File No: STD/1394

July 2011

NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

FULL PUBLIC REPORT

UU

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 Full Public Report may be inspected at our NICNAS office by appointment only at Level 7, 260 Elizabeth Street, Surry Hills NSW 2010.

This Full 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:

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

Director
NICNAS

TABLE OF CONTENTS

FULL PUBLIC REPORT

UU

1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT(S)
Mark Sensing Australia Pty. Ltd. (ABN 27 005 481 961)
31 Jersey Road
BAYSWATER VIC 3153

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, Non-hazardous impurities, Use details, Import volume, and Identity of recipients.

VARIATION OF DATA REQUIREMENTS (SECTION 24 OF THE ACT)
Variation to the schedule of data requirements is claimed as follows: Hydrolysis as a function of pH, Dissociation constant, Induction of Germ Cell Damage, Acute inhalation and Bioaccumulation

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT(S)
None

NOTIFICATION IN OTHER COUNTRIES
EU (2004)

2. IDENTITY OF CHEMICAL

MARKETING NAME(S)
UU (product containing the notified chemical at >70%)

MOLECULAR WEIGHT
>500 Da

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

3. COMPOSITION

DEGREE OF PURITY >70%

HAZARDOUS IMPURITIES/RESIDUAL MONOMERS None

NON HAZARDOUS IMPURITIES/RESIDUAL MONOMERS (> 1% by weight)
Several unidentified impurities (over 20) are traced in a HPLC. Most are below 1%, total between 3 and 35%. The typical concentration of unknown impurities is 16.2% (w/w) with a lower limit of 3% and an upper limit of 35%.

ADDITIVES/ADJUVANTS None

4. PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE AT 20°C AND 101.3 kPa: White powder

Property	Value	Data Source/Justification
Melting Point	Between 160 and 200°C	Measured
Boiling Point	Not determined	Expected to decompose at temperatures above 220°C
Density	1250 Kg/m^3 at 20°C	Measured
Vapour Pressure	6 x 10^{-10} kPa at 25°C	Measured
Water Solubility	≤ 3 × 10^{-5} g/L at 20°C	Measured
Hydrolysis as a Function of pH	Not determined	Hydrolysis is expected to be very slow under environmental conditions due to the limited water solubility of the notified chemical
Partition Coefficient (n-octanol/water)	log Kow = 2.0 at 25°C	Measured
Adsorption/Desorption	log K_{oc} = 4.5 at 25°C	Measured
Dissociation Constant	Not determined	The notified chemical is not expected to dissociate under environmental conditions (pH 4 – 9)
Particle Size	Inhalable fraction (<105 μm): 51.5% Respirable fraction (<10.4 μm):5.9%	Measured
Flash Point	Not determined	The notified chemical is a solid
Flammability (Solid)	Not highly flammable	Measured
Relative Autoignition Temperature for Solids	>400°C	Measured
Oxidising properties	Non-oxidising	Measured
Explosive Properties	Not explosive	Measured

DISCUSSION OF PROPERTIES

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

Reactivity

Under normal conditions, the notified chemical does not react with water or air. In addition, the notified chemical does not possess oxidizing properties nor it is explosive when exposed to shock/friction or to intense heat.

Dangerous Goods classification

Based on the submitted physical-chemical data in the above table, the notified chemical is not classified according to the Australian Dangerous Goods Code (NTC, 2007). However, the data above do not address all Dangerous Goods endpoints. Therefore, consideration of all endpoints should be undertaken before a final decision on the Dangerous Goods classification is made by the introducer of the chemical.

5. INTRODUCTION AND USE INFORMATION

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

The notified chemical will not be manufactured in Australia & will be imported in a powder form at >70% concentration.

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
Melbourne

TRANSPORTATION AND PACKAGING
The notified chemical will be imported in multilayer 20kg bags within a card board box wrapped on to pallets and transported by trucks from the port and for any further delivery.

USE
Colour developer for thermal paper

OPERATION DESCRIPTION
The notified chemical will not be manufactured in Australia. It will be imported in a powder form at >70% concentration.

The notified chemical will be reformulated with water into a 'premix formulation' containing approximately <25% of the notified chemical by manually charging the neat notified chemical into a mixer containing water and a stirrer. The mixer is half (lid) covered with dust extraction fitted onto it. Local Exhaust Ventilation (LEV) is also used during the addition of powdered notified chemical and other chemicals during 'premix formulation'. The premix water dispersion formulation gets stirred inside this vessel. The solution is later transferred into a closed mill.

During the next step, the premix formulation, blended with other ingredients and water in an enclosed system, is converted into a coating formulation to be used for coating and printing. The final concentration of the notified chemical in the coating/printing product is <10%.

The coating/printing mill is a closed unit, the coater is semi-enclosed (roto gravure process) multi station printing press, with a closed circuit where the liquid is continuously being pumped and coated.

Maintenance and service are mainly linked to mechanical /electrical repairs other than cleaning processes, which result in running out or dumping formulation (via chemical recycling firms) and cleaning the print cylinders.

6. HUMAN HEALTH IMPLICATIONS

6.1. Exposure Assessment

6.1.1. Occupational Exposure

NUMBER AND CATEGORY OF WORKERS

Category of Worker	*Number*	*Exposure Duration (hours/day)*	*Exposure Frequency (days/year)*
Formulator (in powder form), (in dispersion)	3	(10 min) (8 hour shift)	12
Printer	6	8	12
Maintenance & servicing personnel	-	-	-

EXPOSURE DETAILS
Transport and storage
During transportation, warehousing, and distribution of products containing the notified chemical, exposure to the notified chemical is not expected, except in the unlikely event of an accident where the packaging is damaged.

Reformulation into coatings/printing formulations
There is potential for dermal, ocular and inhalation exposure to the notified chemical during various reformulation steps leading to the final coating/printing formulation. Although the neat notified chemical is manually charged into a mixer containing water and a stirrer, the use of dust extraction fitted into the lid of the mixer and also the use of LEV will minimise any potential exposure to the notified chemical. Furthermore, the use of personal protective equipment (PPE) such as overalls, chemical resistant gloves, closed goggles, half or full facemasks with various filters, will also limit the potential of any exposure to the notified chemical.

The premix water dispersion formulation containing the notified chemical is transferred into a closed mill for further blending with other ingredients. Although there is potential for dermal, ocular and inhalation exposure to the notified chemical when the premix water dispersion formulation containing the notified chemical is blended with other intermediate formulations and water, exposure is expected to be limited due to the use of an enclosed system and the use of PPE as stated above. There may be some exposure as a result of drips and spills during the connection and disconnection of transfer pipes, but exposure will be minimised by the use of PPE as stated above.

Coating and printing
There is also a potential for dermal, ocular and inhalation exposure to the notified chemical during coating and printing processes. However, exposure is expected to be limited as the coating/printing mill is a closed circuit where the liquid is continuously pumped and coated. Although the coater is semi-enclosed (roto gravure process) multi station printing press, the use of PPE such as overalls, safety shoes, and occasionally gloves/protective glasses, will limit any exposure to the notified chemical.

Maintenance and Servicing
Dermal, ocular and inhalation exposure to the notified chemical may occur during maintenance and servicing of various equipments used in the reformulation of the notified chemical into coatings/printing formulations and coating/printing equipments. Workers will wear PPE as required to minimise any possible exposure.

