |

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 phrase(s):

R22: Harmful if swallowed
R35: Causes severe burns

### 6.3. Human Health Risk Characterisation

#### 6.3.1. Occupational Health and Safety
The notified chemical is harmful via the oral route, corrosive to the skin and expected to cause damage to the eyes. Given that workers will be exposed to the notified chemical at up to 1.5% concentration, irritation effects represent the greatest risk posed by the notified chemical.

Manufacture of products
Given the engineering controls in place during manufacture to limit exposure, workers most at risk of irritation effects from the notified chemical will be those involved in handling the notified chemical at up to 1.5% concentration during quality control analysis and cleaning of manufacturing equipment. Therefore, provided these workers wear PPE to limit exposure, the risk to the health of workers during manufacture of the formulated product containing the notified chemical is not considered to be unreasonable.

End-use
During end use workers will only be exposed to the notified chemical at up to 1.5% concentration when diluting the sanitiser product (KX-6228) to the desired end use concentration for spray and immersion applications; hence workers involved in this operation will be most at risk of irritation effects from the notified chemical. Therefore, provided these workers wear PPE to limit exposure, the risk to the health of workers under the proposed use for the notified chemical is not considered unreasonable.

### 6.3.2. Public Health
The public is not expected to be exposed to the notified chemical to any significant level; hence, the risk to public health is not considered unreasonable under the proposed use of the notified chemical.

## 7. ENVIRONMENTAL IMPLICATIONS

### 7.1. Environmental Exposure & Fate Assessment

### 7.1.1. Environmental Exposure

RELEASE OF CHEMICAL AT SITE
Manufacture of the notified chemical in situ as a component of the formulated product is expected to occur in Australia. During manufacture, raw materials are combined in a mixing vessel. Vapours generated during mixing are expected to be collected and displaced through a closed vent system where the vapours are captured to prevent release to the atmosphere.

Equipment is cleaned after batches of the notified chemical are manufactured. Batches are manufactured in succession to minimise the need for equipment cleaning and water use. Approximately 0.22% of the total import volume of notified chemical is expected to be disposed via wastewater as a result of cleaning processes. Wastewater is expected to undergo treatment before being released to sewer.

After manufacture, the notified chemical is packaged and stored before being dispatched to customer sites. Releases to the environment may occur following accidental spills during transport or storage. Notified chemical that is spilled is expected to be adsorbed onto a suitable material and collected for disposal in accordance with local regulations.

RELEASE OF CHEMICAL FROM USE
The notified chemical is expected to be used in highly controlled industrial environments. Accidental release of the notified chemical is not expected.

The majority of the notified chemical used as a sanitiser in the food and beverage industry is expected to be used in a fully enclosed, automated system. After completion of a sanitising cycle, the notified chemical is expected to be collected and undergo waste water treatment before being released to sewer. The sanitising cycle is expected to be conducted on 200 days per year nationwide. Approximately 11% of the total import volume of the notified chemical is expected to be released to sewer.

Some of the notified chemical may be used to sanitise external surfaces and small equipment using spray application and immersion baths. Surfaces are thoroughly rinsed after treatment with the notified chemical. The notified chemical in the immersion baths and the washings generated during rinsing are expected to be collected and undergo waste water treatment before being released to sewer.

RELEASE OF CHEMICAL FROM DISPOSAL
After use, the majority of the notified chemical is expected to undergo waste water treatment before being released to sewer. Up to 0.5% of the total import volume of the notified chemical is expected to be present as residues in empty product containers. These residues are expected to be disposed of to landfill.

### 7.1.2. Environmental Fate

The majority of the notified chemical is expected to be released to sewer following waste water treatment. Based on its water solubility, any notified chemical released to sewer is expected to remain in the water column and be released to surface waters. The notified chemical disposed of to landfill is expected to be mobile based on its high water solubility and adsorption/desorption coefficient (Koc) and may also migrate to surface waters. The notified chemical is not readily biodegradable. However, it is not expected to persist in landfill or the aquatic environment as it readily hydrolyses. Based on its potential to hydrolyse and high water solubility, the notified chemical is not expected to bioaccumulate. Ultimately, the notified chemical is expected to degrade via biotic and abiotic processes to form water and oxides of carbon. For the details of the environmental fate studies please refer to Appendix C.

