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NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

FULL PUBLIC REPORT

2,6-di(2-hydroxyethyl)aminotoluene

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Director
Chemicals Notification and Assessment

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FULL PUBLIC REPORT

2,6-di(2-hydroxyethyl)aminotoluene

1. APPLICANT DETAILS

APPLICANT(S)
Schwarzkopf Pty Ltd (ACN No.: 000 076 782)
20 Rodborough Road
FRENCHS FOREST NSW 2086

2. IDENTITY OF CHEMICAL

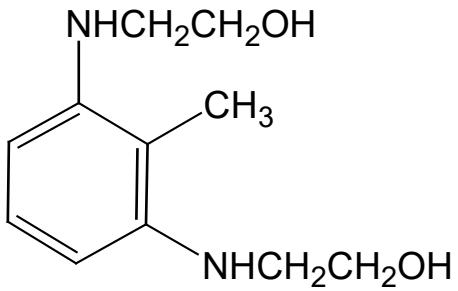
CHEMICAL NAME
Ethanol, 2,2'-[(2-methyl-1,3-phenylene)diimino]bis-

OTHER NAME(S)
2,6-di(2-hydroxyethyl)aminotoluene (INCI name)
1-methyl-2,6-bis(2-hydroxyethylamine)benzene

CAS NUMBER
149330-25-6

MOLECULAR FORMULA
$C_{11}H_{18}N_2O_2$

STRUCTURAL FORMULA



MOLECULAR WEIGHT
210.3

SPECTRAL DATA

METHOD	Infrared spectroscopy.
Remarks	Major absorbance peaks (cm^{-1}): 3400, 3350, 2930, 2850, 1600, 1460, 1400, 1250, 1150, 1060, 760.

METHODS OF DETECTION AND DETERMINATION

ANALYTICAL METHOD	Infrared spectroscopy.

3. COMPOSITION

DEGREE OF PURITY
99.5%

HAZARDOUS IMPURITIES/RESIDUAL MONOMERS
None.

NON HAZARDOUS IMPURITIES/RESIDUAL MONOMERS (>1% by weight)
None.

ADDITIVES/ADJUVANTS
None.

Typical imported formulations contain a number of components as indicated on the submitted MSDS.

4. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will be imported as a 1% component of a hair dye preparation in a 60 mL aluminium tube inside a cardboard carton.

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

Year	*1*	*2*	*3*	*4*	*5*
Tonnes	0.1	0.1	0.1	0.1	0.1

USE
A component of hair dye preparations.

5. PROCESS AND RELEASE INFORMATION

5.1. Distribution, Transport and Storage

PORT OF ENTRY
Sydney.

IDENTITY OF MANUFACTURER/RECIPIENTS
Notifier.

TRANSPORTATION AND PACKAGING
Products containing the notified chemical will be imported as a 1% (w/w) component of hair dye products contained in 60 mL aluminium tubes inside cardboard cartons.

5.2. Operation Description

The notified chemical will be transported from the dockside to the notifier's warehouse and then to hair salons and retail outlets. Hair salon workers unpack boxes containing the imported product and place them on shelves for use in the salon and for sale to the public. Hairdressers manually mix the cream hair dye with a cream developer in a small plastic container and apply the mixture to a customer's hair with a dedicated brush. The hair is then combed to evenly distribute the product through the hair and rinsed with water after a 20 minute development period.

5.3. Occupational exposure

Number and Category of Workers

Category of Worker	*Number*	*Exposure Duration*	*Exposure Frequency*
Waterside workers	1 – 2	1 – 2 hours per day	10 – 15 days per year
Transport workers	1 – 2	1 – 2 hours per day	10 – 15 days per year
Warehouse workers	2 – 4	1 – 2 hours per day	10 – 15 days per year
Hair salon and retail outlet workers	> 1000	1 hour per day	200 – 240 days per year

Exposure Details

Transport and storage workers should only come into contact with the notifier chemical in the event of an accidental breach of the packaging.

Hair salon workers can potentially come in contact with the notified chemical during mixing and application of the hair dye to customers' hair and while rinsing it from the hair. Exposure is controlled by the use of impervious gloves.