Overall, as stated above and based on the use of engineering controls and PPE by workers, exposure during various procedures/processes involving the use of notified chemical is expected to be low.

6.1.2. Public Exposure
The notified chemical and the product containing the notified chemical will only be available to industrial users and will not be sold to the general public. Therefore, the general public will not be exposed to the notified chemical as such. However, the general public may be exposed to the notified chemical through contact with the coated/printed thermal paper. Once coated/printed on the thermal paper, the notified chemical becomes an inert form of the thermal paper and is not bioavailable for exposure. Therefore, exposure to the general public from the use of the notified chemical is considered to be low.

6.2. Human Health Effects Assessment

The results from toxicological investigations conducted on the notified chemical are summarised in the table below. Details of these studies can be found in Appendix B.

Endpoint	*Result and Assessment Conclusion*
Rat, acute oral toxicity	LD50 >2000 mg/kg bw; low toxicity
Rat, acute dermal toxicity	LD50 > 3161 mg/kg bw, low toxicity
Rabbit, skin irritation	non-irritating
Rabbit, eye irritation	non-irritating
Guinea pig, skin sensitisation – adjuvant test	no evidence of sensitisation
Rat, repeat dose oral toxicity – 28 days.	NOEL 1000 mg/kg/day
Mutagenicity – bacterial reverse mutation	non mutagenic
Genotoxicity–in vitro-Chinees Hamster CHL/IU Cells	non genotoxic

Toxicokinetics, metabolism and distribution.
No data were available to assess toxicokinetics, metabolism and distribution of the notified chemical. Although dermal absorption may occur due to low log Kow and molecular weight (>500), low water solubility may be a limiting factor in dermal absorption. This is consistent with the lack of systemic effects in the acute dermal toxicity study.

Acute toxicity.
The oral LD50 was >2000 mg/kg bw in rats and dermal LD50 was >3161 mg/kg bw in rats. Therefore, the notified chemical was not considered to be harmful via the oral and dermal route. Although information was not submitted on inhalation toxicity, due to low vapour pressure, inhalation toxicity is expected to be low.

Irritation and Sensitisation.
The notified chemical was not irritating to the skin and eyes of rabbit, and was also not a skin sensitiser in guinea

pigs.

Repeated Dose Toxicity (sub acute, sub chronic, chronic).
In an oral toxicity study in rats, the notified chemical was administered orally by gavage once daily (7 days/week) for 28 days at 0, 40, 200 or 1000 mg/kg bw/day. A recovery group of the high dose group was also observed for 14 days after the termination of the main study.

Significant abnormality at necropsy and during histological examination was not detected in any treatment groups at the termination of dosing and recovery periods. The only noted effect was increased absolute and relative adrenal gland weights in males only at 40 mg/kg bw/day. However, as no such effect was noted in females of the 40 mg/kg bw/day group and in animals from the 200 and 1000 mg/kg bw/day groups, this effect was unlikely to be test substance related and is therefore not considered to be an adverse effect. In addition, histopathological examination did not reveal any abnormality in adrenal glands of the 40 mg/kg bw/day animals group.

No significant finding was noted in urinalysis at the termination of treatment and recovery periods. Minor alterations were observed in some clinical chemistry and haematology parameters. However, these alterations were considered to be incidental, not toxicologically significant, and were unlikely to be test substance related.

The No Observed Effect Level (NOEL) was established as 1,000 mg/kg bw/day in this study, based on lack of effect at the highest tested dose (1,000 mg/kg bw/day).

Mutagenicity.
The notified chemical was found to be negative in a bacterial reverse mutation test, and also showed no evidence of clastogenicity in a Mammalian Chromosome Aberration Test, using Chinese Hamster CHL/IU Cells. Based on these results, the notified chemical is not expected to be genotoxic.

Carcinogenicity.:
No data were available to assess the potential for carcinogenicity.

Toxicity for reproduction.
No data were available to assess the potential for toxicity for reproduction.

Health hazard classification
Based on the submitted data, the notified chemical is not classified as hazardous, according to the *Approved Criteria for Classifying Hazardous Substances* (NOHSC, 2004).

6.3. Human Health Risk Characterisation

6.3.1. Occupational Health and Safety
The notified chemical was of low acute oral and dermal toxicity in rats and was neither irritating to the skin and eyes of rabbit nor was a skin sensitiser in guinea pigs. In a 28 day subacute oral toxicity study in rats, the NOEL was established as 1,000 mg/kg bw/day, based on lack of effect at the highest tested dose. The notified chemical is unlikely to be genotoxic. Therefore, the notified chemical is considered to be a non-hazardous chemical.

However, as around 5.9% particles of notified chemical are in the respirable range (<10.4 μm), the primary concern for workers handling the notified chemical is the potential of adverse respiratory effects if airborne dusts are inhaled. This potential is not considered to be significant, due to the enclosed nature of the reformulation systems, which are fitted with dust extraction systems and LEV, as well as the use of respiratory protection by workers.

Given the proposed use of PPE and the engineering controls in place and the non-hazardous nature of the notified chemical, the risk to workers using the notified chemical is not considered to be unreasonable.

6.3.2. Public Health

The notified chemical and the product containing the notified chemical will only be available to industrial users and will not be sold to the general public. No exposure is expected from the coated/printed thermal paper as the notified chemical will not be in bioavailable form. Therefore, as exposure to the general public is not expected, the notified chemical does not pose an unreasonable risk to the public.

7. ENVIRONMENTAL IMPLICATIONS

7.1. Environmental Exposure & Fate Assessment

7.1.1. Environmental Exposure

RELEASE OF CHEMICAL AT SITE

The notified chemical will be imported as a powder for reformulation into coating/printing formulations. The potential for release of the notified chemical during reformulation is anticipated to be very low as the process will be conducted in a closed system. Residues of the notified chemical in import containers are expected to be disposed of to landfill. Spills of the notified chemical are expected to be collected and disposed of to landfill. Reformulation equipment wash water and excess coating containing the notified chemical are expected to be disposed of by licensed waste disposal companies.

RELEASE OF CHEMICAL FROM USE

The notified chemical will be applied to thermal paper. No significant release of the notified chemical is anticipated as it will be used in closed system in industrial settings. After introduction of the notified chemical into thermal paper, the notified chemical cures and becomes an inert part of the paper.

RELEASE OF CHEMICAL FROM DISPOSAL

Thermal paper containing the notified chemical is not likely to enter the paper recycling process stream at significant levels, and is mainly expected to be disposed of to landfill. Hence, the majority of the imported quantity of notified chemical will eventually be disposed of to landfill.

7.1.2. Environmental Fate

The majority of notified chemical will be applied to paper and cured, and is therefore not expected to be bioavailable. The majority of paper containing the notified chemical is expected to be disposed of to landfill where the notified chemical will eventually degrade by biotic and abiotic processes to form water and oxides of carbon, nitrogen and sulfur.

