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NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

FULL PUBLIC REPORT

CIM-09

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 the Environment, Water, Heritage and the Arts.

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 334-336 Illawarra Road, Marrickville NSW 2204.

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:

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Director
NICNAS

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1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT(S)
Canon Australia Pty Ltd (ABN: 66 005 002 951)
1 Thomas Holt Drive
North Ryde NSW 2113

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

EXEMPT INFORMATION (SECTION 75 OF THE ACT)
Data items and details claimed exempt from publication:
Chemical Name, Other Names, CAS Number, Molecular and Structural Formulae, Spectral Data, Molecular Weight, Purity, Impurities, Import Volume, Use Details.

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

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT(S)
LVC/743

NOTIFICATION IN OTHER COUNTRIES
USA (2007); UK (2007); Switzerland (2008); Japan (2008); Korea (2008); Philippines (2008)

2. IDENTITY OF CHEMICAL

MARKETING NAME(S)
CIM-09, C-BW1, Brown C-BW1, Brown C-BW1 Liq.

MOLECULAR WEIGHT
>1000 Da

ANALYTICAL DATA
Reference NMR, IR, HPLC, LC/MS, UV/vis spectra were provided.

3. COMPOSITION

DEGREE OF PURITY >80%

ADDITIVES/ADJUVANTS None

4. PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE AT 20°C AND 101.3 kPa: Dark brown powder

Property	Value	Data Source/Justification
Melting Point/Freezing Point	Not observed below 360°C	Measured
Boiling Point	Not determined	Decomposed prior to melting
Density	1600 kg/m^3 at 22.5 ± 0.5°C	Measured
Vapour Pressure	< 1.9 x 10^{-8} kPa at 25°C	Measured
Water Solubility	262 - 278 g/L at 20°C	Measured

Hydrolysis as a Function of pH	Stable (half-life at 25ºC > 1 year at ph 4, 7 and 9)	Measured
Partition Coefficient (n-octanol/water)	log Pow = -2.73 at 21.5ºC	Measured
Surface Tension	71.7 mN/m at 22.0 ± 0.2ºC	Measured
Adsorption/Desorption	log K_{oc} = < 1.25 at 30°C	Measured
Dissociation Constant	pKa = -1.63 to -0.71, 1.22 and 2.56	Calculated
Particle Size	Inhalable fraction (<100 μm): 27% Respirable fraction (<10 μm): 4.58%	Measured
Flash Point	Not determined	Low vapour pressure solid
Solid Flammability	Not highly flammable	Measured
Autoignition Temperature	Does not self-ignite below 400ºC	Measured
Explosive Properties	Not explosive	Measured
Oxidising Properties	Not expected to be oxidising	Estimated

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

Reactivity
The notified chemical is not expected to be reactive under normal conditions of use.

5. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will be imported as a component (<5%) of inkjet printer ink contained within sealed ink cartridges.

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

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

PORT OF ENTRY
Sydney

IDENTITY OF MANUFACTURER/RECIPIENTS
Canon Australia Pty Ltd and office equipment retailers and offices nationwide.

TRANSPORTATION AND PACKAGING
Imported ink cartridges (5 mL – 900 mL) containing the notified chemical (each individually sealed in a plastic bag and packaged in a box) will be stored at the notifier's warehouse prior to distribution to offices and office equipment retailers nationwide.

USE
The notified chemical will be used as an ink component (<5%) in ink cartridges for use in inkjet printers.

OPERATION DESCRIPTION
No manufacture or reformulation will occur in Australia. Sealed ink cartridges containing the notified chemical will be distributed to commercial and retail centres and handled by service technicians, office workers or the public, who will replace spent cartridges in printers as necessary.

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)*
Importation/ Waterside	50	<8	10-50
Storage and Transport	15	<8	10-50
Office worker/ consumer	2,000,000	10 seconds/day	2
Service Technicians	100	1	170

EXPOSURE DETAILS
Storage and transport workers will only handle the sealed cartridges containing the notified chemical and therefore exposure is not expected unless the packaging is accidentally breached.

Service technicians and office workers may be exposed to the ink containing the notified chemical (< 5%) when replacing used ink cartridges and repairing and cleaning ink jet printers. Dermal exposure is expected to be the most likely route of exposure. Instructions on how to replace the cartridges safely are included with the cartridge to minimise exposure. However, occasional dermal exposure during use of the printer may occur if the printed pages were handled inadvertently before the ink had dried, or if ink-stained parts of the printer were touched. Once the ink dries, the chemical would be bonded to the printed paper, and therefore dermal exposure to the notified chemical from contact with dried ink is not expected.

6.1.2. Public exposure
Home users may encounter dermal exposure to the ink containing the notified chemical (< 5%) when replacing used ink cartridges similar to the exposure experienced by office workers. However, home users are expected to handle ink cartridges and print less frequently, therefore exposure is expected to be less frequent when compared to that of office workers.