5.4. Release

RELEASE OF CHEMICAL AT SITE

The notified chemical will be imported in a finished hair dye product at a concentration of 1%. There will be no reformulation or repackaging of the product. Release of the chemical during transport or storage at the importer's warehouse is not expected, except in the event of a transport accident. The small size of the containers (60 mL) would limit accidental releases, should there be an accident and the containers rupture.

RELEASE OF CHEMICAL FROM USE

The product containing the notified chemical will be used in hair salons by professional hairdressers to colour hair. The product is applied to the hair, left to develop for 20 minutes, and then washed off. As such, almost all (95%) of the notified chemical will be released into the sewer when washed from the hair during rinsing. A small amount (5%) may remain as residues in empty containers.

5.5. Disposal

Waste from hair salons is expected to be disposed of via domestic garbage collection. Residues in empty containers are expected to be disposed of in landfill with the used containers.

5.6. Public exposure

The public may come in contact with the notified chemical in the imported product during application and rinsing off for up to 1 hour, 5–6 times a year. The public may potentially come in contact with the product in the event of a transport accident.

6. PHYSICAL AND CHEMICAL PROPERTIES

There are limited physico-chemical data on the notified chemical. However, it appears to be thermally stable up to the melting point.

Appearance at 20ºC and 101.3 kPa	Light brown-grey powder.
Melting Point	118-121ºC
Boiling Point	Not determined.
Density	Not determined.
Vapour Pressure	Not determined.

Remarks The vapour pressure of a range of toluenediamines is less than 10 kPa at temperatures above 150ºC.

Water Solubility 27 g/L at 20°C

Remarks ACD laboratory software was used to calculate a water solubility of 1,089 g/L for the neutral molecule. The water solubility reported by Marquardt (1994) in the mammalian toxicity report for HC Violet AS is 27 g/L, but no details of the test were provided.

Hydrolysis as a Function of pH Not determined

Partition Coefficient (n-octanol/water) log Pow at 22.5 - 23.2°C = 0.031

METHOD EC Directive 92/69/EEC A.8 Partition Coefficient (shake-flask method)

Remarks Following a preliminary test, a storage solution was prepared by adding 224.7 mg of test substance made up to 100 mL with 1-octanol (saturated with water), equating to a concentration of 2328.86 mg/L. The solution was treated in an ultrasonic bath for 10 minutes prior to filtering through a membrane filter. Two replicates of test solutions containing 10, 5, or 2.5 mL storage solution and 0, 5, and 7.5, mL octanol and 10 mL water (pH = 7.8) were prepared. The phases were mixed by inversion rotation (40 RPM) for 3 hours at a temperature of 22.5 - 23.2°C. Samples were left to stand for 2 hours to separate phases. The concentrations of the test substance in the aqueous and octanol phases were analysed by HPLC. Partition coefficients (Pow) were determined to be between 0.986 and 1.1208, with a mean of 1.075. The results indicate the notified chemical has a poor affinity for lipids.

TEST FACILITY Casella AG (1993)

Adsorption/Desorption Not determined

An adsorption/desorption coefficient was calculated using the software package KOWWIN. The log Koc was determined to be 1.4 for the neutral molecule. This value indicates a very high mobility in soils (McCall et al. 1980). The protonated substance would be expected to sorb strongly to soils and sediments due to the positive charge (Nabholz, 1993)

Dissociation Constant Not determined

Remarks The notified chemical has two protonated amines expected to show typical acidity. Based on the pK_b (pK_b = 5.12) of N-ethyl aniline, the notified chemical is expected to be predominantly in the protonated form in the environmental pH range, particularly below pH 8 (CRC Handbook of Chemistry & Physics, 1977).

Particle Size Not determined.

Remarks Not required as the notified chemical is imported at 1% (w/w) in a cream.

Flash Point Not determined.

Flammability Limits Not determined.

Autoignition Temperature Not determined.

Explosive Properties Not determined.

Remarks Not expected to be explosive based on structure.

Reactivity Not determined.

Remarks Expected to be stable under normal environmental conditions.