A minor amount of the paper containing the notified chemical may be recycled. During recycling processes, waste paper is repulped using a variety of chemical agents which, amongst other things, enhance detachment of ink from the fibres. Very little of the notified chemical is expected to partition to the supernatant water which is released to the sewer. Moreover, based on its high log K_{oc} the notified chemical released to sewer during the recycling process is anticipated to sorb to sludge and sediment where it is also expected to degrade biotically and abiotically. Sludge from water treatment plants is expected to be sent to landfill or used for soil remediation.

The notified chemical was found to be not readily biodegradable based on a study submitted by the notifier. However, the notified chemical is not anticipated to bioaccumulate due to its low partition coefficient.

For the details of the environmental fate study, refer to Appendix A.

7.1.3. Predicted Environmental Concentration (PEC)

Worst-case aquatic PECs (ocean and river) have been calculated assuming that 50% of notified chemical will reach the aquatic compartment due to releases from thermal paper recycling. This is a conservative upper limit as thermal paper is not expected to enter the paper recycling stream to a significant extent. It was also assumed there would be no removal of the notified chemical by sewerage treatment plants (STPs) and release of the notified chemical will occur over 260 days per annum into the total Australian effluent volume. This corresponds to release from recycling processes only on working days, based on a 5 day work week.

Predicted Environmental Concentration (PEC) for the Aquatic Compartment		
Total Annual Import/Manufactured Volume	10,000	kg/year
Proportion expected to be released to sewer	50%	
Annual quantity of chemical released to sewer	5,000	kg/year
Days per year where release occurs	260	days/year
Daily chemical release:	19.23	kg/day
Water use	200.0	L/person/day
Population of Australia (Millions)	21.161	million
Removal within STP	0%	
Daily effluent production:	4,232	ML
Dilution Factor - River	1.0	
Dilution Factor - Ocean	10.0	
PEC - River:	4.54	µg/L
PEC - Ocean:	0.45	µg/L

Based on its high log Koc, the notified chemical is anticipated to sorb to sludge and sediment in STPs. However based on the worst case assumption that none of the notified chemical is removed from waste water in STPs, the estimated quantity of notified chemical anticipated to be released to agricultural land via STP effluent re-use is presented below.

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 4.544 µg/L may potentially result in a soil concentration of approximately 30.29 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 152 µg/kg and 303 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*
Fish Toxicity (96 h)	LL50 > 100 mg/L (WAF)	Not harmful up to the limit of its solubility in water
Daphnia Toxicity (48 h)	EL50 > 100mg/L (WAF)	Not harmful up to the limit of its solubility in water
Daphnia Toxicity (21 d)	NOEL = 0.05 mg/L	Not harmful up to the limit of its solubility in water
Algal Toxicity (72 h)	E_rL50 > 100 mg/L (WAF)	Not harmful up to the limit of its solubility in water
Algal Toxicity (72 h)	NOEL = 100 mg/L (WAF)	Not harmful up to the limit of its solubility in water
Inhibition of Bacterial Respiration (3 h)	IL50 > 100 mg/L	Does not inhibit respiration of waste water microorganisms

Under the Globally Harmonised System of Classification and Labelling of Chemicals (United Nations, 2009) the notified chemical is classified as not harmful to fish, aquatic invertebrates and algae up to the limit of its solubility in water. The reported endpoints are based on nominal loading rates of the water accommodated fraction (WAF) used for testing, consistent with international best practice (OECD, 2000), as the notified chemical is part of a multi-component substance with low aqueous solubility.

7.2.1. Predicted No-Effect Concentration

A PNEC was not calculated since the results from ecotoxicological investigations indicate that the notified chemical is not harmful to aquatic organisms up to its limit of solubility in water.

7.3. Environmental Risk Assessment

It was not considered meaningful to calculate a PNEC, nor a risk quotient Q (= PEC/PNEC), since no effects to aquatic organisms were reported up to the limit of solubility of the notified chemical in the submitted ecotoxicity studies. The exposure of the chemical to aquatic compartment is expected to be very low as the majority of the thermal paper containing the notified chemical is expected to be disposed of to landfill. The amount of thermal paper entering the paper recycling stream is not expected to be significant and, moreover, the majority of notified chemical released from recycling processes is expected to sorb to sludge and sediment in STPs resulting in a limited potential for release to surface waters. Based on its low log Kow the notified chemical is not expected to bioaccumulate. The notified chemical is therefore not considered to pose an unreasonable risk to the aquatic environment from its assessed use pattern.

8. CONCLUSIONS AND REGULATORY OBLIGATIONS

Hazard classification

Based on the submitted data, the notified chemical is not classified as hazardous according to the *Approved Criteria for Classifying Hazardous Substances* [NOHSC:1008(2004)].

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

CONTROL MEASURES

Occupational Health and Safety

- Employers should implement the following engineering controls to minimise occupational exposure to aerosols (particles) of the notified chemical during reformulation:
  - Local Exhaust Ventilation
  - Enclosed and automated systems

- Employers should ensure that the following personal protective equipment is used by workers to minimise occupational exposure to aerosols (particles) of the notified chemical during reformulation:
  - Respiratory protection

- Service personnel should wear cotton or disposable gloves and ensure adequate ventilation is present during cleaning processes involving the notified chemical and during routine maintenance and repairs.

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

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

- If products and mixtures containing the notified chemical are classified as hazardous to health in accordance with the *Approved Criteria for Classifying Hazardous Substances* [NOHSC:1008(2004)] workplace practices and control procedures consistent with provisions of State and Territory hazardous substances legislation must be in operation.

Disposal

- The notified chemical should be disposed of to landfill.

Emergency procedures

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

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(2) of the Act; if
- the function or use of the chemical has changed from as a colour developer for thermal paper, or is likely to change significantly;
- the amount of chemical being introduced has increased from up to 10 tonnes/annum, 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.

No additional secondary notification conditions are stipulated.

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

APPENDIX A: PHYSICAL AND CHEMICAL PROPERTIES

Melting Point Between 160 and 200ºC

Method	OECD TG 102 Melting Point/Melting Range. EC Directive 92/69/EEC A.1 Melting/Freezing Temperature.
Remarks	The notified chemical was found to melt between approximately 160 and 200ºC, although Differential Scanning Calorimetry (DSC) indicated melting onset at approximately 140 ºC.
Test Facility	Huntingdon Life Sciences Ltd (2002)

Density 1250 Kg/m^3 at 20ºC

Method	OECD TG 109 Density of Liquids and Solids. EC Directive 92/69/EEC A.3 Relative Density.
Remarks	Determined using a pyknometer at 20ºC
Test Facility	Huntingdon Life Sciences Ltd (2002)

Vapour Pressure 6×10^{-10} kPa at 25ºC

Method	OECD TG 104 Vapour Pressure. EC Directive 92/69/EEC A.4 Vapour Pressure.
Remarks	Determined using a vapour pressure balance
Test Facility	Huntingdon Life Sciences Ltd (2002)

Water Solubility $\leq 3 \times 10^{-5}$ g/L at 20ºC

Method	OECD TG 105 Water Solubility. EC Directive 92/69/EEC A.6 Water Solubility.
Remarks	Column Elution Method. Test substance concentration was determined by HPLC. All water solubility measurements were close to or below the limit of detection (LOD; 0.03 mg/L) hence the solubility was reported to be ≤ LOD.
Test Facility	Huntingdon Life Sciences Ltd (2002a)

Partition Coefficient (n-octanol/water) log Kow = 2.0 at 25ºC

Method	OECD TG 117 Partition Coefficient (n-octanol/water). EC Directive 92/69/EEC A.8 Partition Coefficient.
Remarks	HPLC Method. The partition coefficient was determined by interpolation from a calibration curve constructed from known standards (log Kow range 1.0 – 4.5) in accordance with the guidelines above.
Test Facility	Huntingdon Life Sciences Ltd (2002a)

Adsorption/Desorption log K_{oc} = 4.5 at 25ºC

Method	OECD TG 121: Estimation of the Adsorption Coefficient (Koc) on Soil and Sewage Sludge using High Performance Liquid Chromatography
Remarks	HPLC Method. The adsorption coefficient was determined by interpolation from a calibration curve constructed from known standards (log K_{oc} range 1.43-5.38) in accordance with the guidelines above.
Test Facility	Huntingdon Life Sciences Ltd (2002a)

Particle Size 6% by mass of the notified chemical is smaller than 10 μm.

