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NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

FULL PUBLIC REPORT

Wolfwood

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Director
Chemicals Notification and Assessment

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1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT(S)
Firmenich Ltd
73 Kenneth Road
Balgowlah NSW 2093

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

EXEMPT INFORMATION (SECTION 75 OF THE ACT)
Data items and details claimed exempt from publication:
Chemical name
Other names
CAS number
Molecular formula
Structural formula
Molecular weight
Spectral data
Identity and weight percent of toxic or hazardous impurities
Identity and weight percent of non-toxic or non-hazardous impurities
Identity and weight percent of additives and adjuvants

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)
Low Volume Chemical: Permit 32, LVC 32 (1994), Permit 243, LVC 273 (1998), Permit 385, LVC 428 (2001).

NOTIFICATION IN OTHER COUNTRIES
USA (1992), Switzerland (1995), Canada (2003), EU (1992, 1999) (ELINCS 430-460-1).

2. IDENTITY OF CHEMICAL

MARKETING NAME(S)
Wolfwood

METHODS OF DETECTION AND DETERMINATION

ANALYTICAL METHOD	UV/visible spectrophotometry, Infrared (IR) spectroscopy, 1H and 13C NMR spectroscopy, Mass spectrometry

Remarks
TEST FACILITY Firmenich Laboratories, Europe

3. COMPOSITION

DEGREE OF PURITY

Typically 92% (range 89-95%)

HAZARDOUS IMPURITIES/RESIDUAL MONOMERS
All identified impurities were present in the sample of notified chemical used for toxicity testing.

ADDITIVES/ADJUVANTS
None

4. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will not be manufactured in Australia. It will be introduced as a small component (maximum 5%) of compounded fragrances.

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

Year	*1*	*2*	*3*	*4*	*5*
Kilograms	50	75	100	125	150

USE
The notified chemical will be used as a fragrance ingredient in a variety of cosmetic and domestic products. It will be imported in liquid compounded fragrances, which will be reformulated in Australia to produce the final consumer products. In final products, the concentration of the notified chemical will be a maximum of 1% in fine perfumes, and a maximum of 0.025% in other cosmetic products and domestic products.

5. PROCESS AND RELEASE INFORMATION

5.1. Distribution, transport and storage

PORT OF ENTRY
The notified chemical will be imported through Sydney, by wharf or airport, as a small component of fragrance preparations.

IDENTITY OF MANUFACTURER/RECIPIENTS
The fragrance preparations containing the notified chemical will be imported by Firmenich Ltd and will be reformulated locally. The fragrance preparations containing the notified chemical will initially be stored and distributed from the notifier's site. Customers (manufacturers of cosmetics, toiletries and household products) will receive the fragrance preparations for blending into a wide variety of cosmetics, toiletries and household products.

TRANSPORTATION AND PACKAGING
The fragrance preparations containing the notified chemical will be transported by road from the wharf or airport of entry to the Firmenich Ltd warehouse for storage. Firmenich Ltd will forward them directly to the clients. These fragrance preparations will be imported and distributed in tightly closed lacquered drums, typically of 180 kg size, but also 100, 50, 25 10 or 5 kg. Final consumer products will be sold in a variety of small package sizes, typical of consumer-sized containers that will be transported to retail stores for distribution.

5.2. Operation description

The fragrance preparations containing the notified chemical will be reformulated at customer sites. Domestic products will be produced in a continuous mixing process, which will involve a regulated feed of the fragrance mixture into an automated system. Cosmetic products will be produced in a batch process, which may involve open vessels and manual addition of the fragrance preparations containing the notified chemical, but usually batches will be produced by blending all ingredients together in a large mixer, usually closed, followed by automatic filling in containers of various sizes.
The final consumer products will be distributed to retail outlets, displayed and sold to the public.

5.3. Occupational exposure

Number and Category of Workers

Category of Worker	*Number*	*Exposure Duration (Hours/day)*	*Exposure Frequency (Days/year)*
FIRMENICH workers: storage, maintenance and quality control (and, if need arises, repack containers)	**3**	**1**	**1**
Customers' workers: Drum handling/storage/transport Mixer/weighing/formulation Drum cleaning/washing Maintenance Quality assurance Packaging	 **5** **5** **8** **3** **2** **8**	 **2** **4** **2** **2** **½** **4**	 **2** **2** **2** **1** **1** **2**

Exposure Details

Import; Transport to & from Warehouse

The notified chemical will be imported as a component of fragrance preparations. These fragrance preparations will be transported by road to the Firmenich warehouse, and then distributed to clients for reformulation. Transport and warehouse workers will only be exposed to the notified chemical in the event of container breakage and/or accidental spillage.

Formulation of consumer products

Following distribution to clients, import containers of fragrance preparations containing the notified chemical will be opened and re-formulated into consumer products. The major occupational exposure to the notified chemical will be during these processes. Workers at these sites may be exposed to the notified chemical during warehouse, production line, cleaning and sampling or analysis tasks. The main route of exposure is by skin contact; however, inhalation may occur in some circumstances.

The notifier identified two classes of consumer products that will be made using fragrance preparations containing the notified chemical: household cleaning products and cosmetics.

Household cleaning products will be formulated in a continuous mixing process, with a regulated feed of the fragrance mixture into an automated system, and automated packing lines. Cosmetics will be produced in large batches of several thousand kilograms, with mixing vessels that may be open or closed, with exposure a possibility in open systems. However, for large batches employing a number of hazardous components, typically this will necessitate use of closed lines, local exhaust ventilation where vapours or aerosols are produced, and automated packing lines. If open vessels are used for mixing, ventilation will be provided to remove aerosols that may be generated. Where exposure is possible through manual handling, opening and closing drums, and cleaning operations, workers are expected to wear suitable gloves, eye and face protection and protective clothing.

End Use

Workers exposed to end use products may include professional cleaners (household cleaning products) and beauticians (cosmetics). These workers can be expected to use minimal PPE. However, the final concentration of notified chemical in cleaning and cosmetic products (other than fine perfumes) will be less than 0.025%.

5.4. Release

No manufacturing of the notified substance will occur in Australia. Environmental release of the notified substance is unlikely during importation, storage and transportation, and accidental spills, leaks and catastrophic mechanical failure during a transport accident is the most likely reason for environmental release. Established engineering controls (eg. 180 kg sealed and lacquered drum specifications) and established emergency clean-up procedures will limit the impact on the environment of such incidents

RELEASE OF CHEMICAL AT SITE

Release of the notified chemical to the environment during blending of the cosmetic and household products is expected to be minimal due to the relatively small import quantity and the enclosed automated processes used. Potential sources of release include spills, equipment washing, and container residues. The drum size of the fragrance preparation containing the new chemical will determine the amount of environmental release in the event of an accidental spill. The notifier

estimates that up to 0.1% of waste may be generated as a result of spills. No release is anticipated from cleaning of formulation equipment. It is expected that this equipment will be cleaned using water and the aqueous solution reused for new purposes. The average amount of residue in empty containers after removal by vacuum pump is estimated to be < 0.1%. Therefore a total of 0.2% or up to 0.3 kg of waste may be generated each year as a result of formulation activities. Spilled material will be either reclaimed and reused or disposed of by incineration or landfill. Emptied imported containers of perfume compositions containing the notified chemical will either be recycled or disposed of through an approved waste management procedure (eg. metal recycling, drum reconditioning).

RELEASE OF CHEMICAL FROM USE

The majority of the notified chemical will enter the sewerage system after use of the consumer products (cosmetics, toiletries, household products) into which it is incorporated, when cosmetics and toiletries are washed off the hair and skin, and cleaning agents enter the sewer during or after cleaning activities (i.e. up to ~150 kg/y). A widespread and diffuse use and disposal pattern is expected. The notifier estimates that finished product container residues of the notified chemical will vary depending of size, construction material (glass, plastic, metal, paperboard, etc) of the containers, physical state and viscosity of the consumer products and the way the consumers finish their products; however, residues of 0.1-3% of the consumer product may be assumed (i.e. 0.03% or 50 g/yr of the notified chemical based on 1% content). These emptied consumer containers will be disposed of into domestic rubbish and ultimately landfill.