### 7.1.3. Predicted Environmental Concentration (PEC)

The predicted environmental concentration (PEC) was calculated using a conservative scenario where the total annual import volume is release to sewer over 200 working days per year across the nation. This scenario also assumes no removal of the notified chemical during sewage treatment plant (STP) processes.

| ***Predicted Environmental Concentration (PEC) for the Aquatic Compartment*** | | |
|---|---|---|
| Total Annual Import/Manufactured Volume | 5,000 | kg/year |
| Proportion expected to be released to sewer | 100 % | |
| Annual quantity of chemical released to sewer | 5,000 | kg/year |
| Days per year where release occurs | 200 | days/year |
| Daily chemical release: | 25 | kg/day |
| Water use | 200 | L/person/day |
| Population of Australia (Millions) | 22.613 | million |
| Removal within STP | 0% | |
| Daily effluent production: | 4,523 | ML |
| Dilution Factor - River | 1 | |
| Dilution Factor - Ocean | 10 | |
| PEC - River: | 5.53 | µg/L |
| PEC - Ocean: | 0.55 | µg/L |

STP effluent re-use for irrigation occurs throughout Australia. The agricultural irrigation application rate is assumed to be 1000 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 1500 kg/m$^3$). Using these assumptions, irrigation with a concentration of 5.53 µg/L may potentially result in a soil concentration of approximately 36.9 µg/kg. Assuming accumulation of the notified chemical in soil for 5 and 10 years under repeated irrigation, the concentration of notified chemical in the applied soil in 5 and 10 years may be approximately 184 µg/kg and 369 µg/kg, respectively.

## 7.2. Environmental Effects Assessment

The results from ecotoxicological investigations 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* |
|---|---|---|
| ***Acute Toxicity*** | | |
| Fish Toxicity (96 h) | LC50 = 0.75 mg/L | Very toxic to fish |
| Daphnia Toxicity (48 h) | EC50 = 3.05 mg/L | Toxic to aquatic invertebrates |
| Algal Toxicity (72 h) | ErC50 = 5.44 mg/L | Toxic to algae |
| Inhibition of Bacterial Respiration (3 h) | EC50 = 216 mg/L | Not expected to be inhibitory to microbial activity. |
| ***Chronic Toxicity*** | | |
| Algal Toxicity (72 h) | NOEC = 1.66 mg/L | Not harmful |

Under the Globally Harmonised System of Classification of Chemicals (GHS; United Nations, 2009), the notified chemical is considered very toxic to fish, toxic to aquatic invertebrates and toxic to algae on an acute basis. Based on the acute toxicity endpoint for the most sensitive species, fish, the notified chemical is formally classified under the GHS as 'Acute Category 1; Very toxic to aquatic life'. The notified chemical is not classified under the GHS on an acute basis based on its rapid degradability via hydrolysis.

#### 7.2.1. Predicted No-Effect Concentration

The predicted no-effect concentration (PNEC) was calculated using the value for the most sensitive endpoint, the median lethal concentration (LC50) for fish. An assessment factor of 100 was used as study reports are available for three trophic levels.

| Predicted No-Effect Concentration (PNEC) for the Aquatic Compartment | | |
|---|---|---|
| LC50 (Fish, 96 h) | 0.75 | mg/L |
| Assessment Factor | 100 | |
| PNEC: | 7.5 | µg/L |

### 7.3. Environmental Risk Assessment

| Risk Assessment | PEC µg/L | PNEC µg/L | Q |
|---|---|---|---|
| Q - River | 5.53 | 7.5 | 0.737 |
| Q - Ocean | 0.553 | 7.5 | 0.0737 |

The Risk Quotients (Q = PEC/PNEC) for a conservative discharge scenario have been calculated to be < 1 for the river and ocean compartments. The notified chemical is not expected to be readily biodegradable however it is not expected to be persistent based on its potential to readily hydrolyse. The notified chemical is not expected to bioaccumulate based on its water solubility and potential to hydrolyse. Therefore, the notified chemical is not expected to pose an unreasonable risk to the environment based on its assessed use pattern.

## APPENDIX A: PHYSICAL AND CHEMICAL PROPERTIES

**Melting Point/Freezing Point** -48.1 to -31.7 °C

Method OECD TG 102 Melting Point/Melting Range.

Remarks Capillary tube method. The experiment was performed in duplicate and the results averaged.

Test Facility LAUS GmbH (2011a)

**Boiling Point** 100.2 °C (with decomposition) at 101.3 kPa

Method OECD TG 103 Boiling Point.