Method OECD TG 110 Particle Size Distribution/Fibre Length and Diameter Distributions.

Range (μm)	*Mass (%)*
0.5 – 10.4	5.9
10.4 – 30.0	16.1
30.0 – 60.0	17.2
60.0 - 105	12.3
>105	1.9
>125	46.6

Remarks The particle size distribution was initially examined using sieve analysis. As greater than 10% by weight of the test substance was found to pass a 75 micron sieve, it was further examined by image analysis.
Test Facility Huntingdon Life Sciences Ltd (2002)

Flammability (Solids) The notified chemical was not highly flammable.

Method EC Directive 92/69/EEC A.10 Flammability (Solids).
Remarks Determined using a test mould and an ignition source.
Test Facility Huntingdon Life Sciences Ltd (2002)

Relative Autoignition Temperature for Solids > 400°C.

Method EC Directive 92/69/EEC A.16 Relative Self-Ignition Temperature for Solids.
Remarks There was no exothermic reaction of UU, indicating that it does not self-ignite below 400°C.
Test Facility Huntingdon Life Sciences Ltd (2002)

Explosive Properties The notified chemical was not explosive.

Method EC Directive 92/69/EEC A.14 Explosive Properties.
Remarks Koenen test apparatus was used for determination of sensitivity to heat (flame), a fall hammer for determination of sensitivity to shock and a friction test apparatus for determination of sensitivity to shock and a friction test apparatus for determination of sensitivity to friction.
Test Facility Huntingdon Life Sciences Ltd (2002)

Oxidizing Properties The notified chemical is not oxidising.

Method EC Directive 92/69/EEC A.17 Oxidizing Properties (Solids).
Test Facility Huntingdon Life Sciences Ltd (2002)

APPENDIX B: TOXICOLOGICAL INVESTIGATIONS

B.1. Acute toxicity – oral

TEST SUBSTANCE	Notified chemical (>70%)
METHOD	OECD TG 401 Acute Oral Toxicity. EC Directive 92/69/EEC B.1 Acute Toxicity (Oral).
Species/Strain	Rat/Hsd:Sprague-Dawley(CD)
Vehicle	1% w/v aqueous methylcellulose.
Remarks - Method	Animals were treated by oral gavage.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	5 males	2000	0
2	5 females	2000	0

LD50	>2000 mg/kg bw
Signs of Toxicity	All animals were considered to have achieved satisfactory bodyweight gains throughout the study. Clinical signs of reaction to treatment were confined to piloerection, seen in all rats accompanied by abnormal faeces in all females.
Effects in Organs	No macroscopic abnormalities were observed for animals killed at study termination on Day 15.
CONCLUSION	The notified chemical is of low toxicity via the oral route.
TEST FACILITY	Huntingdon Life Sciences Ltd (1999a)

B.2. Acute toxicity – dermal

TEST SUBSTANCE	Notified chemical (>70%)
METHOD	OECD TG 402 Acute Dermal Toxicity. EC Directive 92/69/EEC B.3 Acute Toxicity (Dermal).
Species/Strain	Rat/Hsd:Sprague-Dawley(CD)
Vehicle	1% w/v aqueous methylcellulose.
Type of dressing	Semi-occlusive.
Remarks - Method	All animals received a single topical application of the test substance for 24 hours.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	5 males	3161	0
2	5 females	3161	0

LD50	> 3161 mg/kg bw
Signs of Toxicity - Local	Slight to well-defined dermal irritation (Grade 1 or 2 erythema with or without oedema Grade 1) was seen in four females and one male following removal of the dressings, resolving completely by Day 4. No dermal irritation was noted in any other animal during the study.
Signs of Toxicity - Systemic	Apart from two animals, all other animals were considered to have achieved satisfactory bodyweight gains throughout the study. One animal recorded no change in body weight on Day 8 with a low bodyweight gain on Day 15, while the other animal has a low bodyweight gain on Day 8.
Effects in Organs	None

CONCLUSION	The notified chemical is of low toxicity via the dermal route.
TEST FACILITY	Huntingdon Life Sciences Ltd (1999b)

B.3. Irritation – skin

TEST SUBSTANCE	Notified chemical (>70%)
METHOD	OECD TG 404 Acute Dermal Irritation/Corrosion. EC Directive 92/69/EEC B.4 Acute Toxicity (Skin Irritation). EPA Health Effects Test Guidelines, OPPTS 870.2500 Acute Dermal Irritation, August 1998.
Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Vehicle	Test substance was moistened with distilled water.
Observation Period	72 hours
Type of Dressing	Semi-occlusive.
Remarks - Method	A single dermal dose of 0.5 g of the test substance was applied for 4 hours.

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	0	0	0
Oedema	0	0	0	0	0	0

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

Remarks - Results	No dermal irritation was observed following a single semi-occlusive application of the test substance for four hours.
CONCLUSION	The notified chemical is non-irritating to the skin.
TEST FACILITY	Huntingdon Life Sciences Ltd (2000a)

B.4. Irritation – eye

TEST SUBSTANCE	Notified chemical (>70%)
METHOD	OECD TG 405 Acute Eye Irritation/Corrosion. EC Directive 92/69/EEC B.5 Acute Toxicity (Eye Irritation). EPA Health Effects Test Guidelines, OPPTS 870.2400 Acute Eye Irritation, August 1998.
Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Observation Period	72 hours
Remarks - Method	Each rabbit was administered a single ocular dose of 0.1 mL of the test substance (mean weight 81 mg).

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	0	0	0	0	0
Conjunctiva: chemosis	0	0	0	0	0	0
Conjunctiva: discharge	-	-	-	-	-	-
Corneal opacity	0	0	0	0	0	0
Iridial inflammation	0	0	0	0	0	0

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

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

TEST FACILITY Huntingdon Life Sciences Ltd (2000b)

B.5. Skin sensitisation

TEST SUBSTANCE Notified chemical (>70%)

METHOD OECD TG 406 Skin Sensitisation – Magnusson and Kligman Method.
EC Directive 96/54/EC B.6 Skin Sensitisation - Magnusson and Kligman Method.
EPA Health Effects Test Guidelines OPPTS 870.2600 'Skin sensitisation' EPA 712-C-98-197. August 1998

Species/Strain Guinea pig/ Dunkin/Hartley

PRELIMINARY STUDY Maximum Non-irritating Concentration:
intradermal: 1% w/v in liquid paraffin
topical: 70% w/v in liquid paraffin

MAIN STUDY

Number of Animals Test Group: 10 Control Group: 5

INDUCTION PHASE Induction Concentration:
intradermal: 1% w/v in liquid paraffin.
topical: 70% w/v in liquid paraffin.