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	low oral toxicity LD50 > 2000 mg/kg bw
Rat, acute dermal toxicity	low dermal toxicity LD50 > 2000 mg/kg bw
Rabbit, skin irritation	non-irritating
Rabbit, eye irritation	slightly irritating
Mouse, skin sensitisation – Local lymph node assay	no evidence of sensitisation
Rat, repeat dose oral toxicity – 28 days.	NOEL 1000 mg/kg bw/day
Mutagenicity – bacterial reverse mutation	non mutagenic
Mutagenicity – bacterial reverse mutation (incorporating Prival and Mitchell modification for azo colourants)	non mutagenic
Genotoxicity – in vitro chromosome aberration	non genotoxic

Toxicokinetics, metabolism and distribution
Many azo dyes are unlikely to be absorbed through the skin because of their size and polarity (Øllgaard, 1998). Smaller species may undergo percutaneous absorption, dependent on their properties. There was no evidence from the toxicological investigations to suggest that the notified chemical was absorbed following dermal exposure. Absorption through the skin is not expected to be significant, given its relatively high molecular weight (>500 Da), high water solubility (>10 g/L), and low partition coefficient (log P < 0). These parameters demonstrate its low lipophilicity, indicating that it is not likely to cross the stratum corneum (European Commission, 2003). However, bacterial skin microflora have been reported to be able to break down azo dyes into smaller species, which may be more readily absorbed, through azo reduction (SCCNFP, 2002).

Highly sulfonated azo dyes are generally poorly absorbed from the intestine after oral intake. However, cleavage

of the azo linkage may take place in the gastrointestinal tract, through the action of azo reductases (see below). The sulfonated aromatic amines are rapidly absorbed. The orange coloured stomach contents of one high dose male and the yellow/red coloured urine observed mainly in high dose animals during the oral repeat dose study suggest that the notified chemical can be absorbed, perhaps following reduction, from the gastrointestinal tract after oral exposure.

Ultimately, the metabolites of azo dyes are expected to be excreted in the urine and bile (Brown & DeVito, 1993, referenced in Øllgaard, 1998). Sulfonation of azo dyes appears to decrease their toxicity by enhancing their urinary excretion, and that of their metabolites. The coloured urine observed in animals that had been orally administered with the notified chemical (mainly high dose animals of the repeat dose study) could be indicative of urinary excretion of metabolites of the notified chemical.

A significant proportion of the notified chemical (27%) is of inhalable particle size (<100 μm) and only a small fraction (4.58%) is of respirable size (<10 μm). The particles of inhalable size are expected to diffuse or dissolve into the mucus lining of the respiratory tract and be retained in the mucus and then transported out of the respiratory tract.

Acute toxicity
The notified chemical was found to be of low acute oral and dermal toxicity in studies conducted on rats (LD50 > 2,000 mg/kg bw). No information on the acute inhalation toxicity was available.

Irritation and Sensitisation
The notified chemical was found to be slightly irritating to the eye, though not enough to warrant hazard classification, and non-irritating to the skin.

The notified chemical was not a skin sensitiser when tested in a mouse local lymph node assay up to a concentration of 25%. Therefore, the notified chemical is unlikely to cause skin sensitisation in exposed humans.

Repeat dose toxicity
The No Observed Adverse Effect Level (NOEL) in a 28-day oral repeat dose study in rats was established as 1000 mg/kg bw/day (highest dose tested), based on the absence of toxicologically significant changes in the parameters measured at all dose levels.

Mutagenicity and Carcinogenicity
Azo dyes are a concern for their potential induction of mutagenicity and carcinogenicity, mainly through their reduction to aromatic amines in the body. Exposure to heat or sunlight has also been reported to result in breakdown of azo dyes, including some that are similar to the notified chemical (Brown and DeVito, 1993).
The notified chemical is not expected to be reductively cleaved to release any of the restricted aromatic amines specified in either the Appendix to EC Directive 76/769/EEC (EC, 2004) or the annexes of EU SCCNFP/0495/01 (SCCNFP, 2002). However, it may be degraded to form species that resemble these arylamines, some of which are suspected of being mutagenic. However, due to their significant structural modification and the suggestive negative test data of these species, the amine species may not exhibit mutagenicity.

In addition, azo dyes are known for their content of impurities, particularly for the presence of component arylamines (SCCNFP, 2002; Øllgaard *et al* 1998). The HPLC trace provided by the notifier indicates that the sample of the notified chemical contains a number of impurities that have not been identified, each present at <1%. These impurities are both more and less polar than the notified chemical and are likely to be free amine species and/or sulfonation variants. Free amines may exhibit higher toxicity than the notified chemical as they are likely to be more readily absorbed across the skin.

The notifier supplied test results showing that the notified chemical was found to be not mutagenic in bacteria (under the conditions of the Ames test used), and did not induce chromosomal aberrations in mammalian cells *in vitro*. Furthermore, the notifier also supplied a summary of test results from a study showing that the notified chemical was found to be not mutagenic using the modified Ames test for azo dyes (Prival and Mitchell, 1982). This modified test is thought to yield a greater detection of mutagenic azo dyes as it utilises a reductive pre-incubation step (during which the azo dye is reduced to amine species) before the test is carried out.

Overall, on the basis of weight of evidence the notified chemical is not expected to be genotoxic.

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

6.3. Human health risk characterisation

6.3.1. Occupational health and safety
Based on the data supplied, the notified chemical has no identified hazards. Dermal exposure of workers to the notified chemical should occur infrequently and be of small amounts, given the containment of the notified chemical within ink cartridges.

The level of repeat dermal exposure for service technicians and office workers handling sealed cartridges of printing inks containing the notified chemical at < 5% is not expected to be significant compared to the NOEL of 1000 mg/kg bw/day established in the 28 day rat study.