7. TOXICOLOGICAL INVESTIGATIONS

A summary of a number of toxicological studies was available for evaluation. In addition, as the notified chemical belongs to a class which includes known carcinogens, the bacterial mutagenicity test was positive and there are well-defined quantitative structure activity relationship equations for predicting the carcinogenic potency of aromatic amines, an analysis was requested. The analysis consisted of Hansch equations to predict carcinogenic potency and discriminant functions to separate active from inactive compounds. The discriminant functions suggested the chemical would not be carcinogenic in male or female rats and male or female mice. The Hansch equations suggested that the carcinogenic potency of the notified chemical would be well below the values for the least active compounds used to derive the equations and supports the conclusions reached via the discriminant functions. These results are consistent with the weak genotoxicity of the chemical as found in various short term tests described below.

Endpoint and Result	*Assessment Conclusion*
Rat, acute oral LD50 > 2000 mg/kg bw	low toxicity
Rabbit, skin irritation	non-irritating (1% (w/v) solution)
Rabbit, eye irritation	non-irritating (1% (w/v) solution)
Guinea pig, skin sensitisation - adjuvant test.	no evidence of sensitisation.
Rat, oral repeat dose toxicity - 90 days.	NOAEL = 100 mg/kg/day bw
Genotoxicity - bacterial reverse mutation	mutagenic
Genotoxicity – in vitro mammalian cell gene mutation test	non genotoxic
Genotoxicity – in vitro human lymphocytes chromosomal aberration test	non genotoxic
Genotoxicity – in vitro unscheduled DNA synthesis in primary rat hepatocytes	non genotoxic
Genotoxicity – in vitro sister chromatid exchange in CHO cells	non genotoxic
Genotoxicity – in vivo mouse bone marrow micrnucleus test	non genotoxic
Toxicokinetic studies	rapid elimination mainly via the urine
Developmental and reproductive effects	none
Carcinogenicity (cell transformation assay)	negative

7.1. Acute toxicity – oral

TEST SUBSTANCE — Notified chemical.

METHOD
- Species/Strain: Rat/Sprague-Dawley.
- Vehicle: Distilled water.
- Remarks - Method: A summary of the test report only was submitted.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	5/sex	2000	None.

- LD50: > 2000 mg/kg bw
- Signs of Toxicity: Hunched posture and lethargy on the day of dosing.
- Effects in Organs: None.

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

TEST FACILITY — Safepharm (1991a).

7.2. Irritation – skin

TEST SUBSTANCE	1% (w/v) Solution of Notified chemical.
METHOD	OECD TG 404 Acute Dermal Irritation/Corrosion.
Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Vehicle	Distilled water.
Observation Period	72 hours.
Type of Dressing	Not stated.
Remarks - Method	A summary of the test report only was submitted.
CONCLUSION	A 1% (w/v) solution of the notified chemical is classified as non-irritating to skin.
TEST FACILITY	Safepharm (1991b).

7.3. Irritation - eye

TEST SUBSTANCE	1% (w/v) Solution of Notified chemical.
METHOD	OECD TG 405 Acute Eye Irritation/Corrosion.
Species/Strain	Rabbit/New Zealand White
Number of Animals	3
Observation Period	72 hours.
Remarks - Method	A summary of the test report only was submitted.
CONCLUSION	A 1% (w/v) solution of the notified chemical is classified as non-irritating to the eye.
TEST FACILITY	Safepharm (1991c).

7.4. Skin sensitisation

TEST SUBSTANCE	Notified chemical.
METHOD	OECD TG 406 Skin Sensitisation – Maximisation Test.
Species/Strain	Guinea pig/Dunkin Hartley.
Remarks - Method	A summary of the test report only was submitted.
PRELIMINARY STUDY	Not stated.
MAIN STUDY	
Number of Animals	Test Group: 20 Control Group: 10
INDUCTION PHASE	Induction Concentration intradermal injection, 0.5% topical application, 1%
Signs of Irritation	Not stated.
CHALLENGE PHASE	
1st challenge	topical application: 1%
CONCLUSION	There was no evidence of reactions indicative of skin sensitisation to the notified chemical under the conditions of the test.
TEST FACILITY	CIT (1991).