Remarks Siwoloboff method. The test substance showed signs of decomposition.

Test Facility LAUS GmbH (2011b)

**Density** 1048.4 kg/m$^3$ at 20°C

Method OECD TG 109 Density of Liquids and Solids.

Remarks Determined using the pycnometer method

Test Facility LAUS GmbH (2011c)

**Water Solubility** 43.8 g/L at 20 °C

Method OECD TG 105 Water Solubility.

Remarks Flask Method. The notified chemical contains surface active functionality and is expected to be water dispersible.

Test Facility TOXI-COOP ZRT (2011a)

**Hydrolysis as a Function of pH** $t_{½}$ < 365 days at pH 4, 7 and 9
$t_{½}$ < 1 day at pH 7 and 8 (HYDROWIN v2.00)

Method OECD TG 111 Hydrolysis as a Function of pH.

| *pH* | *T (°C)* | *$t_{½}$ <hours or days>* |
|---|---|---|
| 4 | 25 | < 365 days |
| 7 | 25 | < 365 days |
| 9 | 25 | < 365 days |

Remarks A preliminary test (Tier 1) was conducted on the test substance and the half-life was determined to be less than 365 days at 25 ºC. A Tier 2 test was not conducted. The recoveries of the test substance at 50 ºC after 1 day were 53.1%, 35.6% and 12.2% at pH 4, 7 and 9, respectively.

As a Tier 2 study was not conducted, further investigation into hydrolysis of the notified chemical indicated that it is likely to have a short half-life. As the study indicates, the half-life of the notified chemical decreases with increasing pH. HYDROWIN v2.00 estimates that the notified chemical is expected to have a hydrolysis half-life of less than 1 day at 25 ºC and pH 7 and 8 (US EPA 2011). Therefore, the notified chemical is expected to readily hydrolyse.

Test Facility Charles River (2012a)

**Partition Coefficient (n-octanol/water)** log Pow ≤ 5.04 at 20 °C

Method OECD TG 117 Partition Coefficient (n-octanol/water).

Remarks HPLC Method. The test substance is a UVCB substance without a defined n-octanol/water partition coefficient. The log Pow for peaks eluting from the HPLC with peak areas greater than 5% was measured. The log Pow for two peaks were determined to be < 1.6 and ≤ 5.04. Therefore, log Pow ≤ 5.04.

Test Facility TOXI-COOP ZRT (2011b)

**Adsorption/Desorption** log $K_{oc}$ = < 5.28 at 20 °C

Method OECD TG 121 Estimation of the Adsorption Coefficient (Koc) on Soil and on Sewage Sludge
Remarks HPLC Method. The test substance was unretained toup to log Koc = 5.28.
Test Facility Charles River (2011a)

**Flash Point** 62.5 °C at 101.3 kPa

Method EC Council Regulation No 440/2008 A.9 Flash Point.
Remarks Pensky-Martens closed cup method.
Test Facility LAUS GmbH (2011d)

**Explosive Properties**

Method EC Council Regulation No 440/2008 A.14 Explosive Properties.
Remarks No explosion was observed in the tests for thermal sensitivity and mechanical sensitivity (shock using an energy of 40 J)
Test Facility LAUS GmbH (2011e)

**Surface Tension** 32.9 mN/m at 20 °C

Method OECD TG 115 Surface Tension of Aqueous Solutions.
EC Council Regulation No 440/2008 A.5 Surface Tension.
Remarks Concentration: 756.38 mg/L
Test Facility LAUS GmbH (2011f)

# 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/ Sprague-Dawley Crl:CD(SD) |
| Vehicle | "Labrasol" (Caprylocaproylmacrogoglycerides EP/ Caprylocaproylpolyoxyglycerides) USP/NF in acetate buffer (0.1M acetic acid and 0.1M sodium acetate anhydrous |
| Remarks - Method | Formulation analysis with regard to concentration, homogeneity and stability was not performed in this study. Owing to the low concentration of the test substance in the vehicle, the test substance was administered as two dose fractions for the 300 mg/kg bw dose. Due to insufficient formulation, a new formulation of test substance was administered on the second dose to two animals in group II. This resulted in a 6 h separation between dosing, rather than 3 h as was observed for the other animals in the 300 mg/kg bw dose groups.<br><br>The test substance was only administered at up to 300 mg/kg bw for humane reasons since the test substance was believed to cause corrosion at higher concentrations. |