Overall, the risk presented by the notified chemical to the health and safety of workers is not considered to be unacceptable.

6.3.2. Public health
The exposure and hazard of the notified chemical to the members of the public during the use of inkjet printers are expected to be identical or similar to that experienced by office workers. Therefore, the risk of the notified chemical to the health of the public is not considered to be unacceptable.

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 into Australia as a component of a printer ink final product in ready-to-use cartridges. No manufacturing and reformulation of the notified chemical will take place in Australia. Environmental release of the notified chemical is unlikely to occur during importation, storage and transportation.

RELEASE OF CHEMICAL FROM USE
The ink cartridges are designed to prevent leakage and will not be opened during transport, use, installation or replacement. Therefore, release of ink containing the notified chemical to the environment is not expected under normal conditions of use. Workers at large businesses will undertake installation and replacement. If leakage or spillage does occur, the ink will be wiped up with a wet cloth or paper and disposed to landfill in accordance with federal, state and local regulations.

Cartridges are contained within the printer until the contents are consumed and then they are removed and sent for recycling or disposed to landfill. Around 5% of the ink containing the notified chemical will remain in "empty" cartridges.

Most of the notified chemical (95%) will be bound to printed paper, which will be disposed to landfill, recycled or possibly thermally decomposed.

RELEASE OF CHEMICAL FROM DISPOSAL
Around 5% of the ink containing the notified chemical will remain in "empty" cartridges. The notifier will collect the used cartridges by setting up the collection boxes in general merchandising stores and post offices, etc. The collected cartridges are sent to the subcontractor. The subcontractor disassembles the used cartridges and recycles as raw materials, for example a plastic material to be used to make plastic goods. The remaining ink separated from the used cartridges is disposed of under Australian regulations. The notifier will not recycle the used cartridges to be renewed as new cartridges by refilling the ink. The other cartridges which are not collected will be disposed to landfill.

The majority of the notified chemical will be bound within the cured printing matrix, adhering to the paper product articles and it will share the fate of the articles into which it is incorporated. It will be disposed to

landfill, recycled or possibly thermally decomposed. Recycling of treated paper may result in the release of a proportion of the notified chemical to the aquatic compartment. Waste paper is re-pulped using a variety of chemical treatments, which result in fibre separation and ink detachment from the fibres. The wastes are expected to go to trade waste sewers. Approximately 50% of the ink printed on paper will enter paper recycling and a minor proportion of the ink may be recovered during recycling in the sludge. Any quantities of notified chemical recovered with sludge during the recycling process will be disposed to landfill.

7.1.2 Environmental fate

The notified chemical is water soluble and not readily biodegradable, and could therefore be expected to pass through sewage treatment works and disperse in receiving waters. In practice, the notified chemical can be expected to precipitate during sewage treatment and in surface waters as sparingly soluble calcium salts. Bioaccumulation is not expected as the notified chemical has high molecular weight and is water soluble. For the details of the environmental fate studies please refer to Appendix C.

7.1.3 Predicted Environmental Concentration (PEC)

The PEC is estimated below based on the assumption that 50% of the imported quantity will enter paper recycling streams and be discharged in aqueous effluent following detachment from the fibre. Note that the assumption of complete release to surface water is highly conservative as the notified chemical is expected to precipitate as calcium salts.

Predicted Environmental Concentration (PEC) for the Aquatic Compartment		
Total Annual Import/Manufactured Volume	< 1000	kg/year
Proportion expected to be released to sewer	0.5	
Annual quantity of chemical released to sewer	< 500	kg/year
Days per year where release occurs	365	days/year
Daily chemical release:	< 1.37	kg/day
Water use	200.0	L/person/day
Population of Australia (Millions)	21.37	million
Removal within STP	0%	
Daily effluent production:	4,275	ML
Dilution Factor - River	1.0	
Dilution Factor - Ocean	10.0	
PEC - River:	< 0.32	µg/L
PEC - Ocean:	< 0.03	µg/L

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	LC50 > 100 mg/L	Not harmful
Daphnia Toxicity	EC50 > 100 mg/L	Not harmful
Algal Toxicity	E_rC50 > 100 mg/L	Not harmful
Lemna Toxicity	E_rC50 > 100 mg/L	Not harmful
Inhibition of Bacterial Respiration	IC50 > 1000 mg/L	Not harmful

The results from testing indicate that the notifed chemical is not harmful to aquatic life, consistent with its water solubility. The algal test was discontinued in favour of the lemna test because of indications that algal growth was inhibited by light absorption.

7.2.1 Predicted No-Effect Concentration

The PNEC can be estimated as outlined below by application of a 100-fold assessment factor, as data are available for three trophic levels.

Predicted No-Effect Concentration (PNEC) for the Aquatic Compartment		
Aquatic toxicity	> 100	mg/L
Assessment Factor	100	
PNEC:	> 1000	µg/L

7.3. Environmental risk assessment

Risk Assessment	PEC µg/L	PNEC µg/L	Q
Q - River	0.32	> 1000	**< 0.00032**
Q - Ocean	0.032	> 1000	**< 0.000032**

The notified chemical is not considered to pose a risk to the environment as risk quotients are well below one, even under the hypothetical worst case assumption that the notified chemical will be discharged to surface waters after detachment from paper fibres during recycling.