5.5. Disposal

Disposal via incineration or landfill is recommended for wastes generated during the formulation of the products containing the fragrance preparations. The majority of the notified chemical will ultimately be disposed of to sewer after use, with a minor proportion to landfill. The emptied imported drums may potentially be rinsed and re-used, sent to a recycler, or sent to landfill for disposal. Drum rinse water may be reused in additional batches. Following use, emptied consumer product containers are disposed of through domestic garbage disposal and hence will enter landfill or recycling.

5.6. Public exposure

Public exposure to the notified chemical as imported as a component of fragrance compositions could only occur in the event of transport accident or spillage. Public exposure from the reformulation process is unlikely. Public exposure to the notified chemical will occur during day-to-day usage of consumer products (cosmetics, toiletries and household products) containing the notified chemical at a maximum concentration of 1%.

6. PHYSICAL AND CHEMICAL PROPERTIES

Appearance at 20°C and 101.3 kPa Colourless liquid

Melting/Freezing Point 3-30°C (range)

METHOD EC Directive 92/69/EEC A.1 Melting/Freezing Temperature.
TEST FACILITY SafePharm Laboratories Ltd (1998a)

Boiling Point 324.3-337.5°C (range) at 101.3 kPa

METHOD EC Directive 84/449/EEC A.2 Boiling Temperature
TEST FACILITY Toxicol Laboratories Ltd (1992a)

Density 1.03 kg/m^3 at 20°C

METHOD EC Directive 92/69/EEC A.3 Relative Density.
TEST FACILITY SafePharm Laboratories Ltd (1998a)

Vapour Pressure 9.6 x 10^{-6} kPa at 25°C

METHOD EC Directive 92/69/EEC A.4 Vapour Pressure.

Remarks The vapour pressure was measured using a vapour pressure balance system (sensitivity 0.1 μg) with replicate measurements (9 runs) at several temperatures (20-45°C) and linear regression analysis used to calculate the average vapour pressure at 25°C. The test material did not change in appearance during the test. The test substance is slightly volatile (Mensink et al., 1995).

TEST FACILITY SafePharm Laboratories Ltd (1998a)

Water Solubility 31.67 mg/L at 20°C

METHOD EC Directive 84/449/EEC A.6 Water Solubility.

Remarks Flask Method. Only a very brief test report is available. The mass concentration of the test material in solution was determined by gas chromatography (GC). Test substance concentrations at 24, 48 and 72 h were 31.11, 30.45 and 33.44 ppm, respectively. Test solution pH 8.35. The test substance is moderately soluble in water (Mensink et al., 1995).

TEST FACILITY Toxicol Laboratories Ltd (1992a)

Hydrolysis as a Function of pH

METHOD EC Directive 92/69/EEC C.7 Degradation: Abiotic Degradation: Hydrolysis as a Function of pH.

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

Remarks Stock solution (1000 mg/L in methanol) was diluted in replicate by a factor of 100 using 3 buffer solutions of pH 4, 7 and 9. The solutions were shielded from light. Samples were maintained at 50±0.5°C for 5 days prior to analysis by GC after C18 solid phase extraction and elution in methanol. Sample concentrations for pH 4, 7, and 9 solutions were $1.12x10^{-2}$, $1.02x10^{-2}$ and $1.12x10^{-2}$ g/L, respectively, at day 0. Duplicate standards of test material were prepared at a nominal concentration of 100 mg/L. A maximum of 10% hydrolysis occurred over the 5 day test period at 50°C.

TEST FACILITY SafePharm Laboratories Ltd (1998a)

Partition Coefficient (n-octanol/water) log Pow = 4.57 at 20ºC

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

Remarks HPLC Method. Both preliminary (based on the approximate solubilities of the test material in n-octanol and distilled water by visual assessment) and definitive tests were performed. An aliquot (0.0155 g) of test substance was diluted to 100 mL with methanol. In the latter, 6 reference substances of known log Pow were also tested, and dead time was determined by the retention time of thiourea (17 mg/L as supplied). The range finding test gave a log Pow of >3.6.

TEST FACILITY SafePharm Laboratories Ltd (1998a)

Adsorption/Desorption log Koc = 3.14

METHOD OECD (1997) Draft Guideline (July 1997): Estimation of the Adsorption Coefficient (Koc) on Soil and Sewage Sludge using High Performance Liquid Chromatography (HPLC).

Remarks An aliquot (0.0138 g) of test material was diluted to 100 mL with methanol. Dead time was determined by measuring the retention time of formamide (820 mg/L). Solutions of 10 reference standards of known log Koc were also prepared in methanol.

TEST FACILITY SafePharm Laboratories Ltd (1998a)

Dissociation Constant Not determined. There are no acidic or basic groups on the molecule able to dissociate.

Particle Size Test not conducted as the notified chemical is a liquid.

Flash Point 163°C (pressure not reported)

METHOD EC Directive 44/449/EEC A.9 Flash Point.
Remarks The determination was carried out using the Pensky-Martens Closed Tester according to the ASTM-IP method (ASTM D93-77-IP34/80).
TEST FACILITY Toxicol Laboratories Ltd (1992a)

Flammability Limits Test not conducted since experience in use indicate that negative results would be obtained.

Autoignition Temperature 274°C +/- 5 °C

METHOD 92/69/EEC A.15 Auto-Ignition Temperature (Liquids and Gases).
TEST FACILITY SafePharm Laboratories Ltd (1998a)

Explosive Properties Not explosive

METHOD EC Directive 92/69/EEC A.14 Explosive Properties.
Remarks A negative result was obtained for sensitivity to shock and to heat. The test for friction is not applicable to liquids.
TEST FACILITY SafePharm Laboratories Ltd (1998a)

Reactivity

Remarks The notified chemical is expected to be stable under normal environmental conditions. No test of oxidising properties was performed, however the notified chemical has not structural indications of oxidising properties or other unusual reactivity.

ADDITIONAL TESTS

Fat (or n-octanol) Solubility Soluble in all proportions in coconut oil at 37°C

METHOD EC Directive 84/449/EEC A7: Fat Solubility.
Remarks Proportions tested included 1:1, 1:3, 3:1, 1:5, 5:1, 1:9 and 9:1 by volume.
TEST FACILITY Toxicol Laboratories Ltd (1992a)

Surface Tension 55 mN/m at 20.5ºC

METHOD EC Directive 92/69/EEC A.5 Surface Tension.
Remarks ISO 304 Ring Method using a White Electrical Interfacial Tension Balance and platinum ring. Test solution was prepared by dilution of test substance (0.1011 g) in 200 mL of glass double distilled water. After ultrasonication, the supernatant was filtered (prewashed 0.45 μm and 0.2 μm) and filtrate was diluted to 90% of its former concentration with the dilution water. The sample solution was then transferred to the measuring vessel. Surface tension readings were repeated until constant values were obtained. Readings were taken of the force required to detach the ring from the surface of the liquid. Test substance concentration was determined (in duplicate) by GC (mean sample concentration was $3.47x10^{-2}$ g/L). The test substance is a surface active substance (<60 mN/m) at the concentration tested.
TEST FACILITY SafePharm Laboratories Ltd (1998a)

7. TOXICOLOGICAL INVESTIGATIONS

Endpoint	*Assessment Conclusion*
Rat, acute oral	low toxicity LD50 >2000 mg/kg bw
Rat, acute dermal	low toxicity LD50 >2000 mg/kg bw
Rabbit, skin irritation	corrosive
Rabbit, eye irritation	Not performed because corrosive to skin
Guinea pig, skin sensitisation –adjuvant test.	Evidence of sensitisation.
Guinea pig, skin sensitisation – Buehler test	Evidence of sensitisation
Rat, repeat dose oral toxicity – 28 days.	NOAEL 150 mg/kg bw/day
Skin sensitisation-Human repeated insult patch test	No evidence of sensitisation to 1% preparation
Skin sensitisation-Human repeated insult patch test	No evidence of sensitisation to 5% preparation
Genotoxicity – bacterial reverse mutation	non mutagenic
Genotoxicity – in vitro chromosome aberration	non genotoxic