RESULTS

| *Group* | *Number and Sex of Animals* | *Dose mg/kg bw* | *Mortality* |
|---|---|---|---|
| I | 3F | 50 | 0/3 |
| II | 3F | 300# | 0/3 |
| III | 3F | 300# | 0/3 |

# Dose administered as 2 equal fractions

| | |
|---|---|
| LD50 | > 300 mg/kg bw |
| Signs of Toxicity | There were no mortalities during the study. One female in the 300 mg/kg bw group displayed hunched posture after approximately 1 h and 13/4 h after the second dose fraction. Body weight gains were in the normal range. |
| Effects in Organs | No macroscopic abnormalities were recorded |
| Remarks - Results | The oral LD50 value of the test substance was determined to be > 300 mg/kg bw. |
| CONCLUSION | The study is inconclusive for determining if the notified chemical is harmful via the oral route. |
| TEST FACILITY | Charles River (2011b) |

## B.2. Irritation – skin (in vitro)

| | |
|---|---|
| TEST SUBSTANCE | Notified chemical |
| METHOD | OECD TG 431 In vitro Skin Corrosion - Human Skin Model Test <Human Skin Model EST-1000™> |
| Vehicle | None |
| Remarks - Method | The positive control used was 8.0 M KOH and the negative control was deionised water. Cell viability was measured by MMT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide, thiazolyl blue) assay. |

RESULTS

| *Test material* | *Exposure Time* | *Mean $OD_{570}$ of two tissues* | *Relative mean Viability (%)* |
|---|---|---|---|
| Negative control | 3 min | 1.074 | 100.0 |
| Test substance | 3 min | 0.201 | 18.7 |
| Positive control | 3 min | 0.472 | 44.0 |
| Negative control | 1 h | 1.476 | 100.0 |
| Test substance | 1 h | 0.024 | 2.5 |
| Positive control | 1 h | 0.015 | 1.2 |

OD = optical density

Remarks - Results The test item was considered to be corrosive to the skin since the viability was less than 50% for the 3 minute exposure and less than 15% for 1 hour exposure.

CONCLUSION The notified chemical was corrosive to the skin under the conditions of the test.

TEST FACILITY Harlan (2011a)

**B.3. Repeat dose toxicity**

TEST SUBSTANCE Notified chemical

METHOD OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents.
Species/Strain Rat/Sprague Dawley (Crl:CD(SD))
Route of Administration Oral – gavage
Exposure Information Total exposure days: 28
Dose regimen: 7 days per week
Post-exposure observation period: 14 days
Vehicle Deionised water
Remarks - Method No significant protocol deviations

RESULTS

| *Group* | *Number and Sex of Animals* | *Dose mg/kg bw/day* | *Mortality* |
|---|---|---|---|
| control | 5M/5F | 0 | 0 |
| low dose | 5M/5F | 5 | 0 |
| mid dose | 5M/5F | 15 | 0 |
| high dose | 5M/5F | 50 | 0 |
| control recovery | 5M/5F | 0 | 0 |
| high dose recovery | 5M/5F | 50 | 0 |

*Mortality and Time to Death*
No unscheduled deaths occurred during the study.

*Clinical Observations*
No treatment related changes were observed by physical examination, sensory reactivity assessment or motor skills. There were no adverse bodyweight or food consumption.

*Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis*
No treatment related abnormalities were noted related to clinical chemistry or haematology. Urine volumes were lower in the mid- and high dose group than in controls with the differences statistically significant for females. This change was no longer observed at the end of the recovery period. The lower urine volumes were not considered toxicologically significant as there was no correlating change in kidney weight or histopathology findings in the kidney.

*Effects in Organs*

Dark foci in the lungs that were shown to correlate after microscopal examination with alveolar haemorrhage were observed at higher incidence in males of the high dose group than in controls. However the high dose recovery group showed no significant difference in the incidence of dark foci in the lung between treated and control animals and was therefore not considered by the study authors as biologically significant.

Speckling of the thymus that was correlated after microscopal examination to minimal multifocal haemorrhage was observed in the high dose recovery group. This observation was considered to be a common background finding by the study authors.

No treatment related organ weight changes were noted during the study.

Remarks – Results
The results of the study showed no treatment related effects at the highest dose tested.