8. CONCLUSIONS AND REGULATORY OBLIGATIONS

Hazard classification
Based on the available data the notified chemical is not classified as hazardous under 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 unacceptable risk to the health of workers.

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

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

Recommendations

CONTROL MEASURES
Occupational Health and Safety

- No specific engineering controls, work practices or personal protective equipment are required for the safe use of the notified chemical itself, however, these should be selected on the basis of all ingredients in the formulation.

  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 disposal.

Regulatory Obligations

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

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

(1) Under Section 64(1) of the Act; if
- the importation volume exceeds one tonne per annum notified chemical;

(2) Under Section 64(2) of the Act; if
- the function or use of the chemical has changed from a component of inkjet printer ink, or is likely to change significantly;
- the amount of chemical being introduced has increased from one tonne per 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.

Material Safety Data Sheet
The MSDS of products containing 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/Freezing Point Not observed below 360 ºC

Method OECD TG 102 Melting Point/Melting Range.
EC Directive 92/69/EEC A.1 Melting/Freezing Temperature.
Remarks Differential scanning calorimetry.
There was evidence of gradual decomposition during the test.
Test Facility SafePharm (2007a)

Boiling Point Not determined

Remarks The boiling temperature was not determined as the notified chemical was found to decompose prior to melting (see above).
Test Facility SafePharm (2007a)

Density 1600 kg/m^3 at 22.5 ± 0.5ºC

Method OECD TG 109 Density of Liquids and Solids.
EC Directive 92/69/EEC A.3 Relative Density.
Remarks Gas comparison pycnometer.
Test Facility SafePharm (2007b)

Vapour Pressure < 1.9 x 10^{-8} 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. Balance readings were low and variable and thus statistical analysis was not meaningful. A regression slope was imposed on a chosen data point to provide an estimate of the maximum value for the vapour pressure at 25°C.
Test Facility SafePharm (2007c)

Water Solubility 262 - 278 g/L at 20ºC

Method OECD TG 105 Water Solubility.
EC Directive 92/69/EEC A.6 Water Solubility.
Remarks Flask Method. A visual assessment was conducted since there were difficulties filtering those test concentrations that near the solubility limit. A preliminary test indicated the water solubility was in the range 199 – 593 g/L.
Test Facility SafePharm (2007a)

Hydrolysis as a Function of pH Half-life at 25ºC > 1 year at ph 4, 7 and 9

Method OECD TG 111 Hydrolysis as a Function of pH.
EC Directive 92/69/EEC C.7 Degradation: Abiotic Degradation: Hydrolysis as a Function of pH.

pH	*T (°C)*	*$t_{½}$ (hours)*
4	50	> 120
7	50	> 120
9	50	> 120

Remarks At pH 4, 7 and 9, hydrolysis was not detected (HPLC) after 5 days at 50ºC which is equivalent to a half-life greater than 1 year at 25ºC.
Test Facility SafePharm (2007b)

Partition Coefficient (n-octanol/water) log Pow = -2.73 at 21.5ºC

Method OECD TG 107 Partition Coefficient (n-octanol/water).

EC Directive 92/69/EEC A.8 Partition Coefficient.

Remarks Flask Method with analysis by HPLC. A preliminary estimate of < -3.4 was obtained from the approximate solubilities in n-octanol (< 22 mg/L) and water (> 54 g/L).

Test Facility SafePharm (2007a)

Surface Tension 71.7 mN/m at 22.0 ± 0.2°C

Method EC Directive 92/69/EEC A.5 Surface Tension.
Interfacial tension balance and procedure based on the ISO 304 ring method.

Remarks Concentration: 0.942 g/L
Not considered to be a surface active material.

Test Facility SafePharm (2008a)

Adsorption/Desorption – screening test log K_{oc} = < 1.25 at 30°C

Method OECD TG 121 Estimation of the Adsorption Coefficient on Soil and on Sewage Sludge Using HPLC.

Remarks The test substance eluted from the column before the reference substance acetanilide.

Test Facility SafePharm (2007b)

Dissociation Constant pKa (est) = -1.63 to -0.71, 1.22 and 2.56

Method Predicted using ACD/I-Lab Web Service (ACD/pKa 8.03)

Remarks Some of the dissociation constants were estimated. There was no pKa within the environmental pH range, as expected. The notified chemical is a water soluble salt that is expected to be ionised in the environment.

Test Facility SafePharm (2007b)

Particle Size

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

Range (μm)	*Mass (%)*
<100	27
<10.0	4.58
<5.5	0.793

Remarks The fraction <100 μm was determined by passing through a sieve. The fraction <10 μm was determined using a cascade impactor, with results averaged from three separate determinations.
Too few particles were of size <10 μm to allow accurate determination of the mass median aerodynamic diameter.

Test Facility SafePharm (2007b)

Solid Flammability Not highly flammable

Method EC Directive 92/69/EEC A.10 Flammability (Solids).

Test Facility SafePharm (2007d)

Autoignition Temperature Does not self-ignite below 400°C

Method EC Directive 92/69/EEC A.16 Relative Self-Ignition Temperature for Solids.

Remarks The notified chemical appeared to have decomposed during the test.

Test Facility SafePharm (2007c)

Explosive Properties Not explosive

Method EC Directive 92/69/EEC A.14 Explosive Properties.