7.5. Repeat dose toxicity

TEST SUBSTANCE	Notified chemical.
METHOD	OECD TG 408 Repeated Dose 90-Day Oral Toxicity Study in Rodents.
Species/Strain	Rat/Wistar.
Route of Administration	Oral – gavage.
Exposure Information	Total exposure days: 90 or 91 days (males) and 91 or 92 days (females) or 92 days (recovery groups); Dose regimen: 7 days per week; Post-exposure observation period: 28 days.
Vehicle	0.5% aqueous Na-carboxymethyl cellulose.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw/day*	*Mortality*
I (control)	15/sex	0	None
II (low dose)	"	100	1 male (accidental)
III (mid dose)	"	316	None
IV (high dose)	"	1000	"
V (control recovery)	10/sex	0	"
VI (high dose recovery)	"	1000	"

Clinical Observations

A dose-dependent light to dark staining of the skin, fur, urine and bedding material. In all high dose animals transient apathy was seen in the first two weeks of treatment within a half hour after gavage; salivation was observed in 1 mid dose and most high dose animals; abnormal head posture and stereotype was observed occasionally.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis

Clinical chemistry Bilirubin was elevated in high dose males; creatinine was lower in mid and high dose females.

Urinalysis Bilirubin and urobilinogen were elevated in mid and high dose animals; urine pH was lower in high dose males.

Effects in Organs

Organ weights Increased relative liver weight and both absolute and relative kidney weights were seen in high dose recovery males; high dose females exhibited increase relative kidney weight and high dose recovery females had increased absolute adrenal weight.

Histopathology Renal tubular epithelial basophilia was observed in high dose males.

Remarks – Results

Target organs were the liver (increased serum bilirubin, urine bilirubin and urobilinogen, organ weight changes) and the kidney (lower creatinine, organ weight changes and histopathology).

CONCLUSION

The No Observed Adverse Effect Level (NOAEL) was established as 100 mg/kg bw/day in this study, based on effects on the liver and kidneys.

TEST FACILITY Austrian Research Center Seibersdorf (1994a).

7.6. Genotoxicity - bacteria

TEST SUBSTANCE	Notified chemical.
METHOD	Maron and Ames (1983) Plate incorporation procedure.
Species/Strain	*S. typhimurium*:

	TA1538, TA1535, TA1537, TA98, TA100.
Metabolic Activation System	Phenobarbital/β-naphthoflavone-induced rat liver S9 fraction.
Concentration Range in Main Test	a) With metabolic activation: 0 – 5000 µg/plate. b) Without metabolic activation: 0 – 5000 µg/plate.
Vehicle	Demineralised water.
Remarks - Method	A summary of the test report was submitted in addition to a full test report in German.
Remarks - Results	5000 µg/plate was not cytotoxic. The notified chemical was mutagenic to TA 98 in the presence of metabolic activation in a dose-dependent manner (maximum 13.8 times the spontaneous rate) and also to TA 1538 although the increase is marginal (maximum 2.3 times the spontaneous rate).
CONCLUSION	The notified chemical was mutagenic to bacteria under the conditions of the test.
TEST FACILITY	Fraunhofer Institute for Toxicology and Aerosol Research (1990).