CHALLENGE PHASE

1st challenge topical: 35% w/v and 70% w/v in liquid paraffin

Remarks - Method Control animals were treated with liquid paraffin.

Signs of irritation during induction:
Intradermal injections: Well-defined irritation was seen in test animals at sites receiving the test substance, 1% w/v in liquid paraffin and slight irritation was observed in control animals receiving liquid paraffin.
Topical application: No erythema was observed in test or control animals

RESULTS

Animal	*Challenge Concentration*	*Number of Animals Showing Skin Reactions after: 1st challenge*	
		24 h	*48 h*
Test Group	35%	0	0
	70%	0	0
Control Group	35%	0	0
	70%	0	0

Remarks - Results Following the challenge phase, no signs of skin reaction were noted for

	animals in both the control and test groups. No concurrent positive control was used in this study. However, the sensitivity of the method is checked periodically at the laboratory with a known moderate sensitizer, hexyl cinnamic aldehyde (HCA).
CONCLUSION	There was no evidence of reactions indicative of skin sensitisation to the notified chemical under the conditions of the test.
TEST FACILITY	Huntingdon Life Sciences Ltd (2001)

B.6. Repeat dose toxicity

TEST SUBSTANCE	Notified chemical (>70%)
METHOD	OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents. Notification No. 700 - Planning and Coordination Bureau, Environmental Agency Notification No. 1039 – Pharmaceutical Affairs Bureau, Ministry of Health and Welfare Notification No. 1014 – Basic Industries Bureau, Ministry of International Trade and Industry (MITI), December 1986
Species/Strain	Rat/Crj: CD(SD) IGS
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	Gum arabic solution
Remarks – Method	No significant deviation from test protocol. A 14-day preliminary repeated-dose oral toxicity study was carried out at three doses of 50, 250 or 1,000 mg/kg bw/day. Abnormalities were noted in haematological and blood chemical examinations at 1,000 mg/kg bw/day. Therefore, high dose level at 1,000 mg/kg bw and two lower doses at 200 and 40 mg/kg bw/day were set for the main study. Recovery groups were prepared in the 1,000 mg/kg bw/day and vehicle control groups. Recovery animals were sacrificed after 14-day treatment-free recovery period

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw/day*	*Mortality*
Control	6/sex	0	0
Low dose	6/sex	40	0
Mid dose	6/sex	200	0
High dose	6/sex	1000	0
Control recovery	6/sex	0	0
High dose recovery	6/sex	1000	0

Mortality and Time to Death

All animals survived until schedule necropsy.

Clinical Observations

The notified chemical did not produce any abnormal clinical signs, and had no effects on body weights or food intake.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

In haematological examinations at the termination of the dosing period, decreases in activated partial

thromboplastin time in the 40, 200 and 1000 mg/kg male rats and an increase in white blood cell count in the 1000 mg/kg female rats were noted. At the termination of recovery period, only change noted was an increase in mean corpuscular haemoglobulin in the males of 1000 mg/kg bw/day. These changes were considered to be incidental and were not toxicologically significant.

In clinical chemistry examination, only change noted in treatment groups was an increase in cholinesterase activity in the males of 40 mg/kg bw/day group at the termination of dosing period. However, as no such effect was noted in females of 40 mg/kg bw/day group and in animals from 200 and 1000 mg/kg bw/day, this effect was unlikely to be test substance related and is therefore, not considered to be an adverse effect.

At the termination of recovery period, a decrease in glucose level and an increase in total bilirubin levels were noted in the males of 1000 mg/kg bw/day group. In the females of the same group, an increase in cholinesterase activity, total protein and albumin levels and A/G ratio were noted. The changes were considered to be incidental as these were not noted at the termination of the dosing period and changes in the recovery animals were not suspected as a result of delayed toxicity of the test substance.

No significant finding was noted in urinalysis at the termination of treatment and recovery periods.

Effects in Organs

Increased absolute and relative adrenal gland weights were noted in males only at 40 mg/kg bw/day. However, as no such effect was noted in females of 40 mg/kg bw/day group and in animals from 200 and 1000 mg/kg bw/day, this effect was unlikely to be test substance related and is therefore, not considered to be an adverse effect. In addition, histopathological examination did not reveal any abnormality in adrenal glands of 40 mg/kg bw/day animals. Also, no differences to the mean absolute and relative organ weights were seen in all treatment groups after the treatment and recovery periods, when compared with the control animals of either sex.

Significant abnormality at necropsy and during histological examination was not detected in any treatment groups at the termination of dosing and recovery periods.

CONCLUSION

The No Observed Effect Level (NOEL) was established as 1,000 mg/kg bw/day in this study, based on lack of effect at the highest tested dose (1,000 mg/kg bw/day).

TEST FACILITY	Hita Laboratory (2000a)

B.7. Genotoxicity – bacteria

TEST SUBSTANCE	Notified chemical (>70%)
METHOD	OECD TG 471 Bacterial Reverse Mutation Test.
Species/Strain	*S. typhimurium*: TA1535, TA1537, TA98, TA100 *E. coli*: WP2uvrA
Metabolic Activation System	S9 fraction Phenobarbital/β-naphthoflavone induced rat liver
Concentration Range in Main Test	a) With metabolic activation: 313 - 5000 µg/plate b) Without metabolic activation: 313 - 5000 µg/plate
Vehicle	Distilled water, dimethylsulfoxide (for some positive controls)
Remarks - Method	No significant protocol deviations. In the pre-experiment, the concentration range of the test item was 5-5000 µg/plate. Neither toxic effect of the test substance nor increases in the number of the revertant colonies were observed, regardless of the presence of metabolic activation or the absence of metabolic activation. On the other hand, precipitation of the test substance was detected at the dose of more than 20 µg/plate, in the absence and presence of metabolic activation, but it did not have a bad influence on the observation of the revertant colonies.

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/plate) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	>5000	>5000	313	Negative
Test 2	Not performed	>5000	313	Negative
Present				
Test 1	>5000	>5000	313	Negative
Test 2	Not performed	>5000	313	Negative

Remarks - Results

No substantial increase in revertant colony numbers of bacterial strains was observed following treatment with the notified chemical at any dose level, neither in the presence nor absence of metabolic activation.

The positive controls, 2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide, sodium azide, 9-aminoacridine, benzo[a]pyrene, 2-aminoanthracene) produced a significant increases in the number of the revertant colonies, as compared with the negative control with all bacterial strains. These results confirmed the efficacy of the test system.

CONCLUSION

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

TEST FACILITY

Genetic Laboratory (2000)

B.8. Genotoxicity – in vitro

TEST SUBSTANCE

Notified chemical (>70%)

METHOD

OECD TG 473 In vitro Mammalian Chromosome Aberration Test.