Remarks The notified chemical is not considered to be explosive as it was not thermally sensitive,

shock sensitive or friction sensitive.
Note that during each of the three repeats of the thermal sensitivity test performed with the 2mm orifice plate, and two of the three performed with the 6mm orificie plate, explosions were observed at approximately 80 seconds. In each case the tube was recovered in two pieces. However, as it did not fragment into three or more pieces, according to the test guideline, the notified chemical is not considered to be explosive.

Test Facility SafePharm (2007c)

Oxidizing Properties Not oxidising

Method EC Directive 92/69/EEC A.17 Oxidizing Properties (Solids).
Remarks The molecular structure of the test substance suggests that it is unlikely to have oxidising properties. Expert statement
Test Facility SafePharm (2007c)

APPENDIX B: TOXICOLOGICAL INVESTIGATIONS

B.1. Acute toxicity – oral

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 420 Acute Oral Toxicity – Fixed Dose Method.
EC Directive 2004/73/EC B.1 bis Acute Oral Toxicity – Fixed Dose Procedure.

Species/Strain	Rat/Sprague-Dawley CD (Crl:CD(SD)IGS BR)
Vehicle	Distilled water
Remarks - Method	No significant protocol deviations.

RESULTS

Dose mg/kg bw	*Number and Sex of Animals*	*Mortality*
2000	5F	0

LD50	>2000 mg/kg bw
Remarks - Results	There were no signs of systemic toxicity.

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

TEST FACILITY: Safepharm (2007e)

B.2. Acute toxicity – dermal

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 402 Acute Dermal Toxicity – Limit Test.
EC Directive 92/69/EEC B.3 Acute Toxicity (Dermal) – Limit Test.

Species/Strain	Rat/ Sprague-Dawley CD (Crl:CD(SD)IGS BR)
Vehicle	Moistened with arachis oil BP
Type of dressing	Semi-occlusive.
Remarks - Method	No significant protocol deviations.

RESULTS

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

LD50	>2000 mg/kg bw
Signs of Toxicity - Local	There were no signs of dermal irritation.
Signs of Toxicity - Systemic	There were no signs of systemic toxicity.

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

TEST FACILITY: Safepharm (2008b)

B.3. Irritation – skin

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 404 Acute Dermal Irritation/Corrosion.

	EC Directive 2004/73/EC B.4 Acute Toxicity (Skin Irritation).
Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Vehicle	Moistened with distilled water
Observation Period	72 hr
Type of Dressing	Semi-occlusive.
Remarks - Method	No significant protocol deviations.

RESULTS

Remarks - Results	No evidence of skin irritation was noted in any of the test animals throughout the study. Brown/orange-coloured staining was noted at all treated skin sites at the one hour observation and then in one animal up to the 48 hr observation. The staining did not affect evaluation of the skin reactions.

CONCLUSION	The notified chemical is non-irritating to the skin.
TEST FACILITY	Safepharm (2007f)

B.4. Irritation – eye

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 405 Acute Eye Irritation/Corrosion. EC Directive 2004/73/EC B.5 Acute Toxicity (Eye Irritation).
Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Observation Period	72 hr
Remarks - Method	No significant protocol deviations.

RESULTS

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

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

Remarks - Results	Moderate conjunctival irritation was noted in all treated eyes one hour after treatment. Brown-coloured staining of the fur around all of the treated eyes was noted throughout the study.
CONCLUSION	The notified chemical is slightly irritating to the eye.
TEST FACILITY	Safepharm (2007g)

B.5. Skin sensitisation – mouse local lymph node assay (LLNA)

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 429 Skin Sensitisation: Local Lymph Node Assay. EC Directive 2004/73/EC B.42 Skin Sensitisation (Local Lymph Node Assay).

Species/Strain	Mouse CBA/Ca (CBA/Ca CruBR) strain
Vehicle	1% pluronic L92 in distilled water
Remarks - Method	No significant protocol deviations. The test substance was not suitable for dosing in many of the solvents recommended in the OECD test guideline. The following solvents were found to be suitable for dosing: 25% in DMSO and 1% pluronic L92 in distilled water. The solvent chosen was 1% pluronic L92 in distilled water as this had been used in a previous batch.

RESULTS

Concentration (% w/w)	*Proliferative response (DPM/lymph node)*	*Stimulation Index (Test/Control Ratio)*
Test Substance		
0 (vehicle control)	511.76	1.0
5	317.16	0.62
10	431.44	0.84
25	534.63	1.04
*Positive Control**		
1	Not reported	1.39
10	Not reported	11.33
20	Not reported	19.34

* 2,4-Dinitrobenzenesulfonic acid, sodium salt

CONCLUSION	There was no evidence of induction of a lymphocyte proliferative response indicative of skin sensitisation to the notified chemical.
TEST FACILITY	Safepharm (2007h)

B.6. Repeat dose toxicity

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents. EC Directive 96/54/EC B.7 Repeated Dose (28 Days) Toxicity (Oral).
Species/Strain	Rat/Sprague-Dawley Crl:CD(SD) IGS BR
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	Distilled water
Remarks - Method	Doses were corrected to account for the 94.34% purity of the test substance.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw/day*	*Mortality*
control	5M, 5F	0	0
low dose	5M, 5F	25	0
mid dose 1	5M, 5F	150	0
mid dose 2	5M, 5F	300	0
high dose	5M, 5F	1000	0
control recovery	5M, 5F	0	0
high dose recovery	5M, 5F	1000	0

Mortality and Time to Death

There were no unscheduled deaths.