7.1. Acute toxicity – oral

TEST SUBSTANCE Notified chemical

METHOD OECD TG 401 Acute Oral Toxicity– Limit Test.
EC Directive 84/449/EEC B.1 Acute Toxicity (Oral) – Limit Test.
Species/Strain Rat / Crl:CD(SD)Br (VAF plus)
Vehicle Polyethylene glycol (PEG 400)
Remarks - Method Single dose, oral gavage.
Observation period: 14 days.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	5 per sex	2000	0/10

LD50 >2000 mg/kg bw

Signs of Toxicity	Piloerection was observed in all animals but full recovery had occurred within 24 hours of dosing. No effect on bodyweight.
Effects in Organs	At necropsy no abnormalities were found in any animal.
Remarks - Results	

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

TEST FACILITY — Toxicol Laboratories Ltd (1991a)

7.2. Acute toxicity - dermal

TEST SUBSTANCE — Notified chemical

METHOD — OEC D TG 402 Acute Dermal Toxicity – Limit Test.
EC Directive 84/449/EEC B.3 Acute Toxicity (Dermal)

Species/Strain	Rat / Crl:CD(SD)Br (VAF plus)
Vehicle	Used as supplied
Type of dressing	Semi-occlusive.
Remarks - Method	Limit test in which the test article was dermally applied under a semi-occlusive dressing to the clipped backs of rats at a dose level of 2000 mg/kg/bodyweight. After a contact period of 24 hours, the dressings were removed and the treated skin cleansed with water. The animals were observed on the day of dosing and daily thereafter for a further 14 days at the end of which, they were killed and subjected to necropsy.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	5 per sex	2000	0/10

LD50	>2000 mg/kg bw
Signs of Toxicity - Local	Skin irritation at and around the application site was observed in a proportion of animals.
Signs of Toxicity - Systemic	Perinasal staining was noted in most of the males and some females on the day of dosing only. Piloerection, of short duration, was observed in most of the males. No effect on bodyweight.
Effects in Organs	None of the necropsy findings were considered to be treatment related. Abnormal findings consistent with background pathology for this strain of rat.
Remarks - Results	

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

TEST FACILITY — Toxicol Laboratories Ltd (1991b)

7.4. Irritation – skin

TEST SUBSTANCE — Notified chemical

METHOD — OECD 404 Acute Dermal Irritation/Corrosion
EC Directive 84/449/EEC B.4 Acute Toxicity (Skin Irritation).

Species/Strain	Rabbit/New Zealand White
Number of Animals	4 females
Vehicle	Used as supplied
Observation Period	4 hours

Type of Dressing Semi-occlusive.
Remarks - Method

RESULTS

Lesion	*Mean Score* Animal No.*				*Maximum Value*	*Maximum Duration of Any Effect*	*Maximum Value at End of Observation Period*
	1	2	3	4			
Erythema/Eschar	2.0	2.66	2.66	3.5	4	14 days	2
Oedema	2.66	2.33	3.0	2.66	3	7 days	0

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

Remarks - Results

Erythema and oedema were noted on the backs of all animals from one hour after the end of dosing and developed progressively during the following 72 hours. A necrotic lesion, 5 mm in diameter, formed on the back of one rabbit 24 hours after the end of dosing and by the 48 hour assessment the lesion had spread to cover an area 25 x 30 mm and was surrounded by an area of severe erythema. In view of this, the rabbit was killed.
At the 72 hour assessment, moderate to severe erythema and slight or moderate oedema were present on the backs of all 3 remaining animals.
Seven days after dosing skin thickening had occurred at the application site in all 3 rabbits. Oedema was reduced or absent in all 3 animals and erythema had begun to subside in one of them.
14 days after dosing, no sign of skin irritation or skin thickening was apparent in 2 rabbits. Oedema and skin thickening had entirely subsided but a well defined erythema persisted on the back of the 3rd rabbit.
The calculated mean score for erythema or eschar formation was 2.6, and for oedema 2.7.
The notified chemical is severely irritating to the skin of rabbit. In 2/3 of the rabbits, this irritation was reversible within 14 days.

CONCLUSION The notified chemical is corrosive to the skin.

TEST FACILITY Toxicol Laboratories Ltd (1991c)

7.5. Irritation - eye

Test not carried out in view of the result of the skin irritation test. The substance is corrosive to skin and assumed to be irritating to eye.

7.6.1 Skin sensitisation-Magnusson & Kligman test

TEST SUBSTANCE Notified chemical

METHOD OECD 406 Skin Sensitisation – Magnusson and Kligman
EC Directive 84/449/EC B.6 Skin Sensitisation – Maximisation Test – Magnusson and Kligman.

Species/Strain Guinea pig/ Albino, Dunkin-Hartley

PRELIMINARY STUDY Maximum Non-irritating Concentration:
intradermal: 25% in liquid paraffin caused localised response, suitable for the main study. All doses (down to 1%) caused observable irritation effects (erythema and/or oedema).
topical: 100%

MAIN STUDY

Number of Animals Test Group: 20 Control Group: 10

INDUCTION PHASE Induction Concentration:
intradermal: 25% in liquid paraffin
topical: 100%

Signs of Irritation No observations reported after Induction Phase

CHALLENGE PHASE

Topical induction only — Challenge concentration: 100% and 50% in ethanol

Remarks - Method

RESULTS

Animal	*Challenge Concentration (Topical Application)*	*Number of Animals Showing Skin Reactions after: 1st challenge*		*2nd challenge*	
		24 h	*48 h*	*24 h*	*48 h*
Test Group	100%	20/20	20/20	N/A	N/A
Test Group	50%	19/20	12/20	N/A	N/A
Control Group	100%	0/10	0/10	N/A	N/A
Control Group	50%	0/10	0/10	N/A	N/A

Remarks - Results: Following application of undiluted test article, all test animals responded positively to either 50% diluted test article or undiluted test article.

CONCLUSION: There was evidence of reactions indicative of skin sensitisation to the notified chemical under the conditions of the test.

TEST FACILITY: Toxicol Laboratories Ltd (1991d)

7.6.2 Skin sensitisation-Buehler test

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 406 Skin Sensitisation – Buehler test

Species/Strain: Guinea pig/Albino, Dunkin-Hartley

PRELIMINARY STUDY: Not performed. Dose information from previous study (above) used.

MAIN STUDY

Number of Animals: Test Group: 10 Control Group: 10

INDUCTION PHASE: Induction Concentration:
topical: 10% v/v in ethanol
Control group untreated (naïve)

Signs of Irritation: No observations recorded for Induction Phase.

CHALLENGE PHASE

1st challenge: topical: 10% and 5% in ethanol

2nd challenge: topical: 1% and 0.5% in ethanol

Remarks - Method: Further to the result of the previous test (Toxicol, 1992e), this study was performed to determine concentrations of the test article incapable of eliciting a sensitising reaction.

RESULTS

Animal	*Challenge Concentration*	*Number of Animals Showing Skin Reactions after: 1st challenge*		*2nd challenge*	
		24 h	*48 h*	*24 h*	*48 h*
Test Group	10%	10/10	10/10		
	5%	8/10	9/10		
	1%			1/10	2/10
	0.5%			0/10	0/10
Control Group	10%	3/10	4/10		
	5%	2/10	3/10		
	1%			1/10	1/10
	0.5%			0/10	1/10

Remarks - Results: On day 29 all animals, both test and control, were challenged with 2 concentrations of the test article. When challenged with the 10%

concentration, all test animals and 4 control animals exhibited responses. Responses were also exhibited by 9 of the test animals and 3 controls when challenged with a 5% concentration of the test article.
4 days after completion of the initial challenge, a second application was made, in an attempt to determine a no effect level.
When challenged at 1%, 2 test and 2 control animals exhibited responses. Similar response was noted on one animal of the control group when challenged at 0.5%. No response was exhibited by any animal of the test group when challenged at 0.5%. Based on the results obtained from this study it is concluded that the test article produced delayed dermal hypersensitivity in the guinea pig.