Species/Strain: Chinese Hamster

Cell Type/Cell Line: Chinese hamster lung fibroblasts (CHL/IU cells)

Metabolic Activation System: S9 fraction Phenobarbital/β-naphthoflavone induced rat liver

Vehicle: 0.5% Carboxymethyl Cellulose Sodium Salt

Remarks - Method: No significant protocol deviations.
Mitomycin C (MMC) and cyclophosphamide monohydrate (CPA) were used as positive controls.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	0*, 500*, 1580*, 5000*	6	24
Test 2	0*, 500*, 1580*, 5000*	24	24
Present			
Test 1	0*, 500*, 1580*, 5000*	6	24
Test 2	Not performed		

*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	Not performed	>5000	≥500	Negative
Test 2	Not performed	>5000	≥500	Negative
Present				
Test 1	Not performed	>5000	≥500	Negative
Test 2	Not performed			

Remarks - Results	Under the experimental conditions reported, the test substance did not induce chromosome aberrations as determined by the chromosome aberration test in CHU/IU cells (Chinese hamster lung fibroblast). MMC and CPA were used as positive controls and showed distinct increases in cells with chromosomal aberrations.
CONCLUSION	The notified chemical was not clastogenic to Chinese Hamster CHL/IU Cells treated in vitro under the conditions of the test.
TEST FACILITY	Hita Laboratory (2000b)

APPENDIX C: ENVIRONMENTAL FATE AND ECOTOXICOLOGICAL INVESTIGATIONS

C.1. Environmental Fate

C.1.1. Ready biodegradability

TEST SUBSTANCE	UU (product containing the notified chemical at >70%)
METHOD	OECD TG 301 C Ready Biodegradability: Modified MITI Test (I).
Inoculum	Activated sludge samples from 4 sewage plants, 3 rivers, 2 bays and 1 lake
Exposure Period	28 days
Auxiliary Solvent	None reported
Analytical Monitoring	HPLC
Remarks - Method	The oxygen uptake of the test substance (100 mg/L) in inoculated medium was measured over 28 days in a darkened enclosed respirometer, conducted in accordance with the guidelines above. A reference control (aniline) was run in parallel. Biodegradation is expressed as the percentage oxygen uptake, corrected for the blank, of the theoretical uptake (ThOD). Test conditions were: 25 ± 1°C, pH 7.0.

RESULTS

Test substance		*Aniline*	
Day	*% Degradation**	*Day*	*% Degradation*
7	0.33	7	59
14	1.00	14	78
21	1.00	21	80
28	1.00	28	78

*Mean of 3 replicates

Remarks - Results	The pass level (60% of ThOD) was not reached by the test substance over the test period, thus it is not considered to be readily biodegradable. The percentage degradation of the reference substance (aniline) surpassed the 40% and 65% pass levels by days 7 and 14 respectively, thereby validating the test.
CONCLUSION	The test substance, and by inference the notified chemical, is not readily biodegradable
TEST FACILITY	Kurume Laboratory (2000)

C.2. Ecotoxicological Investigations

C.2.1. Acute toxicity to fish

TEST SUBSTANCE	UU (product containing the notified chemical at >70%)
METHOD	OECD TG 203 Fish, Acute Toxicity Test – Semi static EC Directive 92/69/EEC C.1 Acute Toxicity for Fish – Semi static
Species	Rainbow trout (*Oncorhynchus mykiss*)
Exposure Period	96 hours
Auxiliary Solvent	Acetone (0.01% v/v)
Water Hardness	162 mg $CaCO_3$/L
Analytical Monitoring	HPLC
Remarks – Method	After a range-finding test, a limit test with a nominal concentration of 100 mg/L was conducted along with a diluent control and a solvent control (100 µL acetone/L), according to the guidelines above. A water

accommodated fraction (WAF) of the test substance was prepared as follows. Acetone (1.5 mL) was added to test substance (1.5 g) and the mixture was dispersed in approximately 1.5 L of diluent water. The medium was treated with ultrasound for 30 min and the volume adjusted to 2 L with diluent water. The medium was poured into one of two glass aspirators and made up to a volume of 15 L. The procedure was conducted in duplicate to provide a total volume of 30 L. The mixture was then stirred in darkness for approximately 15 – 22 hours. After being allowed to stand for between 1 - 3 hours, approximately 50 mL were withdrawn from the middle of the vessel and discarded. A sample (20 L) of the aqueous phase (the WAF) was removed mid-vessel and was used as the test medium. The fish, 10 per test solution, were observed for mortality and sublethal responses at 0.25 h, 2 h, 24 h and then every 24 hours. Test conditions were: 13.8 – 15.1°C, pH 7.8-8.4, and 73 – 105% ASV dissolved O_2. There was a daily batchwise renewal of the media.

RESULTS

Concentration mg/L		*Number of Fish*	*Mortality*				
Nominal	*Measured*		*2 h*	*24 h*	*48 h*	*72 h*	*96 h*
Diluent Control	< LOD	10	0	0	0	0	0
Solvent control	< LOD	10	0	0	0	0	0
100	0.542*	10	0	0	0	0	0

*Geometric mean of measurements at 0, 24, 72 and 96 h; LOD = 0.03 mg/L

LL50	> 100 mg/L at 96 hours (based on loading rate, WAF)
NOEL	100 mg/L at 96 hours (based on loading rate, WAF)
Remarks – Results	No deaths or adverse effects on the fish were noted due to the test substance or diluent control. Aggressive behavior was exhibited by one fish in the solvent control vessel after 15 minutes of exposure, so this fish was isolated behind a screen in the vessel. After two hours, the fish had escaped from the isolation area; since no further aggression towards the other fish was noted, the screen was removed from the test vessel. This is not thought to have affected the reliability of the test. The aqueous mixture of the test substance was a white, non-homogeneous, hazy dispersion with undissolved material on the base of the preparation vessel. Throughout the study, the WAF was a white non-homogeneous hazy dispersion with undissolved material on the base of the test vessel.

CONCLUSION The test substance, and by inference the notified chemical, is not harmful to fish up to the limit of its solubility in water

TEST FACILITY Huntingdon Life Sciences Ltd. (2002b)

C.2.2. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE UU (product containing the notified chemical at >70%)

METHOD OECD TG 202 *Daphnia* sp. Acute Immobilisation Test - Static
EC Directive 92/69/EEC C.2 Acute Toxicity for Daphnia - Static

Species	*Daphnia magna*
Exposure Period	48 hours
Auxiliary Solvent	Acetone (0.01% v/v)
Water Hardness	260 mg $CaCO_3$/L
Analytical Monitoring	HPLC

Remarks - Method	After a range-finding test, a limit test at a nominal concentration of 100 mg/L was conducted along with a diluent control and a solvent control, according to the guidelines above. A water accommodated fraction (WAF) of the test substance was prepared as follows. Acetone (0.2 mL) and test substance (200 mg) were added to diluent medium (2 L). The medium was treated with ultrasound for 30 min, covered to exclude light and stirred for approximately 15 h. After being allowed to stand for about 4 h, approximately 50 mL were withdrawn from the middle of the vessel and discarded. A sample (1400 mL) of the aqueous phase (the WAF) was removed mid-vessel and used as the test medium. Test conditions were: 20.5 – 20.9°C, pH 7.8-7.9, and 96 - 98% ASV dissolved O_2.