Clinical Observations

There were isolated observations of red/orange and yellow fur staining during the study. These were not considered to be of toxicological significance. There were no other toxicologically relevant clinical effects observed.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

There were some statistically significant changes in a few blood chemistry and haematology parameters. These were not considered to be of toxicological relevance due to either the absence of dose related responses, the absence of corresponding histopathological changes, or comparison of effects in non-recovery animals (if the effects were only observed in recovery animals).

Yellow/orange coloured urine was observed in animals from the high dose group and one male from the 300 mg/kg/day dose group. This was considered to be a result of administration of the coloured test substance and not to be of toxicological significance.

Effects in Organs

There were some statistically significant changes in spleen and liver weights in high dose animals. These were not considered to be of toxicological relevance due to the absence of associated histopathological effects.

One male treated with 1000 mg/kg/day had orange coloured contents in the non-glandular and glandular region of the stomach at necropsy. This was considered to be a result of administration of the coloured test substance and not to be of toxicological significance.

One male and two females of the high dose group displayed acanthosis/hyperkeratosis of the limiting ridge of the stomach. This effect was not observed in animals of the other dose groups or the control group. However, given that this effect is occasionally observed in control animals, it is unlikely to be a toxicologically significant effect in the present study.

Minimal vacuolation of the superficial transitional epithelial cells of the urinary bladder was observed in males from all treated groups. It was also observed in the control recovery groups, though not the non-recovery control group. In the high dose group, one male exhibited this effect with slight severity. In female animals, this effect was observed (with minimal severity) in only the mid dose groups and the high dose recovery group. This was not considered to be a biologically significant effect as there was not a clear dose response relationship.

There were some other isolated histopathological effects of minor severity in several organs that were not considered to be of toxicological significance.

CONCLUSION

The No Observed Effect Level (NOEL) was established as 1000 mg/kg bw/day in this study, based on the absence of toxicological significant changes in the parameters measured at all dose levels.

TEST FACILITY	Safepharm (2008c)

B.7. 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
Species/Strain	*S. typhimurium*: TA1535, TA1537, TA98, TA100 *E. coli*: WP2uvrA$^-$
Metabolic Activation System	S9 fraction from phenobarbitone/β-naphthoflavone-induced rat liver.
Concentration Range in Main Test	a) With metabolic activation: 50-5000 µg/plate b) Without metabolic activation: 50-5000 µg/plate
Vehicle	Distilled water
Remarks - Method	Formulated concentrations were adjusted to allow for the purity of the test substance. The preliminary test was performed with TA100 and

WP2uvrA$^-$. The frequency of revertant colonies was assessed using a Domino colony counter, except at doses of 5000 µg/plate, due to an intense test material colouration. No significant protocol deviations.

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 for TA1537 > 5000 for other strains	> 5000	Negative
Test 2		150 for TA1537 > 5000 for other strains	> 5000	Negative
Present				
Test 1	> 5000	> 5000	> 5000	Negative
Test 2		> 5000	> 5000	Negative

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

TEST FACILITY Safepharm (2007i)

B.8. 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.
Pre incubation procedure
Test 1 of the study utilised the standard Ames method.
Test 2 of the study incorporated the Prival and Mitchell modification for azo compounds (Prival and Mitchell, 1982).

Species/Strain
Test 1:
S. typhimurium: TA1535, TA1537, TA98, TA100
E. coli: WP2uvrA/pKM101
Test 2:
S. typhimurium: TA98, TA100

Metabolic Activation System
Test 1:
S9-mix (details not given)
Test 2:
Hamster liver homogenate metabolising system. Hamster S9 was not treated with any enzyme inducers.

Concentration Range in Main Test
a) With metabolic activation: 313 - 5000 µg/plate
b) Without metabolic activation: 313 - 5000 µg/plate

Vehicle
Water

Remarks - Method
Study summary was provided. As such, it could not be determined whether all of the appropriate modifications for the Prival and Mitchell method were performed.

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	> 5000	Negative
Present				
Test 1	> 5000	> 5000	> 5000	Negative
Test 2	> 5000	> 5000	> 5000	Negative

Remarks - Results: The Prival-Mitchell modification positive control, Trypan Blue, used in the test induced marked increases in the frequency of the TA98 and TA100 revertant colony with metabolic activation only. Thus, the sensitivity of the assay and the efficacy of the uninduced hamster liver S9-mix was validated.

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

TEST FACILITY: Canon (2006)

B.9. Genotoxicity – in vitro

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 473 In vitro Mammalian Chromosome Aberration Test.
EC Directive 2000/32/EC B.10 Mutagenicity - In vitro Mammalian Chromosome Aberration Test.

Species/Strain: Chinese hamster
Cell Type/Cell Line: Chinese hamster lung (CHL/IU) cells
Metabolic Activation System: Rat liver S9-mix induced by a combination of phenobarbitone/β-naphthoflavone
Vehicle: Eagle's Minimal Essential Medium (MEM)
Remarks - Method: The purity of the test substance was accounted for in the formulations. No significant protocol deviations.