Conclusion — There was evidence of reactions indicative of skin sensitisation to the notified chemical under the conditions of the test.

TEST FACILITY — Toxicol Laboratories Ltd (1992b)

7.6.3 Skin sensitisation-Repeated Insult Patch Test (1%)

TEST SUBSTANCE — Notified chemical

METHOD — Human Repeated Insult Patch Test

Subjects — 118 subjects ranging in age from 19 to 74 (males and females) of which 104 completed the study.

Vehicle — Diethyl phthalate

MAIN STUDY

Number of Subjects — Test Group: 118 (104 completed) Control Group: 0

INDUCTION PHASE — The notified chemical was diluted to 1% in a final volume of 0.2 mL. The test substance was applied to the skin of the upper back in occluded patches, for a total of 9 applications. The patches were removed 24 hours after application.

Signs of Irritation — No signs of irritation during 9 induction applications.

CHALLENGE PHASE

1st challenge — After a rest period of 14 days following the ninth application, a challenge patch was applied to the original sites. Sites were evaluated at 48 and 72 hours after application.

Remarks - Method

RESULTS

Subject	*Challenge Concentration*	*Number of Subjects Showing Skin Reactions after: 1st challenge*	
		48 h	*72 h*
Test Group	1%	0/104	0/104

Remarks - Results — There was no evidence of sensitisation in the 104 subjects who completed the study.

CONCLUSION — The notified chemical was non-irritating and non-sensitising under the conditions of the test.

TEST FACILITY — TKL Research, Inc. (1995)

7.6.4 Skin sensitisation-Repeated Insult Patch Test (5%)

TEST SUBSTANCE — Notified chemical

METHOD — Human Repeated Insult Patch Test

Subjects — 124 subjects ranging in age from 19 to 74 (males and females) of which 102 completed the study.

Vehicle	Diethyl phthalate
MAIN STUDY	
Number of Subjects	Test Group: 124 (102 completed) Control Group: 0
INDUCTION PHASE	The notified chemical was diluted to 5% for a final volume of 0.2 mL.
Induction Procedure	The test substance was applied to the skin of the upper back in occluded patches, for a total of 9 applications. The patches were removed 24 hours after application.
Signs of Irritation	No signs of irritation during 9 induction applications.
REST PERIOD	14 days following the ninth application.
CHALLENGE PHASE	
1st challenge	A challenge patch was applied to the original sites. Sites were evaluated at 48 and 72 hours after application.
Remarks - Method	

RESULTS

Subject	*Challenge Concentration*	*Number of Subjects Showing Skin Reactions after: 1st challenge*	
		48 h	*72 h*
Test Group	5%	0/102	0/102

Remarks - Results	There was no evidence of sensitisation in the 102 subjects who completed the study.
CONCLUSION	The notified chemical was non-irritating and non-sensitising under the conditions of the test.
TEST FACILITY	TKL Research, Inc. (1995)

7.7. 28-day repeat dose oral toxicity

TEST SUBSTANCE

METHOD	OECD TG 407 Repeated Dose 28-day Oral Toxicity Study in Rodents.
Species/Strain	Rat/SD Crl:CD BR
Route of Administration	Oral – gavage.
Exposure Information	Total exposure days: 28 days; Dose regimen: 7 days per week; Post-exposure observation period: None
Vehicle	Arachis oil BP
Physical Form	liquid
Remarks - Method	Animals were observed daily. Clinical signs, functional observations, bodyweight, food and water consumption were monitored during the study. Haematology and blood chemistry were evaluated for all animals at the end of the study. All animals were subjected to a gross necropsy examination and histopathological evaluation of selected tissues from 1000 mg/kg/day and control animals was performed.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw/day*	*Mortality*
I (control)	5 per sex	0	0/10
II (low dose)	5 per sex	15	0/10
III (mid dose)	5 per sex	150	0/10
IV (high dose)	5 per sex	1000	0/10

Mortality and Time to Death

There were no deaths during the study

Clinical Observations

Females treated with 1000 mg/kg/day developed piloerection and/or hunched posture from day 15 onwards. One male from this treatment group showed piloerection on day 11 only. No such clinical signs were detected among animals of either sex treated with 150 or 15 mg/kg/day.

Bodyweights and bodyweight gains were unaffected by administration of the test article. Food consumption was not affected by treatment. Animal of either sex treated with 1000 mg/kg/day showed a slight increase in water intake during the final week of treatment when compared with controls. No such effects were evident among animals of either sex treated with 150 or 15 mg/kg/day.

Functional Observations

Detailed behavioural assessments during week 3 revealed hunched posture, piloerection and increased salivation in one 1000 mg/kg/day female. By week 4, all females from this treatment group showed hunched posture during open field observation. Males treated with 1000 mg/kg/day and animals of either sex from the remaining treatment groups showed no treatment related changes in the behavioural parameters assessed.

No treatment related changes were detected in the functional parameters measured and no changes were detected in the sensory scores between treated animals and controls.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

No treatment related changes were detected in the haematological parameters measured.

Male treated with 1000 mg/kg/day showed an increase in calcium levels, total plasma protein and plasma albumin concentration when compared with controls. Females from this treatment group showed increases in plasma aspartate aminotransferase and alanine aminotransferase levels in comparison with controls. Animals of either sex treated with 150 or 15 mg/kg/day showed no treatment related changes in the blood parameters measured.

PATHOLOGY

1. Effects in Organs

Animals of either sex treated with 1000 mg/kg/day showed an elevated liver weight, relative to terminal bodyweight, in comparison with controls. Almost all of the liver weights were outside the normally expected ranges. Females treated with 1000 mg/kg/day showed elevated relative kidney weight. Animals of either sex treated with 150 or 15 mg/kg/day showed no changes in the organ weights measured which could convincingly be associated with test material toxicity. Females treated with 150 mg/kg/day showed a slight increase in relative liver weight compared to controls.

2. Macroscopic Findings

There were no macroscopic abnormalities detected at necropsy following terminal kills.

3. Histopathology

For most endpoints measured, there were no differences in incidence or severity between control and treatment groups. When comparing controls to animals treated with 1000 mg/kg/day, high dose females showed a greater incidence of groups of basophilic tubules, while high dose males showed a slight increase in the incidence (one animal versus no controls) of groups of alveolar macrophages and the appearance of inflammatory cells. However, these changes (as well as others that occurred in treatment and control groups) were considered consistent with the age and strain of the rats, and not of toxicological significance.

Remarks – Results

Repeated oral administration of test material to rats by gavage for a period of 28 consecutive days at dose levels of up to 1000 mg/kg/day resulted in minor toxicologically significant findings at a dose level of 1000 mg/kg/day. There were no toxicologically significant changes in the parameters measured among animals of either sex treated with 150 or 15 mg/kg/day.

CONCLUSION

The No Observed (Adverse) Effect Level (NO(A)EL) was established as 150 mg/kg bw/day in this study, based on increases in liver weight at the top dose of 1000 mg/kg/day and associated changes in blood chemistry parameters.