RESULTS

Concentration mg/L		*Number of D. magna*	*Number Immobilised*	
Nominal	*Measured*		*24 h*	*48 h*
Diluent control	< LOD	20	0	0
Solvent control	< LOD	20	Not reported	5†
100 mg/L	0.239*	20	0	0

*Geometric mean of measurements at 0 and 48 h; †See Remarks – Results; LOD = 0.03 mg/L

EL50	> 100 mg/L at 48 hours (based on loading rate, WAF)
NOEL	100 mg/L at 48 hours (based on loading rate, WAF)
Remarks - Results	The aqueous mixture of the test substance was a colourless, non-homogeneous dispersion with undissolved material on its surface and on the base of the preparation vessel. The WAF was clear and colourless. All of the test organisms in one of the solvent control vessels were immobile after 48 hours. The medium in this vessel was observed to be contaminated with an unidentified yellow/brown coloured substance. As the contaminant was not observed in any other vessel and no other daphnids were affected, the immobility of daphnids was attributed to the presence of the unidentified substance. The test was valid with respect to dissolved O_2 levels but not with respect to mortality in the controls. However, as no daphnia were immobilised in the other control and test substance vessel, the test was considered reliable.
CONCLUSION	The test substance, and by inference the notified chemical, is not harmful to aquatic invertebrates up to the limit of its solubility in water
TEST FACILITY	Huntingdon Life Sciences Ltd. (2002c)

C.2.3. Chronic toxicity to aquatic invertebrates

TEST SUBSTANCE	UU (product containing the notified chemical at >70%)
METHOD	OECD No. 211 *Daphnia magna*, Reproduction test – Semi Static
Species	*Daphnia magna*
Exposure Period	21 days
Auxiliary Solvent	Acetone (0.01% v/v)
Water Hardness	250 – 321 $CaCO_3$ mg/L
Analytical Monitoring	HPLC
Remarks - Method	In a preliminary test to determine the stability of the test substance in the algal nutrient medium, a filtered water accommodated fraction (filtered WAF) of the test substance was prepared as follows. An aqueous mixture was prepared by adding 200 µL of acetone stock solution (1 g test substance/L acetone) to 2 L of test medium to give a loading rate of 100

mg/L test substance. This mixture was stirred for 19 h and filtered through a 0.45 µm membrane filter. The resulting fraction (filtered WAF) was observed to be clear and colourless. Daphnia and algae were added to simulate the test conditions, the solution was left to stand for 72 h , duplicate samples were taken at t=0, 6, 24, 48 and 72 hours and concentrations were analysed.

Based on the preliminary test, a limit test was conducted at a nominal concentration of 0.05 mg/L along with solvent control (0.1 mL acetone/L), in accordance with the guidelines above and in compliance with GLP standards and principles. Test conditions over the exposure period were: 19.3 – 21.1°C, pH 7.2 – 8.7 and 5.2 – 9.7 mg O_2/L. Test solutions were renewed every 48 or 72 h. Twenty vessels contained the solvent control and twenty vessels contained the test substance at a loading rate of 0.05 mg/L, with each vessel containing a neonate (<24 h old) daphnid.

RESULTS

		Day 21		
Nominal Concentration (mg/L)	*Mean Measured Concentration (mg/L)*	*Mean Percent Adult Survival*	*Mean Number of Offspring Produced per female – cumulative*	*Mean body length of surviving parental daphnids (mm)(SD)*
Solvent control	< LOD	80	161.1	4.6 (0.15)
0.05	0.042*	95	167.2	4.7 (0.12)

*Arithmetic mean for 4 concentrations over the 21 day exposure period. Each concentration (days 0-2, 9-12, 14-16 and 19-21) was calculated as a geometric mean of old and fresh solution concentrations. LOD = 0.006 mg/L.

EL50 (reproduction)	> 0.05 mg / L at 21 days (based on loading rate)
NOEL (reproduction)	0.05 mg / L at 21 days (based on loading rate)
Remarks - Results	No significant deviations to protocol were reported and all validity criteria for the test were satisfied.

Test substance concentrations during refreshment periods at day 9-12 and day 14-16 were significantly lower than nominal concentrations with high variations in duplicate samples. This was considered to be due to the low solubility of the test substance. A mean exposure concentration was therefore calculated using all measurements.

No treatment related mortality of parental daphnids was observed. The mean body lengths of the parental daphnids in both test groups at the end of the test were comparable, differing by 2%. No effects on reproduction were observed at the maximum water solubility of the test substance. Statistical analyses were not conducted for reproduction and body length since the control and test organisms were comparable with respect to both parameters.

CONCLUSION The notified chemical, and by inference the notified chemical, is not harmful to aquatic invertebrates with long lasting effects up to the limit of its solubility in water

TEST FACILITY NOTOX B.V. (2006)

C.2.4. Algal growth inhibition test

TEST SUBSTANCE UU (product containing the notified chemical at >70%)

METHOD OECD TG 201 Alga, Growth Inhibition Test.

	EC Directive 92/69/EEC C.3 Algal Inhibition Test.
Species	*Pseudokirchneriella subcapitata* (formerly known as *Selenastrum capricornutum*)
Exposure Period	72 hours
Concentration Range	Nominal: 100 mg/L Actual: 5.06 mg/L (geometric mean of concentrations measured at 0 and 72 h)
Auxiliary Solvent	Acetone (0.01% v/v)
Water Hardness	180 mg $CaCO_3$/L
Analytical Monitoring	HPLC
Remarks - Method	After a range finding test, a limit test at a nominal concentration of 100 mg/L along with an algal nutrient medium control and a solvent control group (100 µL acetone/L) was conducted according to the guidelines above. A water accommodated fraction (WAF) of the test substance was prepared as follows. Acetone (0.2 mL) and test substance (200 mg) were added to sterile algal nutrient medium (2 L). The medium was treated with ultrasound for 30 min, covered to exclude light and stirred for approximately 22 h. After being allowed to stand for about 2.5 h, approximately 50 mL were withdrawn from the middle of the vessel and discarded. A sample (1500 mL) of the aqueous phase (the WAF) was removed mid-vessel and an aliquot of the algal inoculum (5.8 mL) was added to the WAF (1300 mL). An aliquot (100 mL) of the inoculated test medium was added to each test vessel and an aliquot (100 mL) of the remaining medium without test algal cells was added to two additional vessels. Test conditions: 23.6 – 24.1°C, pH 7.9 – 10.0, continuous illumination.