CONCLUSION
The No Observed Effect Level (NOEL) was established as 50 mg/kg bw/day in this study, based no treatment related findings at the highest dose tested.

TEST FACILITY Charles River (2012b)

**B.4. Genotoxicity – bacteria**

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.
Plate incorporation procedure (Test 1) and Pre incubation procedure (Test 2)

| | |
|---|---|
| Species/Strain | *S. typhimurium*: TA1535, TA1537, TA98, TA100<br>*E. coli*: WP2uvrA |
| Metabolic Activation System | S9fraction from phenobarbital/ß-naphthaflavone induced rat liver |
| ConcentrationRange in Main Test | a) With metabolic activation: 3-5000 µg/plate<br>b) Without metabolic activation: 3-5000 µg/plate |
| Vehicle | DMSO |
| Remarks - Method | No significant protocol deviations.<br><br>The negative control was DMSO and positive controls were sodium azide, 4-nitro-o-phenylene-diamine, methyl methane sulfonate without metabolic activation, and 2-aminoanthracene in the presence of metabolic activation. |

RESULTS

| *Metabolic Activation* | *Test Substance Concentration (µg/plate) Resulting in:* | | | |
|---|---|---|---|---|
| | *Cytotoxicity in Preliminary Test* | *Cytotoxicity in Main Test* | *Precipitation* | *Genotoxic Effect* |
| *Absent* | | | | |
| Test 1 | ≥ 2500 | ≥ 2500 | > 5000 | Negative |
| Test 2 | | ≥ 1000 | > 5000 | Negative |
| *Present* | | | | |
| Test 1 | > 5000 | > 5000 | > 5000 | Negative |
| Test 2 | | ≥ 5000 | > 5000 | Negative |

Remarks - Results The test substance was tested up to the maximum dose level of 5000 µg/plate. No significant increases in the frequency of revertant colonies were recorded for any of the bacterial strains, with any dose of the test substance, either with or without metabolic activation.

All the positive control chemicals used in the test induced marked

increases in the frequency of revertant colonies, thus, validating the test system.

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

TEST FACILITY Harlan (2011b)

**B.5. Genotoxicity – in vitro**

TEST SUBSTANCE Notified chemical

METHOD OECD TG 476 In vitro Mammalian Cell Gene Mutation Test.
EC Directive 2000/32/EC B.17 Mutagenicity - In vitro Mammalian Cell Gene Mutation Test.

| | |
|---|---|
| Species/Strain | Chinese hamster |
| Cell Type/Cell Line | V79 cell line |
| Metabolic Activation System | S9fraction from phenobarbital/ß-naphthaflavone induced rat liver |
| Vehicle | DMSO |
| Remarks - Method | No significant protocol deviations |

The negative control was DMSO. Positive controls used in the study were ethylmethane sulfonate (without metabolic activation) and 7,12-dimethylbenz(a)anthracene (with metabolic activation).

| *Metabolic Activation* | *Test Substance Concentration (µg/mL)* | *Exposure Period* | *Expression Time* | *Selection Time* |
|---|---|---|---|---|
| *Absent* | | | | |
| Test 1 | 0.63, 1.3, 2.5*, 5.0*, 10.0*, 15.0*, 20.0* | 4h | 7d | 8d |
| Test 2 | 10.0*, 20.0*, 40.0*, 80.0*, 160.0, 240.0, 320.0 | 24h | 7d | 8d |
| *Present* | | | | |
| Test 1 | 20.0, 40.0*, 80.0*, 160.0*, 320.0*, 460.0*, 600.0 | 4h | 7d | 8d |
| Test 2 | 40.0, 80.0, 160.0*, 320.0*, 360.0*, 400.0*, 440.0* | 4h | 7d | 8d |

*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 | ≥ 20 | ≥ 20 | Negative |
| Test 2 | ≥ 312.5 | ≥ 40 | ≥ 320 | Negative |
| *Present* | | | | |
| Test 1 | ≥ 625 | ≥ 460 | ≥ 600 | Negative |
| Test 2 | | ≥ 400 | ≥ 440 | Negative |

Remarks - Results A single increased value of the mutation frequency relative to the solvent control was noted at 80 µg/mL in the first culture of the second experiment without metabolic activation. However, this increase was judged as being biologically irrelevant by the study authors since the threshold (three times the mutation frequency) was not exceeded and there was no dose dependant trend. No other toxicologically significant increases in the frequency of mutant colonies were recorded, with any dose of the test substance, either with or without metabolic activation.