TEST FACILITY SafePharm Laboratories Ltd (1998d)

7.8. Genotoxicity – bacteria

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 471 Bacterial Reverse Mutation Test. Plate incorporation method
Species/Strain	*S. typhimurium*: TA1535, TA1537, TA98, TA100.
Metabolic Activation System	S9 fraction induced by beta-naphthoflavone and sodium phenobarbitone
Concentration Range in Main Test	a) With metabolic activation: 0.064 to 40 µg/plate. b) Without metabolic activation: 0.064 to 40 µg/plate.
Vehicle	Dimethylsulfoxide
Remarks - Method	

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/plate) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1	40	Not observed at concentrations up to 40 µg/plate	No observations of precipitation reported	Not observed at concentrations up to 40 µg/plate
Test 2		Not observed at concentrations up to 40 µg/plate	No observations of precipitation reported	Not observed at concentrations up to 40 µg/plate
Present				
Test 1	40	Not observed at concentrations up to 40 µg/plate	No observations of precipitation reported	Not observed at concentrations up to 40 µg/plate
Test 2		Not observed at concentrations up to 40 µg/plate	No observations of precipitation reported	Not observed at concentrations up to 40 µg/plate

Remarks - Results	No significant increase in the number of revertant colonies was recorded for any of the bacterial strains with any dose of the test article, either with or without metabolic activation.
CONCLUSION	The notified chemical was not mutagenic to bacteria under the conditions of the test.
TEST FACILITY	Toxicol Laboratories Ltd (1991e)

7.9. Genotoxicity – in vitro

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 473 In vitro Mammalian Chromosome Aberration Test.
Cell Type/Cell Line	Human peripheral blood lymphocytes
Metabolic Activation System	Aroclor 1254 activated S9 fraction
Vehicle	Dimethylsulfoxide
Remarks - Method	Test was performed in duplicate.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Expression Time*	*Selection Time*
Absent				
Test 1	0, 17.03*, 34.06*, 68.13*, 136.25, 272.5, 545, 1090, 2180	4 h	16h	20 h
Test 2	0*, 4.26, 8.52, 17.03*, 34.06*, 51.1, 68.13*	20 h		20 h
Test 2	0*, 17.03, 34.06, 51.1, 68.13*	44 h		44 h
Present				

Test 1	0, 17.03*, 34.06*, 68.13*, 136.25, 272.5, 545, 1090, 2180	4 h	16 h	20 h
Test 2	0*, 4.26, 8.52, 17.03*, 34.06*, 68.13*	4 h	16 h	20 h
Test 2	0*, 17.03, 34.06, 68.13*	4 h	40 h	44 h

*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	136.25	Not observed at concentrations up to 136.25	No observations of precipitation reported	Not observed at concentrations up to 136.25
Test 2		Not observed at concentrations up to 136.25	No observations of precipitation reported	Not observed at concentrations up to 136.25
Present				
Test 1	136.25	Not observed at concentrations up to 136.25	No observations of precipitation reported	Not observed at concentrations up to 136.25
Test 2		Not observed at concentrations up to 136.25	No observations of precipitation reported	Not observed at concentrations up to 136.25

Remarks - Results: No statistically significant increases in the frequency of cells with aberrations or polyploid cells were observed either in the presence or absence of metabolic activation. Appropriate positive controls induced large increases in the number of aberrant cells, indicating that the test system responded appropriately.

CONCLUSION: The notified chemical was not clastogenic to human peripheral blood lymphocytes treated in vitro under the conditions of the test.

TEST FACILITY: SafePharm Laboratories Ltd (1997a)

8. ENVIRONMENT

8.1. Environmental fate

8.1.1. Ready biodegradability

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 301 F Ready Biodegradability: Manometric Respirometry Test.

Inoculum: Mixed biological population from a secondary treated (activated) sludge, Severn Trent Water plc, UK., receiving mainly domestic sewage (pH 7.4; Suspended solids 3.8 g/L).

Exposure Period: 28 days

Auxiliary Solvent: None

Analytical Monitoring: Degradation of the test material was assessed by measurement of daily oxygen consumption.

Remarks - Method: The test material (15.7 mg) was dispersed directly in culture medium (~100 mL) and inoculum (1.8 mL) and ultrasonicated prior to filling to a final volume of 157 mL to give a final test concentration of 100 mg/L. The test chamber was incubated for 28 days (21±1°C) in sealed culture vessels (constantly mixed) at a nominal test substance concentration of

100 mg/L. Controls with inoculum, and standard material (aniline) together with a toxicity control were used for validation purposes. The toxicity control was prepared by dispersion of an amount of test material (9.4 mg) in ~50 mL culture medium with the aid of ultrasonication, prior to addition of an aliquot (9.4 mL) of a 1000 mg aniline/L stock solution, inoculum (1.0 mL) and culture medium to a final volume of 94 mL to give a final test concentration of 100 mg test material/L and 100 mg aniline/L.

RESULTS

Test substance	*ThoD*	*Day 7*		*Day 14*		*Day 28*	
	(mgO2/L)	*BOD (mgO2/L)*	*Degradation (%)**	*BOD (mgO2/L)*	*Degradation (%)*	*BOD (mgO2/L)*	*Degradation (%)*
A	309	185	56	230	70	285	85
B	293	10-15	0-1	15	0	25	1
C	602	120	18	180	27	280	43
Control	---	10-13	---	15	---	22-23	---

A = Aniline + inoculum (100 mg/L); B = Test material + inoculum (100 mg/L); C = Toxicity control + inoculum (test material + aniline, 1000 mg/L). * Degradation (%) = [BODtest – mean BODcontrol]/ThoD] X 100%

Remarks - Results: The biodegradation of the reference substance, aniline was 85% after 28 days, indicating the test conditions were valid.

CONCLUSION: The test material achieved 1% degradation in 28 days and cannot be classed as readily biodegradable under the conditions of the test. The test substance was not toxic to the sewage treatment microbes used in the study.

TEST FACILITY: SafePharm Laboratories (1998i)

8.1.2. Bioaccumulation

No bioaccumulation study was conducted. As the test substance is readily soluble in fat and has an octanol:water partition coefficient (log Pow) of 4.57, a high affinity to lipids is expected and bioaccumulation may potentially occur.

8.2. Ecotoxicological investigations

8.2.1. Acute toxicity to fish

TEST SUBSTANCE: Notified chemical

METHOD: OECD TG 203 Fish, Acute Toxicity Test – flow through

Species	Rainbow trout (*Oncorhynchus mykiss*); juvenile, 4.4 cm length, 1.06 g wt.
Exposure Period	96 h
Auxiliary Solvent	Dimethylformamide (DMF)
Water Hardness	100 mg/L (as CaCO3)
Analytical Monitoring	HPLC with external standard at 0, 24, 48, 72 and 96 h.
Remarks – Method	Range finding and definitive tests were performed. Solvent stock solutions were prepared by dissolving amounts of test material in DMF followed by serial dilution to the nominal test concentrations. Stock solutions were prepared daily. Control and solvent controls were also tested. Test aquaria consisted of 20 L glass containers (no replicates were used). Numbers of mortalities and any sub-lethal effects of exposure in each test and control vessel were determined 3 and 6 hours after the start of exposure and then daily throughout the study until termination after 96 hours. Test temperature was 14±1°C, pH range 6.9-7.1, dissolved oxygen 9.4-9.5

mgO2/L. Photoperiod was 16 light: 8 h dark. Fish were not fed during exposure. Diluent supply was aerated. The LC50 values and associated confidence limits were calculated by the moving average method of Thompson (1947).

RESULTS

Concentration mg/L		*Number of Fish*	*% Mortality*				
Nominal	*Actual**		*3 h*	*24 h*	*48 h*	*72 h*	*96 h*
Control	<LOQ	10	0	0	0	0	0
Solvent control	<LOQ	10	0	0	0	0	0
0.20	0.113-0.360	10	0	0	0	0	0
0.36	0.489-0.970	10	0	0	0	0	0
0.64	0.506-0.864	10	0	0	0	0	0
1.1	0.929-1.39	10	0	0	0	0	0
2.0	1.67-1.96	10	0	0	100	100	100

* LOQ (Limit of Quantitation) = 0.02 mg/L.