Metabolic Activation	*Test Substance Concentration (μg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	0*, 78.1, 156.25, 312.5, 625*, 1250*, 2500*	6	24
Test 2	0*, 39*, 78.1*, 156.25*, 312.5, 468.75, 625	24	24
Present			
Test 1	0*, 19.5, 39, 78.1, 156.25*, 312.5*, 625*	6	24
Test 2	0*, 78.1, 156.25, 312.5*, 625*, 937.5*, 1250*	6	24

*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	312.5	> 2500	> 2500	Negative
Test 2	> 5000	156.25	312.5	Negative
Present				
Test 1	156.25	> 625	> 625	Negative
Test 2	-	> 1250	> 1250	Negative

Remarks - Results: The test material did not induce any statistically significant increases in the frequency of cells with aberrations or the number of polyploid cells in any of the exposure groups.

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

TEST FACILITY	Safepharm (2007j)

APPENDIX C: ENVIRONMENTAL FATE AND ECOTOXICOLOGICAL INVESTIGATIONS

C.1. Environmental Fate

C.1.1. Ready biodegradability

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 301 C Ready Biodegradability: Modified MITI Test (I).

Inoculum	Activated sludge
Exposure Period	28 days
Auxiliary Solvent	None
Analytical Monitoring	Measurement of biochemical oxygen demand (BOD) with a closed system oxygen measuring apparatus; Determination of dissolved organic carbon (DOC) by a total organic carbon analysis (TOC); Determination of test item by HPLC.
Remarks - Method	On-site sludge sampling was carried out at 10 locations in Japan by collecting return sludge, surface water and surface soil that were in contact with atmosphere. The test was conducted in triplicate at 25±1°C at a concentration of 100 mg/L for the notified chemical and 30 mg/L for activated sludge. Aniline was used as a reference item to confirm that the sludge was sufficiently active. Control tests conducted included a blank control of culture medium only, a control test of the notified chemical without sludge and a control with the reference chemical.

RESULTS

Test substance		*Aniline*	
Day	*% Degradation*	*Day*	*% Degradation*
7	-1.3	7	53
14	-2.0	14	69
21	-2.0	21	69
28	-2.3	28	70

Remarks - Results: All the validity criteria of the test were met.
The percentage of biodegradation at 28 days was 0% by BOD, 1% by DOC and 0% by HPLC.
The notified chemical is not considered readily biodegradable based on the test result.

CONCLUSION: The notified chemical is not considered readily biodegradable.

TEST FACILITY: CERI (2007)

C.1.2. Bioaccumulation

Remarks: Bioaccumulation was not tested. The notified chemical is not expected to bioconcentrate in fish because of its high molecular weight and high water solubility.

C.2. Ecotoxicological Investigations

C.2.1. Acute toxicity to fish

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 203 Fish, Acute Toxicity Test – Semi static, 96h.

	EC Directive 92/69/EEC C.1 Acute Toxicity for Fish – Semi static, 96 h.
Species	*Oncorhynchus mykiss* (Rainbow Trout)
Exposure Period	96 hours
Auxiliary Solvent	None
Water Hardness	Approximately 140 mg $CaCO_3$/L
Analytical Monitoring	Spectrophotometric determination of the test material concentrations. The limit of quantitation was 1.6 mg/L.
Remarks – Method	Following a preliminary range-finding test, a limit test was conducted in duplicate at a nominal concentration of 100 mg/L, at a temperature of 14°C, a pH range of 7.8 - 8.1, and the dissolved oxygen concentration as a percentage of the air saturation value ranged 88 – 102%. Although the percentage air saturation value of some of the test vessels was in excess of 100%, due to the presence of microscopic air bubbles in the media super saturating the diluent, it was considered not to have an impact on the validity and outcome of the test.

RESULTS

Concentration mg/L		*Number of Fish*	*Mortality*				
Nominal	*Actual*		*1 h*	*24 h*	*48 h*	*72 h*	*96 h*
0	< LOQ (< 1.6)	7	0	0	0	0	0
100	97 (0 h), 93 (24 h), 95 (96 h)	7	0	0	0	0	0
100	96 (0 h), 99 (24 h), 97 (96 h)	7	0	0	0	0	0

LC50	>100 mg/L at 96 hours.
NOEC	100 mg/L at 96 hours.
Remarks – Results	The test media was renewed on a daily basis. Results are expressed as nominal concentrations, as measured concentrations were close to nominal throughout the exposure period. No sublethal effects were observed.

CONCLUSION	The notified chemical is not toxic to fish.
TEST FACILITY	SafePharm (2008d)

C.2.2. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 202 Daphnia sp. Acute Immobilisation Test and Reproduction Test – Static, 48 h. EC Directive 92/69/EEC C.2 Acute Toxicity for Daphnia - Static, 48 h.
Species	*Daphnia magna*
Exposure Period	48 hours *[acute study]*
Auxiliary Solvent	None
Water Hardness	250 mg $CaCO_3$/L
Analytical Monitoring	Spectrophotometric determination of the test material concentrations. The limit of quantitation was 1.6 mg/L.
Remarks - Method	Following a preliminary range-finding test, a limit test was conducted whereby 4 replicates of 5 daphnids were exposed to a test concentration of 100mg/L of the notified chemical for 48 hours at 21°C. The dissolved oxygen concentration ranged 8.8 – 8.9 mg/L. A positive control test was conducted using potassium dichromate as the reference material at concentrations of 0.32, 0.56, 1.0, 1.8 and 3.2 mg/L for 48 hours at 20°C.