7.7. Genotoxicity – in vitro (HPRT Assay)

TEST SUBSTANCE	Notified chemical.
METHOD	OECD TG 476 In vitro Mammalian Cell Gene Mutation Test.
Cell Type/Cell Line	Chinese Hamster V79 cells.
Metabolic Activation System	Aroclor 1254 induced rat liver S9 fraction.
Vehicle	Tissue culture medium.
Remarks - Method	In assay I, doses tested ranged from 10 to 1000 µg/mL. Six independent experiments were performed, 3 in the absence of S9, 2 with addition of primary hepatocytes as feeder cells and 1 in the presence of S9. In assay II the test substance was dissolved in DMSO and concentrations of 100, 500, 1000, 3000 and 5000 µg/mL were used. Only a summary report was provided for Assay 1. Raw data tables were not provided for Assay II.
Remarks - Results	In assay I a dose-dependent increase of mutant frequency was seen in only one experiment out of three in the absence of S9. Toxic effects were observed at concentrations above 1000 µg/mL in the absence of S9. Addition of hepatocytes or S9 decreased the toxicity of the test substance. No details of the mutagenic or toxic effects were provided. In assay II the test substance showed no toxic effects on the cells in the absence and presence of S9 up to the limit of solubility (5000 µg/mL).
CONCLUSION	The notified chemical was not mutagenic to Chinese Hamster V79 cells treated in vitro under the conditions of the test.
TEST FACILITY	Westendorf J, Univ. Hamburg Med. Sch. (1992a) and Fraunhofer Institute for Toxicology and Aerosol Research (1992).

7.10. Genotoxicity – in vitro (Chromosomal Aberration)

TEST SUBSTANCE	Notified chemical.
METHOD	OECD TG 473 In vitro Mammalian Chromosomal Aberration Test. EC Directive 84/449/EEC B.10

Cell Type/Cell Line	Human lymphocytes.
Metabolic Activation System	Aroclor 1254 induced rat liver S9 fraction.
Vehicle	DMSO.
Remarks - Method	No significant protocol deviations.

Metabolic Activation	*Test Substance Concentration (μg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	1250*, 2500*, 5000*	20 hours	20 hours
Present			
Test 1	1250*, 2500*, 5000*	4 hours	20 hours
Test 2	1250*, 2500*, 5000*	4 hours	30 hours

*Cultures selected for metaphase analysis.

RESULTS

Metabolic Activation	*Test Substance Concentration (μg/mL) Resulting in:*			
	Cytotoxicity in PreliminaryTest	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Absent				
Test 1		Dose-related decrease in mitotic index		Positive
Present				
Test 1				Negative
Test 2				Negative

Remarks - Results	The positive genotoxic effect was statistically significant only if gaps were included but occurred at both 2500 and 5000 μg/mL (up to 8.5% of cells with structural alterations).
CONCLUSION	The notified chemical was not unequivocally clastogenic to human lymphocytes treated in vitro under the conditions of the test.
TEST FACILITY	Safepharm (1992a).

7.11. Genotoxicity – in vitro (Unscheduled DNA Synthesis)

TEST SUBSTANCE	Notified chemical.
METHOD	Butterworth *et al.* (1987) – unscheduled DNA synthesis assay.
Cell Type/Cell Line	Primary rat hepatocytes.
Vehicle	Tissue culture medium.
Remarks - Results	Cells were treated with notified chemical at 1.95 to 2000 μg/mL and no dose-dependent increase of net silver grains over the cell nucleus area was observed. Toxic effects were observed above 500 μg/mL.
CONCLUSION	The notified chemical did not induce unscheduled DNA synthesis in primary rat hepatocytes treated in vitro under the conditions of the test.
TEST FACILITY	Westendorf J, Univ. Hamburg Med. Sch. (1992b).

7.12. Genotoxicity – in vitro (Sister Chromatid Exchange)

TEST SUBSTANCE	Notified chemical.
METHOD	OECD TG 479 Genetic Toxicology: In vitro Sister Chromatid Exchange Assay in Mammalian Cells.

Cell Type/Cell Line	Chinese Hamster Ovary (CHO) cells.
Metabolic Activation System	Aroclor 1254 induced rat liver S9 fraction.
Vehicle	DMSO.
Remarks - Method	Cells were treated with 300 – 2400 µg/mL notified chemical for 3 hours or 24 hours in the absence of S9 and with the same concentrations of the notified chemical in the presence of S9 for 3 hours.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Harvest Time*
Present			
Test 1	300, 600, 1200, 2400	3 hours	27 hours
Absent			
Test 1	100, 200, 300, 600, 1200, 2400	3 hours	27 hours
Test 2	100, 200, 300, 600, 1200, 2400	24 hours	24 hours

RESULTS

Remarks - Results	The notified chemical induced a slight (less than doubling) increase in SCE frequency with and without S9 which was not dose-dependent.