LC50 (mean measured)	1.5 mg/L (nominal) at 96 hours (95% CI 1.1-2.0 mg/L)
NOEC (or LOEC)	0.64 mg/L (nominal) at 96 hours
Remarks – Results	The concentration and stability of the stock solution were verified by chemical analysis at 0, 24, 48, 72 and 96 h. Sublethal effects (i.e. loss of equilibrium, swimming at the bottom and/or of tank) were observed at test concentrations ≥1.1 mg/L. Test concentrations of 0.2 and 2.0 mg/L were clear colourless solutions with no precipitation after 24 h.
CONCLUSION	The notified chemical is toxic (i.e. LC50 1-10 mg/L; United Nations, 2003) to rainbow trout under the conditions tested.
TEST FACILITY	SafePharm Laboratories (1998e)

8.2.2. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 202 *Daphnia* sp. Acute Immobilisation Test – static conditions
Species	Cladoceran (*Daphnia magna*; 1st instar <24 hours old)
Exposure Period	48 hours
Auxiliary Solvent	Dimethylformamide (DMF)
Water Hardness	250 mg/L (as CaCO3)
Analytical Monitoring	HPLC with external standard determined at 0 and 48 hours.
Remarks - Method	Range finding and definitive studies were performed. Test material (200 mg) was dissolved in DMF and the volume adjusted to 10 mL to give the 200 mg/10 mL solvent stock solution. An aliquot (500 µl) of this stock solution was dispersed in reconstituted water and the volume adjusted to 5 L to give the 2.0 mg/L test concentration from which dilutions were made to give the test series. Test aquaria consisted of 250 mL beakers containing 250 mL test solution. Mortality was monitored for at 24 and 48 hours. The EC50 values and associated confidence limits were calculated by the moving average method of Thompson (1947). Daphnids were immobilised when unable to swim for ~15 s after gentle agitation. Water temperature: 21°C, dissolved oxygen 8.1-8.4 mg O2/L and pH 7.9 (acceptable).

RESULTS

Concentration mg/L		*Number of D. magna*	*Number and Percent Immobilised*	
Nominal	*Actual (0 and 48 h)*		*24 h*	*48 h*

Control	<LOQ*	20 (2 replicates of 10)	0	0	0	0
Solvent control	<LOQ**	"	0	0	0	0
0.020	0.0225-0.0222	"	0	0	0	0
0.036	0.0378-0.0395	"	0	0	0	0
0.064	0.0650-0.0602	"	0	0	0	0
0.11	0.107-0.113	"	0	0	0	0
0.20	0.198-0.185	"	0	0	0	0
0.36	0.367-0.348	"	0	0	0	0
0.64	0.620-0.624	"	0	0	9	45
1.1	1.18-1.09	"	3	15	16	80
2.0	1.98-1.94	"	13	65	20	100

* LOQ (limit of quantitation) = 0.0021 mg/L. ** Solvent Control contains 10 µL/L of DMF.

LC50	0.74 mg/L (nominal) at 48 hours (95% CI 0.62-0.87 mg/L)
NOEC (or LOEC)	0.36 mg/L at 48 hours (nominal)
Remarks - Results	Test concentrations were stable as determined by HPLC at 0 and 48 hours, and within 93-113% of nominal (acceptable), and consequently the results are based on nominal test concentrations only. Test of solubility indicated that the concentration of 2.0 mg/L was the highest attainable without visual precipitation.
CONCLUSION	The notified chemical is very toxic (i.e. EC50 <1 mg/L; United Nations, 2003) to Daphnia magna under the conditions tested.
TEST FACILITY	SafePharm Laboratories (1998f)

8.2.3. Algal growth inhibition test

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 201 Alga, Growth Inhibition Test.
Species	Green algae (*Scenedesmus subspicatus*; freshwater unicellular)
Exposure Period	72 hours
Concentration Range	
Nominal	2.0 mg/L
Actual	1.92 to 2.10 mg/L
Auxiliary Solvent	Dimethylformamide (DMF)
Water Hardness	Not measured
Analytical Monitoring	HPLC with external standard at 0 and 72 h
Remarks - Method	Range finding and definitive tests were performed. Precipitation of the test material was observed in the range finding tests at concentrations >2.0 mg/L, therefore this was the highest concentration used in the definitive test. Test material (200 mg) was dispersed in DMF and the volume adjusted to 10 mL to give a 200 mg/10 mL solvent stock solution. From this, a serial dilution was made to give a 20 mg/10 mL solvent stock solution. A solvent control (DMF 100 µL/L) was also tested. Aliquots (20 µL) of each stock solution were separately mixed with algal suspension to give the required test concentration of 2.0 mg/L. Test containers consisted of 250 mL flasks (stoppered) which were incubated at 24±1°C under continuous light (7000 lux) and mixing (100 rpm for 72 h). 0 and 72 h cell densities were ~104 and ~105 cells/mL, respectively, in the controls and treatments. Samples of test solutions were collected at 0, 24, 48 and 72 h for cell counting using Coulter® Multisizer II Particle Counter. Statistically significant differences between test and control groups were determined using Students t-tests.
RESULTS	

Biomass	*Growth*

EbC50 *mg/L at 72 h*	*NOEC* *mg/L*	*ErC50* *mg/L at 0-72 h*	*NOEC* *mg/L*
≥2.0	2.0*	≥2.0	2.0*

* Highest concentration tested

Remarks - Results	No abnormalities were detected in any of the control or test cultures. Analysis of test solutions at 0 and 72 hours showed the measured test concentrations to be near nominal (96-103%) indicating it was stable. Test solution pH increased from 7.4-7.5 (0 h) to 10.2-10.3 (72 h) in controls and treatments, which is not unusual due to CO2 formation.
CONCLUSION	No definitive conclusion can be made as the test substance was not toxic (no growth inhibition) to algae at the single concentration tested of 2.0 mg/L.
TEST FACILITY	SafePharm Laboratories (1998g)

8.2.4. Inhibition of microbial activity

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test.
Inoculum	Mixed biological population from a secondary treated (activated) sludge, Severn Trent Water plc, UK., receiving mainly domestic sewage (pH 7.4; Suspended solids 3.8 g/L).
Exposure Period	3 hours
Concentration Range Nominal	10, 18, 32, 56 and 100 mg/L
Remarks – Method	Range-finding (1.0, 10, 100 and 1000 mg/L) and definitive tests were performed. Test material was aerated for 3 h at 21°C in the presence of activated sewage sludge and synthetic sewage. The rate of respiration was determined at 30 minutes and 3 hours. Amounts of test material (28 and 50 mg) was dispersed in dimethylformamide (DMF) and made up to 250 mL with water. After ultrasonication (30 mins), synthetic sewage sludge (16 mL), activated sewage sludge (200 mL) and water (activated carbon filtered; hardness 100 mg/L as CaCO3) were added to a final volume of 500 mL to give the test material concentration of 56 and 100 mg/L. The solvent control (300 mL) consisted of sewage sludge (16 mL), water, 200 mL inoculum, and DMF 100 µL/L of DMF. Two stock solutions of a reference material (3,5-dichlorophenol) were prepared (50 and 160 mg/L) by direct addition in water and ultrasonication. Aliquots (10 and 100 mL) of the 160 mg/L stock solution were dispersed with DMF (50 µL), activated sewage sludge (200 mL), synthetic sewage (16 mL) and water to give final volume of 500 mL and concentrations of 3.2 and 32 mg/L. Tests were conducted in replicate. After 30 minutes contact time, an aliquot was removed from test containers and poured into a measuring vessel (250 mL darkened BOD bottle) and the rate of respiration was monitored with a DO meter. The rate of respiration was monitored over the linear portion of the oxygen consumption trace for ~10 minutes (between 8.2 mg O2/L and 1.7 mg O2/L). The procedure was repeated after 3 h contact time.

RESULTS

Concentration (mg/L)	Consumption Rate (mg O2/L/min)	% Inhibition
Solvent control	0.38-0.45	0
10	0.42	+1
18	0.34	18
32	0.32	23

56	0.21	49
100	0.13	69
3,5-DCP		
3.2	0.39	6
10	0.14	66
32	0.06	86

EC50	64 mg/L at 3 hours
NOEC	10 mg/L at 3 hours
Remarks – Results	The two solvent control replicates were within 15% of each other (i.e. ±1%), and the EC50 of the reference toxicant was within the range of 5-30 mg/L (i.e. 10 mg/L), thereby validating the test conditions.
CONCLUSION	The test material inhibited the respiration of sewage sludge microbes by various rates after 3 hours exposure to various concentrations of the test material (eg. 69% inhibition at 100 mg/L after 3 h contact time).
TEST FACILITY	SafePharm Laboratories (1998h)

9. RISK ASSESSMENT

9.1. Environment

9.1.1. Environment – exposure assessment

ow volatility (~10^{-6} kPa) and loss to the atmosphere is unlikely to be significant from spills/leaks, sewers and aquatic environment. It is not readily biodegradable (1% biodegradation after 28 days in a manometric respirometry test). It is moderately soluble in water (31.67 mg/L) and has a hydrolysis half-life of greater than 1 year at pH 4, 7 and 9. It has a log Pow of 4.57 and a log Koc of 3.14 indicating that it has the potential to bioaccumulate and the ability to bind tightly to organic matter in soil.