All the positive control chemicals used in the test induced marked increases in the frequency of mutant colonies thus confirming the activity of the S9-mix and the sensitivity of the test.

| | |
|---|---|
| CONCLUSION | The notified chemical was not clastogenic to Chinese hamster cells treated in vitro under the conditions of the test. |
| TEST FACILITY | Harlan (2011c) |

**B.6. Developmental toxicity**

| | |
|---|---|
| TEST SUBSTANCE | Notified chemical |
| METHOD | OECD TG 414 Prenatal Developmental Toxicity Study |
| Species/Strain | Rat/Sprague Dawley (Crl:CD(SD)) |
| Route of Administration | Oral – gavage |
| Exposure Information | Exposure days: once a day for Days 6-19 of gestation<br>Post-exposure observation period: Animals were sacrificed on Day 20 |
| Vehicle | Deionised water |
| Remarks - Method | No significant protocol deviations |

RESULTS

| *Group* | *Number of Animals* | *Dose mg/kg bw/day* | *Mortality* |
|---|---|---|---|
| I (control) | 24 | 0 | 0/24 |
| II (low dose) | 24 | 5 | 0/24 |
| II (low dose) | 24 | 15 | 1/24 |
| III (high dose) | 24 | 50 | 0/24 |

*Mortality and Time to Death*

One animal in the 15 mg/kg bw/day group was found dead on day 18, however, this death was considered to be due to a gavage injury and not test substance related. No unscheduled deaths occurred during this study.

*Effects on Dams*

At 50 mg/kg bw/day one animal showed signs of piloerection (days 19-20) as well as subcutaneous masses on the ventral thorax which were correlated to enlarged lymph nodes at necropsy. A second animal in the same treatment group showed signs of hunched posture, piloerection and weight loss on days 15-20. Necropsy revealed pale, discoloured kidneys and pelvic dilation.

Decreased food consumption and mean body weight was noted in the 50 mg/kg bw/day treatment group; however, this change did not reach statistical significance during the course of the study.

*Effects on Foetus*

Cervical remnants of thymus were noted in more litters at 50 mg/kg bw/day than in other groups but in the same number of foetuses as for animals dosed at 5 mg/kg bw/day. The study authors concluded that since this effect was not dose-dependent it was not significant.

Local thinning of the tendinous region of the diaphragm was noted with higher incidence in the high dose treatment group. This finding was slightly outside of the background control range seen in other Charles river studies, however, this finding was deemed to be of no toxicological significance in the absence of other findings by the study authors.

Remarks - Results

Slight reductions in maternal body weight and food consumption were observed in the dams of the high dose treatment group but this change was deemed non-adverse by the study authors.

CONCLUSION

The foetal No Observed Effect Level (NOEL) and maternal No Observed (Adverse) Effect Level (NO(A)EL) were established as 50 mg/kg bw/day in this study, based on no test substance related effects at this level.

| | |
|---|---|
| TEST FACILITY | Charles River (2012c) |

# 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 |
| Exposure Period | 28 days |
| Auxiliary Solvent | None |
| Analytical Monitoring | Total organic carbon (TOC) |
| Remarks - Method | The test was conducted according to test guidelines using good laboratory practice (GLP) with no significant deviations. |

RESULTS

| *Test substance* | | *Aniline* | |
|---|---|---|---|
| *Day* | *% Degradation* | *Day* | *% Degradation* |
| 7 | 7.9 | 7 | 51.2 |
| 14 | 33.7 | 14 | 74.9 |
| 29 | 55.9 | 29 | 72.8 |

| | |
|---|---|
| Remarks - Results | All relevant test validity criteria were met. A toxicity control indicated the test substance is not toxic towards the inoculum at the concentration tested. |
| CONCLUSION | The notified chemical is not readily biodegradable but has the potential to degrade. |
| TEST FACILITY | LAUS GmbH (2011g) |

## C.2. Ecotoxicological Investigations

### C.2.1. Acute toxicity to fish

| | |
|---|---|
| TEST SUBSTANCE | Notified chemical |
| METHOD | OECD TG 203 Fish, Acute Toxicity Test – Flow-through. |
| Species | *Oncorhynchus mykiss* (Rainbow Trout) |
| Exposure Period | 96 hours |
| Auxiliary Solvent | None |
| Water Hardness | Not reported |
| Analytical Monitoring | Not reported |
| Remarks – Method | The test was conducted according to test guidelines using good laboratory practice (GLP) with no significant deviations. |