CONCLUSION The notified chemical was not unequivocally genotoxic to CHO cells treated in vitro under the conditions of the test.

TEST FACILITY Fraunhofer Institute for Toxicology and Aerosol Research (1991).

7.13. Genotoxicity – in vivo

TEST SUBSTANCE Notified chemical.

METHOD OECD TG 474 Mammalian Erythrocyte Micronucleus Test.
EC Directive 84/449/EEC B.12 Mutagenicity Mammalian Erythrocyte Micronucleus Test.

Species/Strain	Mouse/CD 1.
Route of Administration	Oral – gavage.
Vehicle	Distilled water.
Remarks - Method	In a range finding study, intraperitoneal administration of the notified chemical to 4 animals (2/sex) at 5000 mg/kg bw resulted in deaths of all animals at the 24-hour observation point. Clinical signs in a group of 4 animals (2/sex) administered the notified chemical orally were lethargy, hunched posture, pilo-erection, laboured respiration, ataxia and ptosis.

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Sacrifice Time hours*
1	5/sex	5000	24, 48 and 72 hours

RESULTS

Doses Producing Toxicity	A significant change in the NCE/PCE ratio was observed in the 48- and 72-hour treatment groups. Clinical signs included lethargy, hunched posture, pilo-erection, laboured respiration and ptosis.
Genotoxic Effects	None.

CONCLUSION The notified chemical was not clastogenic in this in vivo micronucleus test under the conditions of the test.

TEST FACILITY Safepharm (1992b).

7.14. Carcinogenicity – Malignant transformation in vitro

TEST SUBSTANCE	Notified chemical.
METHOD	
Cell Type/Cell Line	C3H Mouse M2-Fibroblasts.
Metabolic Activation System	Aroclor 1254 induced rat liver S9 fraction.
Vehicle	DMSO.
Remarks - Method	The test substance was dissolved in DMSO and tested in a concentration range of 50 – 4000 µg/mL (with and without metabolic activation).

Test Substance Concentration (µg/mL)
50, 100, 250, 500, 1000, 2000, 4000

Remarks - Results	The test substance was tested up to concentrations inducing significant cytotoxicity and was detoxified by S9.
CONCLUSION	The notified chemical did not induce malignant transformation in C3H Mouse M2 Fibroblasts.
TEST FACILITY	University Hamburg Medical School (1994).

7.15. Developmental toxicity

TEST SUBSTANCE	Notified chemical.
METHOD	OECD TG 414 Teratogenicity.
Species/Strain	Rat/Wistar.
Route of Administration	Oral – gavage.
Exposure Information	Exposure period: days 6 – 15 of gestation.
Vehicle	Aqueous 0.5% Na carboxymethycellulose solution.

RESULTS

Group	*Number of Animals*	*Dose mg/kg bw/day*	*Mortality*
1	24 females	0	None reported
2	"	40	"
3	"	200	"
4	"	1000	"

Effects on Dams

Slight decrease in body weight at the beginning of the dosing period (days 6 – 11) for high dose animals.

Effects on Foetus

Skeletal effects were only observed in the control group with significantly more litters with foetuses not completely ossified or with unossified hyoid. In one control foetus abnormal curvature of the spine and fused sternebrae were observed.

Remarks - Results

The authors noted there were no test substance related effects on dams or foetuses in the low or mid dose groups.

CONCLUSION

The notified chemical was not teratogenic and exhibited slight maternal toxicity at doses up to 1000 mg/kg/day.

TEST FACILITY Austrian Research Center Seibersdorf (1994b).

7.16. Toxicokinetic study

TEST SUBSTANCE Notified chemical.

METHOD Not stated.

STUDY DESIGN AND OBJECTIVE
The ^{14}C-labelled chemical was integrated in a hair dyeing formulation or used as a solution in water. The hair dyeing formulation was mixed with a 6% hydrogen peroxide solution before application.