Following its widespread use in Australia, the notified chemical will eventually be released into the sewerage system through washing or cleaning activities. As a worst case, if all of the notified substance entered the sewerage system (up to $1.5x10^{11}$ µg/y), a sewage concentration of 0.1 µg/L may be calculated. This assumes an Australian population of 20.1 million people generates 200 L/person/day (i.e. $1.467x10^{12}$ L/y) and no attenuation within the sewerage system. Assuming dilution factors for freshwater and marine environments of 1 and 10, respectively, PEC(freshwater) and PEC(marine) of 0.1 µg/L and 0.01 µg/L may be calculated. A biosolids concentration of <0.1 µg/kg has been estimated, which assumes Australian production of 100 kg/ML of effluent treated. Attenuation of the notified substance within the sewerage system by partitioning to sludge is expected based on its affinity to organic matter. The notifier has indicated that by using the SimpleTreat Model (European Commission, 2003) and assuming a Henry's constant of 0.06 Pa/m3/mole based on measured vapour pressure and water solubility, a Log Pow 4.57 and being not biodegradable, ~14.5% of the chemical may potentially partition to sludge (potential sewage concentration of 0.09 µg/L). However, reanalysis indicates a higher proportion to sludge (21%) based on this procedure and log10 of Henry's Law Constant ($6.62x10^{-2}$ Pa m^3/mole) of -1.18. Thus, sewer attenuation will further reduce the risk to the environment from the potential presence of the notified substance in effluent discharged.

The substance is readily soluble in fat and has an octanol:water partition coefficient (log Pow) of 4.57 and has a high affinity to lipids. However, with the relatively low volume imported and diffuse release to sewer Australia wide bioaccumulate is unlikely.

9.1.2. Environment – effects assessment

Aquatic ecotoxicity data were available for 4 taxonomic groups (fish, invertebrates, algae and sewage microbes). The results of the ecotoxicological data indicate that the notified chemical is toxic to very toxic to aquatic life (United Nations, 2003). The most sensitive species of those tested are daphnids (acute EC50 0.74 mg/L and NOEC 0.36 mg/L). A predicted no effect concentration (PNEC) of 0.0074 mg/L (7.4 µg/L) has been derived by dividing the lowest available EC50 by an assessment (uncertainty) factor of 100 to account for intra and inter species sensitivity to the notified chemical and potential chronic effects. In the absence of marine toxicity data, the PNECfreshwater is tentatively extrapolated to the marine environment, an approach supported by a preliminary review of comparative data by ECETOC (2003). The NOEC for activated sewage sludge microbes is 10 mg/L at 3 hours; however, such concentrations are unlikely in the sewerage system.

9.1.3. Environment – risk characterisation

An indication of risk can be made by comparison between the PEC and the PNEC using a risk quotient (RQ) approach. RQ values for marine and freshwaters received treated sewage effluent of

0.01 (i.e. 0.1 µg/L ÷ 7.4 µg/L) and 0.001 (i.e. 0.01 µg/L ÷ 7.4 µg/L), respectively, may be calculated. As in both cases the RQ is <<1, taking into account the very worst case natures of the PEC calculations, the risk from use of the notified chemical is expected to be low. Sewerage system attenuation of the notified chemical is expected to reduce this risk further. The notified substance is not expected to be mobile within a landfill, and is expected to degrade over time.

9.2. Human health

9.2.1. Occupational health and safety – exposure assessment

Transport & Storage

Occupational exposure to the notified chemical during transport and storage of fragrance preparations containing the notified chemical is only likely in the event of accidental container breakage and/or spillage. Exposure in these circumstances is expected to be infrequent and acute, and can be limited by use of gloves, goggles, masks and protective clothing during clean-up operations.

Formulation

During reformulation of fragrance preparations containing the notified chemical into cosmetics and domestic cleaning products, dermal exposure is the most likely route. Ocular exposure may occur due to accidental splashes. Exposure may occur when workers open the drums containing imported notified chemical at up to 5%, when weighing and transferring the imported fragrance preparations into mixing vessels, during blending operations and when cleaning up spills and equipment. Blending operations can be in open or closed systems, however, the process is often automated and local exhaust ventilation is usually employed.

Exposure to the notified chemical during filling of consumer product containers is expected to be minimal, as the filling of consumer containers is typically automated.

Dermal and inhalation exposure during formulation was estimated using the EASE model (HSE, 1994). Assuming non-dispersive use and intermittent direct handling, the estimated dermal exposure during formulation is 0.1-1 mg/cm^2/day of fragrance preparations containing up to 5% of the notified chemical. This equates to 0.005-0.05 mg/cm^2/day of the notified chemical. Absorption of the notified chemical may be significant, as the substance has a high Log P_{ow} and fat solubility so ready diffusion across membranes would be expected. Therefore, for a 70 kg worker with surface area for hands at 820 cm^2 and forearms at 1140 cm^2, and assuming 100% absorption, systemic exposure is estimated to be 0.14-1.4 mg/kg bw/day of the notified chemical. This exposure would be substantially reduced by the use of protective clothing and gloves.

The estimated atmospheric concentration of notified chemical during formulation is 0 – 0.9 mg/m^3 using the EASE model (HSE, 1994). Therefore for a 70 kg worker, assuming an inhalation rate of 1.3 m^3/hour, a 4 hour exposure time and 100% bioavailability, inhalation exposure is estimated to be 0 – 0.067 mg/kg bw/day. Exposure to the notified chemical would be reduced by the use of local exhaust ventilation and/or personal respiratory equipment.

End Use

Occupational exposure to end use consumer products may occur, for example, with professional cleaners using cleaning products, or beauticians using cosmetic products. These workers are less likely to use extensive PPE; however, the concentration of notified chemical in end use products will be less than 0.025% (except for fine fragrances, which have up to 1%).

Using the EASE model, and assuming wide dispersive use with extensive, direct handling, estimated dermal exposure to end use products is 5-15 mg/cm^2/day of end use products. This equates to 0.001-0.004 mg/cm^2/day of notified chemical at 0.025% in most end use products. (The exception is fine fragrances, which contain up to 1% of notified chemical, and therefore would expose workers to 0.05-0.15 mg/cm^2/day of notified chemical. However, fine fragrances are not likely to be used occupationally.) For a 70kg worker with 1960 cm^2 surface area and assuming 100% absorption (as above), systemic exposure is therefore estimated to be 0.035-0.105 mg/kg bw/day of the notified chemical for cleaning products and cosmetics other than fine fragrances.

9.2.2. Public health – exposure assessment

It is expected that during import, transport, storage, reformulation of fragrance compositions containing the notified chemical, exposure of the general public will be limited, except in the event of an accidental spill.

Consumer products containing the notified chemical (cosmetics, toiletries, household cleaning products) will be sold in the public domain, consequently there is the potential for widespread public exposure. Exposure will be principally via dermal route. Exposure to the notified chemical is considered minimal given the small amount of notified chemical in the final consumer products (maximum 0.025% other than fine fragrances, which have maximum 1%).

9.2.3. Human health - effects assessment

The notified chemical is of low acute oral and dermal toxicity in rats. Acute inhalation toxicity data were not provided. The notified chemical has low volatility and is not expected to cause significant adverse effects by inhalation.

The notified chemical is not mutagenic in a bacteriological test and not clastogenic to human lymphocytes.

The neat notified chemical is corrosive to rabbit skin, and sensitising to guinea pigs in both adjuvant and non-adjuvant studies. However, no skin sensitisation responses were seen in over 100 human volunteers exposed repeatedly to a 5% formulation over 9 weeks. Eye irritation was not assessed, because in view of the result of the skin irritation test, the chemical was assumed to be irritating to eyes.