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 |
| 0.0625 | 0.0629 | 7 | 0 | 0 | 0 | 0 | 0 |
| 0.125 | 0.126 | 7 | 0 | 0 | 0 | 0 | 0 |
| 0.25 | 0.252 | 7 | 0 | 0 | 0 | 0 | 0 |
| 0.5 | 0.505 | 7 | 0 | 0 | 0 | 0 | 0 |
| 1 | 1.01 | 7 | 0 | 3 | 7 | 7 | 7 |

| | |
|---|---|
| LC50 | 0.75 mg/L at 96 hours. |
| NOEC | 0.25 mg/L at 96 hours. |
| Remarks – Results | All relevant test validity criteria were met. |

CONCLUSION The notified chemical is very toxic to fish

TEST FACILITY Charles River (2012d)

**C.2.2. Acute toxicity to aquatic invertebrates**

TEST SUBSTANCE Notified chemical

METHOD OECD TG 202 *Daphnia* sp. Acute Immobilisation Test -

| | |
|---|---|
| Species | *Daphnia magna* |
| Exposure Period | 48 hours |
| Auxiliary Solvent | None |
| Water Hardness | 249 mg $CaCO_3$/L |
| Analytical Monitoring | Iodometric titration |
| Remarks - Method | The test was conducted according to test guidelines using good laboratory practice (GLP) with no significant deviations. |

RESULTS

| *Concentration mg/L Actual* | *Number of D. magna* | *Number Immobilised* | |
|---|---|---|---|
| | | *24 h* | *48 h* |
| Control | 4 × 5 | 0 | 0 |
| 1.28 | 4 × 5 | 0 | 0 |
| 2.69 | 4 × 5 | 0 | 7 |
| 6.89 | 4 × 5 | 18 | 20 |
| 15.61 | 4 × 5 | 20 | 20 |
| 32.91 | 4 × 5 | 20 | 20 |

| | |
|---|---|
| EC50 | 3.05 mg/L at 48 hours |
| NOEC | 1.28 mg/L at 48 hours |
| Remarks - Results | All relevant test validity criteria were met. |

CONCLUSION The notified chemical is toxic to aquatic invertebrates.

TEST FACILITY TOXI-COOP ZRT (2011c)

**C.2.3. Algal growth inhibition test**

TEST SUBSTANCE Notified chemical

METHOD OECD TG 201 Alga, Growth Inhibition Test.

| | |
|---|---|
| Species | *Pseudokirchneriella subcapitata* |
| Exposure Period | 72 hours |
| Concentration Range | Nominal: 0, 0.4, 1.0, 2.6, 6.4, 16.0 and 40.0% v/v<br>Actual: < LOD, < LOD, < LOD, 0.56, 1.66, 3.60 and 8.25 mg/L |
| Auxiliary Solvent | None |
| Water Hardness | Not reported |
| Analytical Monitoring | Iodometric Titration |
| Remarks - Method | The test was conducted according to test guidelines using good laboratory practice (GLP) with no significant deviations. The lowest two concentrations only remained in the measureable range at the start of the test. |

RESULTS

| *Biomass* | *Growth* |
|---|---|

| *EyC50* *mg/L at 72 h* | *NOEyC* *mg/L* | *ErC50* *mg/L at 72 h* | *NOErC* *mg/L* |
|---|---|---|---|
| 2.56 | 1.66 | 5.44 | 1.66 |

Remarks - Results All relevant test validity criteria were met.

CONCLUSION The notified chemical is toxic to algae.

TEST FACILITY TOXI-COOP ZRT (2011d)

**C.2.4. Inhibition of microbial activity**

TEST SUBSTANCE Notified chemical

METHOD OECD TG 209 Activated Sludge, Respiration Inhibition Test.
Inoculum Activated sludge
Exposure Period 3 hours
Concentration Range Nominal: 6, 20, 60, 200 and 600 mg/L
Remarks – Method The test was conducted according to test guidelines using good laboratory practice (GLP) with no significant deviations.

RESULTS
EC50 216 mg/L
NOEC 20 mg/L
Remarks – Results All relevant test validity criteria were met.

CONCLUSION The notified chemical is not expected to be inhibitory to microbial activity.

TEST FACILITY Brixham Environmental Laboratory (2012)

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