RESULTS

Biomass		*Growth*	
E_bL_{50} mg/L at 72 h	*NOEL mg/L*	*E_rL_{50} mg/L at 72 h*	*NOEL mg/L*
> 100	100	> 100	100

Remarks - Results	No significant deviations to protocol were reported and validity criteria were satisfied. The aqueous mixture of the test substance was an off-white, non-homogeneous dispersion with undissolved material on its surface and on the base of the preparation vessels. No microscopic abnormalities of the algal cells were detected. The growth of *Pseudokirchneriella subcapitata* was not inhibited after exposure to a water accommodated fraction of the test substance prepared in algal nutrient medium at a nominal loading rate of 100 mg/L or to a concentration in excess of its limit of aqueous solubility in the medium (5.06 mg/L, measured).
CONCLUSION	The notified chemical, and by inference the notified chemical, is not harmful to algae up to the limit of its solubility in water
TEST FACILITY	Huntingdon Life Sciences Ltd. (2002d)

C.2.5. Inhibition of microbial activity

TEST SUBSTANCE	UU (product containing the notified chemical at >70%)
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test. EC Directive 88/302/EEC C.11 Biodegradation: Activated Sludge

	Respiration Inhibition Test
Inoculum	Activated sewage sludge
Exposure Period	3 hours
Concentration Range	Nominal: 1, 10 and 100 mg/L Actual: Not measured
Remarks – Method	A definitive test was conducted according to the guidelines above at nominal test substance concentrations of 1, 10 and 100 mg/L. A blank control and reference (3,5-dichlorophenol) control were run in parallel. The rate of respiration was determined after 3 h contact time and compared to the results from the control and reference material. Test conditions: 18.6 – 19.6°C, pH 7.4–8.0.

RESULTS

IL50 (3 h)	> 100 mg/L (based on loading rate)
NOEL (3 h)	100 mg/L
Remarks – Results	The validation criteria for the control respiration rates and reference material, (3,5-dichlorophenol) EC_{50} were satisfied.

CONCLUSION	The notified chemical, and by inference the notified chemical, is not expected to inhibit microbial respiration
TEST FACILITY	Huntingdon Life Sciences Ltd. (2002e)

BIBLIOGRAPHY

Genetic Laboratory, JBS, Inc. (2000) Notified chemical: Reverse Mutation Test using microorganism. Final Report 6 July 2000, Test Number: 4065 for Asahi Chemical Industry Co Ltd, Chiyoda-ku 100-8550, JAPAN. Genetic Laboratory, JBS, Inc. 2078-1 Akahama, Yorii-machi, Osato-gun, Saitama, Japan (Unpublished report provided by notifier).

Hita Laboratory (2000a) Notified chemical: Twenty-eight day repeated-dose oral toxicity study in rats. Final Report 26 July 2000, Project Number: B11-0571 for Asahi Kasei Corporation, Kanagawa 210-0863, JAPAN. Hita Laboratory, Chemicals Evaluation and Research Institute, Japan (Unpublished report provided by notifier).

Hita Laboratory (2000b) Notified chemical: Chromosomal aberration test using cultured mammalian cells. Final Report 9 August 2000, Study Number: K06-0779 for Asahi Kasei Corporation, Kanagawa 210-0863, JAPAN. Hita Laboratory, Chemicals Evaluation and Research Institute, Japan (Unpublished report provided by notifier).

Huntington Life Sciences Ltd (1999a) Notified chemical: Acute oral toxicity to the rat. Final Report 28 July 1999, Project Number: ASI 109/993243/AC for Asahi Chemical Industry Co Ltd, Chiyoda-ku 100-8550, JAPAN. Huntingdon Life Sciences Ltd, Huntingdon, Cambridgeshire, PE28 4HS, England (Unpublished report provided by notifier).

Huntington Life Sciences Ltd (1999b) Notified chemical: Acute dermal toxicity to the rat. Final Report 21 September 1999, Project Number: ASI 110/993249/AC for Asahi Chemical Industry Co Ltd, Chiyoda-ku 100-8550, JAPAN. Huntingdon Life Sciences Ltd, Huntingdon, Cambridgeshire, PE28 4HS, England (Unpublished report provided by notifier).

Huntington Life Sciences Ltd (2000a) Notified chemical: Skin irritation to the rabbit. Final Report 11 July 2000, Project Number: ASI 110/993249/AC for Asahi Chemical Industry Co Ltd, Chiyoda-ku 100-8550, JAPAN. Huntingdon Life Sciences Ltd, Huntingdon, Cambridgeshire, PE28 4HS, England (Unpublished report provided by notifier).

Huntington Life Sciences Ltd (2000b) Notified chemical: Eye irritation to the rabbit. Final Report 13 September 2000, Project Number: ASI 146/003191/SE for Asahi Chemical Industry Co Ltd, Chiyoda-ku 100-8550, JAPAN. Huntingdon Life Sciences Ltd, Huntingdon, Cambridgeshire, PE28 4HS, England (Unpublished report provided by notifier).

Huntington Life Sciences Ltd (2001) Notified chemical: Skin sensitisation to the Guinea-Pig (Manusson and Kligman Method). Final Report 4 April 2001, Project Number: ASI 186/012376/SS for Asahi Kasei Corporation, Kanagawa 210-0863, JAPAN. Huntingdon Life Sciences Ltd, Huntingdon, Cambridgeshire, PE28 4HS, England (Unpublished report provided by notifier).

Huntington Life Sciences Ltd (2002a) Notified chemical: Physico-Chemical Properties. Final Report 3 July 2002, Project Number: ASI232/022489 for Asahi Kasei Corporation, Kanagawa 210-0863, JAPAN. Huntingdon Life Sciences Ltd, Huntingdon, Cambridgeshire, PE28 4HS, England (Unpublished report provided by notifier).

Huntingdon Life Sciences Ltd. (2002b) UU Acute toxicity to fish. (Project number ASI 233/023312, 13 August 2002) Huntingdon, England, Huntingdon Life Sciences Ltd (Unpublished report submitted by the notifier).

Huntingdon Life Sciences Ltd. (2002c) UU Acute toxicity to *Daphnia magna* (Project No. ASI 234/023313, 13 August 2002) Huntingdon, England, Huntingdon Life Sciences Ltd (Unpublished report submitted by the notifier).

Huntingdon Life Sciences Ltd. (2002d) UU Algal growth inhibition assay (Project number ASI 235/023314, 13 August 2002) Huntingdon, England, Huntingdon Life Sciences Ltd (Unpublished report submitted by the notifier).

Huntingdon Life Sciences Ltd. (2002e) UU Activated sludge-respiration inhibition test (Project number ASI 236/014504, 25 February 2002) Huntingdon, England, Huntingdon Life Sciences Ltd (Unpublished report submitted by the notifier).

Kurume Laboratory (2000) Biodegradation test of UD by microorganisms (Test No. S99-3489, 13489) Fukuoka, Japan, Kurume Laboratory Chemicals Evaluation and Research Institute (Unpublished report submitted by notifier)

NOHSC (2003) National Code of Practice for the Preparation of Material Safety Data Sheets, 2nd edition [NOHSC:2011(2003)]. National Occupational Health and Safety Commission, Canberra, Australian Government Publishing Service.

NOHSC (1994) National Code of Practice for the Labelling of Workplace Substances [NOHSC:2012(1994)]. National Occupational Health and Safety Commission, Canberra, Australian Government Publishing Service.

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

NOTOX B.V. (2006) *Daphnia magna*, reproduction test with UU (semi-static) (Project No. 450697, 19 June 2006) 's-Hertogenbosch, The Netherlands, NOTOX B.V. (Unpublished report submitted by notifier)

OECD (2000) Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures (No. 23) Paris, Organisation for Economic Co-operation and Development.

NTC (National Transport Commission) 2007 Australian Code for the Transport of Dangerous Goods by Road and Rail (ADG code), 7th Edition, Commonwealth of Australia