In rats, a 4-week repeat dose oral toxicity study showed the NOAEL to be 150 mg/kg/day. This study showed minor treatment related effects, which suggest absorption from the gastro-intestinal tract. The substance has a high Log P_{ow} and fat solubility so ready diffusion across membranes, and hence adsorption, would be expected. It is also moderately water soluble so absorption would probably not be reduced due to deposition at the dosage site and there are no ionisable groups in the parent compound, so absorption will not be pH dependent. No evidence of systemic effects was seen in an acute dermal toxicity study but this does not necessarily indicate absence of absorption, in view of the low toxicity of the substance. Indeed, the lipophilicity of the substance suggests that absorption through the skin might be possible.

Based on the available data, the notified chemical is classified as a hazardous substance and is assigned R34 (causes burns) and R43 (may cause sensitisation by skin contact) in accordance with the NOHSC Approved Criteria for Classifying Hazardous Substances (NOHSC, 2002).

9.2.4. Occupational health and safety – risk characterisation

The notified chemical has been used in Australia since 1994 under a Low Volume Permit. No incident of work-related injury has been reported.

The notified chemical is of low acute toxicity (LD50 >2000 mg/kg for oral or dermal routes). Therefore the risk of acute toxic effects in workers is low. However, the notified chemical is corrosive. If there are accidental spills during transport or storage of the imported fragrance oil containing 5% notified chemical, workers will need skin and eye protection, including gloves, protective clothing and safety glasses or goggles. Given the possible dermal exposure to fragrance oil during formulation of end use products, particularly in open mixing processes, there is a risk of skin and eye irritant effects in workers. Therefore, workers will need protective clothing, gloves and safety glasses when opening the drums, transferring the fragrance oil into mixing vessels and cleaning up spills and equipment.

During formulation, chronic dermal exposure to the notified chemical was estimated to be 0.14–1.4 mg/kg bw/day. The margin of exposure (MOE) for chronic toxicity is based on a NOAEL of 150 mg/kg bw/day. MOE greater than or equal to 100 are considered acceptable to account for intra- and inter-species differences. For dermal exposure, the MOE is calculated as 107-1071 during formulation. Therefore, the risk of chronic systemic toxicity using modelled worker data is acceptable for formulation workers handling fragrance preparations containing up to 5% notified chemical. Occupational risk due to dermal exposure can be further limited by the use of PPE specified in the MSDS.

Chronic inhalation exposure during formulation was estimated to be 0-0.067 mg/kg bw/day. Based on a NOAEL of 150 mg/kg bw/day, the MOE is calculated to be greater than 2200. Therefore the risk to formulation workers is acceptable. Occupational risk due to inhalation exposure can be further limited by the use of local exhaust ventilation.

Dermal exposure to end use products containing up to 0.025% notified chemical is estimated to be 0.035-0.105 mg/kg bw/day. Using the same toxicity data (NOAEL of 150 mg/kg bw/day),

the MOE is calculated as 1429-4286. Therefore the risk to workers handling end use products in the absence of PPE is acceptable.

9.2.5. Public health – risk characterisation

It is expected that public exposure to compounded fragrances containing up to 5% notified chemical for industrial use will be minimal except in the rare event of an accidental spill. There will be public exposure to the notified chemical from dermal, inhalation, oral and ocular exposure to cosmetics, toiletries, and household cleaning products containing up to 0.025% of the notified chemical. Consequently the public risk from exposure to the notified chemical through all phases of its life cycle is considered to be low.

10. CONCLUSIONS – ASSESSMENT LEVEL OF CONCERN FOR THE ENVIRONMENT AND HUMANS

10.1. Hazard classification

Based on the available data the notified chemical is classified as hazardous under the NOHSC Approved Criteria for Classifying Hazardous Substances. The classification and labelling details are:

R34 Causes burns

R43 May cause sensitisation by skin contact

The notified chemical is classified as dangerous for the environment according to the EU criteria with the following risk phrase:

R50/53 Very toxic to aquatic organisms, may cause long-term adverse effects in the aquatic environment.

The notified chemical is classified as a Class 9 Dangerous Good on the basis of its toxicity to aquatic organisms.

As a comparison only, the classification of the notified chemical using the Globally Harmonised System for the Classification and Labelling of Chemicals (GHS) (United Nations, 2003) is presented below. This system is not mandated in Australia and carries no legal status but is presented for information purposes.

	Hazard category	*Hazard statement*
Skin corrosion/irritation	1	Causes severe skin burns and eye damage
Serious eye damage/eye irritation	1	Causes serious eye damage
Skin sensitiser	1	May cause allergic skin reaction
Chronic hazards to the aquatic environment	1	Very toxic to aquatic life with long lasting effects

10.2. Environmental risk assessment

The chemical is not considered to pose a risk to the environment based on its reported use pattern and PEC/PNEC ratio <1.

10.3. Human health risk assessment

10.3.1. Occupational health and safety

There is Low Concern to occupational health and safety under the conditions of the occupational settings described.

10.3.2. Public health

There is No Significant Concern to public health when used as an ingredient in consumer products as described in the notification.

11. MATERIAL SAFETY DATA SHEET

11.1. Material Safety Data Sheet

The MSDS of the notified chemical and fragrance preparations containing the notified chemical provided by the notifier was in accordance with the NOHSC *National Code of Practice for the Preparation of Material Safety Data Sheets* (NOHSC, 2003). It is published here as a matter of public record. The accuracy of the information on the MSDS remains the responsibility of the applicant.

11.2. Label

The label for the notified chemical and fragrance preparations containing the notified chemical) provided by the notifier was in accordance with the NOHSC *National Code of Practice for the Labelling of Workplace Substances* (NOHSC, 1994). The accuracy of the information on the label remains the responsibility of the applicant.

12. RECOMMENDATIONS

REGULATORY CONTROLS

Hazard Classification and Labelling

- The NOHSC Chemicals Standards Sub-committee should consider the following hazard classification for the notified chemical:
  - R34 Causes burns
  - R43 May cause sensitisation by skin contact
  - S24 Avoid contact with skin
  - S26 In case of contact with eyes, rinse immediately with plenty of water and seek medical advice
  - S36/37/39 Wear suitable protective clothing, gloves and eye/face protection
  - S45 In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible)

- Use the following risk phrases for products/mixtures containing the notified chemical:
  - 1-5%: R43 May cause sensitisation by skin contact
  - 5-10%:
    - R43 May cause sensitisation by skin contact
    - R36/38 Irritating to eyes and skin
  - >= 10%:
    - R43 May cause sensitisation by skin contact
    - R34 Causes burns

- The notified chemical should be classified under the ADR code: Class 9 – Miscellaneous Dangerous Goods and Articles

CONTROL MEASURES

Occupational Health and Safety

- Employers should implement the following engineering controls to minimise occupational exposure to the notified chemical:
  - Closed system during mixing and blending of the ingredients with fragrance preparations containing the notified chemical.
  - Local exhaust ventilation if the mixing vessel is open

- Employers should implement the following safe work practices to minimise occupational exposure during handling of the notified chemical:
  - Prevent splashes and spills.

- Employers should ensure that the following personal protective equipment is used by workers to minimise occupational exposure to the notified chemical during formulation of the fragrance preparations containing it with consumer products:

– Chemical resistant gloves, protective overalls and goggles/faceshield

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 NOHSC *Approved Criteria for Classifying Hazardous Substances*, 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 by incineration or landfill.

Emergency procedures

- Spills/release of the notified chemical should be contained as described in the MSDS (i.e. by sand or inert powder) and the material disposed of in accordance with Government regulations.

12.1. Secondary notification

The Director of Chemicals Notification and Assessment must be notified in writing within 28 days by the notifier, other importer or manufacturer:

(1) Under subsection 64(1) of the Act; if
- the importation volume exceeds one tonne per annum notified chemical; or
- The notified chemical is itself manufactured locally or imported

or

(2) Under subsection 64(2) of the Act:
- if any of the circumstances listed in the subsection arise.

The Director will then decide whether secondary notification is required.

No additional secondary notification conditions are stipulated.

Should import levels rise above 1 tonne per annum, a chronic daphnia study should be submitted for assessment.

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