RESULTS

Concentration mg/L	*Number of D. magna*	*Number Immobilised*

Nominal	*Actual*		*24 h*	*48 h*
0	< LOQ (< 1.6)	20	0	0
100	95 (0 h), 101 (48 h)	20	0	0

EC50	>100 mg/L (nominal) at 48 hours
NOEC	100 mg/L (nominal) at 48 hours
Remarks - Results	The 48-Hour EC50 for the reference material to *Daphnia magna* based on nominal concentrations was 0.47 mg/L with 95% confidence limits of 0.43 – 0.53 mg/L. Analysis of the immobilisation data was by the trimmed Spearman-Karber method. Results are expressed as nominal concentrations, as measured concentrations were close to nominal throughout the exposure period. No immobilisation was reported throughout the 48 hours test at 100 mg/L.
CONCLUSION	The notified chemical is not harmful to daphnids
TEST FACILITY	SafePharm (2008e)

C.2.3. Algal growth inhibition test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 201 Alga, Growth Inhibition Test. EC Directive 92/69/EEC C.3 Algal Inhibition Test.
Species	*Desmodesmus subspicatus*
Exposure Period	72 hours
Concentration Range	Nominal: 0.1, 1, 10, 100 mg/L
Auxiliary Solvent	None
Water Hardness	Not reported
Analytical Monitoring	A Coulter® Multisizer II Particle Counter for the analysis of algal cell concentration.
Remarks - Method	A range finding study only was conducted at nominal concentrations of 0.1, 1.0, 10 and 100 mg/L at 23 - 25°C under static conditions and initial cell density 3 – 5 x 10^3. The test material solutions ranged from pale yellow to dark orange; significant light absorption by the test material at the concentrations of 10 and 100 mg/L was measured at 460 nm.

RESULTS

Yield		*Growth*	
E_yC50 mg/L at 72 h	*NOEC mg/L*	*E_yC50 mg/L at 72 h*	*NOEC mg/L*
> 100	100	> 100	100

Remarks - Results	There was an increase in growth in response to the nominal concentrations 0.1 and 1.0 mg/L (2 and 13%, respectively). There was no inhibition or growth at 10 mg/L and a reduction in growth at 100 mg/L. Significant absorption occurred at 460 nm at the test concentrations 10 and 100 mg/L which is the wavelength that affects chlorophyll a (the principal photoreceptor in algae) and hence photosynthesis which gives rise to growth rate reduction.
CONCLUSION	The notified chemical is not harmful to the growth of green algae, notwithstanding its colour and consequent reduction in light intensity available for photosynthesis.
TEST FACILITY	SafePharm (2008f)

C.2.4. Lemna growth inhibition test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 221 *Lemna* sp. Growth Inhibition Test.
Species	*Lemna minor*
Exposure Period	7 days
Concentration Range	Nominal: 100 mg/L (limit test) Actual: 107 mg/L (0 h), 104 mg/L (2 d), 107 mg/L (5 d) and 106 mg/L (7 d)
Auxiliary Solvent	None
Analytical Monitoring	Spectrophotometric determination of the test material concentrations. The limit of quantitation was 1.6 mg/L.
Remarks – Method	Following a preliminary range-finding test, a limit test was conducted whereby 6 replicates (frond number of 10 each) were exposed to a test concentration of 100 mg/L of the notified chemical for 7 days at 22 - 26°C and the pH ranged 6.6 – 9.4. Test solutions were renewed on days 2 and 5. Observations were made on frond size, appearance, root length and number of colonies present. The endpoints were based on frond numbers and dry weights.

RESULTS

Yield		*Growth*	
E_yC50	*NOEC*	*E_rC50*	*NOEC*
mg/L at 72 h	*mg/L*	*mg/L at 72 h*	*mg/L*
> 100	100	> 100	100

Remarks - Results	The doubling time of the control cultures was 1.68 days. The response to the positive control (3,5-dichlorophenol) was within the normal range. Statistical analysis of the average specific growth rate data was carried out using a Student's t-test incorporating Bartlett's test for homogeneity of variance. Results are expressed as nominal concentrations, as measured concentrations were close to nominal throughout the exposure period.
CONCLUSION	The notified chemical is not harmful to lemna minor.
TEST FACILITY	SafePharm (2008g)

C.2.5. Inhibition of microbial activity

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test. EC Directive 88/302/EEC C.11 Biodegradation: Activated Sludge Respiration Inhibition Test
Inoculum	Activated sludge from the aeration stage of the Severn Trent Water Plc sewage treatment plant which treats predominantly domestic sewage.
Exposure Period	3 hours
Concentration Range	Nominal: 1000 mg/L (limit test)
Remarks – Method	Standard protocol was followed. A range-finding study was used to determine the nominal concentration for the final study. 3,5-Dichlorophenol was used as reference material. The study was considered to be valid.
RESULTS	
IC50	> 1000 mg/L
NOEC	1000 mg/L

Remarks – Results	The IC50 for the reference substance was 9.2 mg/L (confidence intervals 7.3 – 12 mg/L). The confidence intervals were calculated using the method of Litchfield and Wilcoxon.
CONCLUSION	The notified chemical is not harmful to the respiration of sewage sludge microorganisms.
TEST FACILITY	SafePharm (2008h)

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