Cutaneous application: The notified chemical was applied to the dorsal skin of Him: OFA rats for 30 minutes prior to washing off either in a formulation and the rats killed after 24 or 72 hours or in a solution and the rats killed after 72 hours. For the rats killed after 24 hours, radioactivity was measured in the blood. For the other two groups the rinsing water, treated skin, urine, faeces, adrenals, blood, brain, fat, femur, heart, kidney, liver, lung, muscle, ovaries, skin (untreated), spleen, testes, thyroids and carcass were examined for radioactivity.

Oral application: Two groups received the test substance by gavage. One group was killed after 24 hours and the radioactivity determined in the blood. The other group was killed after 72 hours and the radioactivity determined in the urine, faeces, organs and carcass without gastrointestinal tract.

RESULTS
The mean percutaneous absorption rate was found to be approximately 0.1%. The test substance was excreted mainly via the urine (82 – 89% of applied radioactivity) and to a lesser extent via the faeces (11 – 18% of eliminated radioactivity). The mean excretion was rapid (88 – 94% eliminated during the first 24 hours) and the concentration of radioactivity was at or below the detection limit in blood or organs at 72 hours after dosing.

CONCLUSION
The notified chemical is not taken up cutaneously to great extent in conditions similar to hair dye application and when absorbed via the gastrointestinal tract is rapidly eliminated in the urine in rats.

8. ENVIRONMENT

8.1. Environmental fate

No environmental fate data were submitted. The notified chemical is not expected to bioaccumulate based on the measured partition coeffiecient.

8.2. Ecotoxicological investigations

Ecotoxicity data were submitted only for the aquatic invertebrate, Daphnia.

8.2.1. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE	HC Violet AS
METHOD	EC Directive 92/69/EWG Part C:C.2. Acute Toxicity for Daphnia
Species	*Daphnia magna*
Exposure Period	48 hours
Auxiliary Solvent	none
Water Hardness	2.51 mmol/L (Ca^{2+} and Mg^{2+})
Analytical Monitoring	Test concentration (method not reported)
Remarks – Method	Four replicates containing 5 daphnia per test group were exposed, without renewal, to a control and 9 test concentrations between 2.0 and 500 mg/L for 48 hours. A sensitivity control test using potassium di-chromate was also performed and resulted in a mean 24 h EC50 of 1.4 mg/L. Water parameters of temperature, dissolved oxygen and pH were measured daily and were in the acceptable range, except for a slight change in pH, which fluctuated toward the alkaline range (7.8-8.1). The fluctuation in pH was not considered to affect the results. The endpoints and concentration effect relationship were determined by probit analysis after 24 and 48 hours.

RESULTS

Concentration		*Number Still mobile*	
mg/L	*Number of daphnia*	*24 h*	*48 h*
0	20	20	20
2.0	20	20	20
4.0	20	20	20
8.0	20	20	20
16.0	20	19	18
31.25	20	17	15
62.5	20	13	4
125	20	7	2
250	20	5	1
500	20	5	0

EC50	45 mg/L (CI 35–57 mg/L) at 48 h
NOEC (or LOEC)	8 mg/L at 48 hours
Remarks – Results	After 24 hours, dead animals were found in concentrations of 16 mg/L and above. In the highest test concentration, all animals were dead after 48 hours. Determination of the EC50 was by an adjusted probit method.
CONCLUSION	The notified chemical is harmful to *Daphnia magna.*
TEST FACILITY	Ingenieurgesellschaft Wasser – Und Tiefbau mbH (1993)

9. RISK ASSESSMENT

9.1. Environment

9.1.1. Environment – exposure assessment

The notified chemical will be imported as a component (1%) of a finished hair dye product for use in hair salons. No manufacturing or reformulation is required. Up to 95% of the notified chemical is expected to end up in the sewer during end use when excess dye is washed from the hair after treatment. A further 5% could end up in landfill as residues in used containers.

The notifier provided a predicted environmental concentration (PEC) in the aquatic environment of 0.005 ppm for metropolitan areas and 0.125 ppm for rural areas. The PEC was calculated assuming release of 0.3 g of notified chemical for each use of the product, with 60 L of water used during the washing process, and no adsorption to the hair. A dilution factor of 1:10 was assumed for the metropolitan sewer, sewage treatment plant, and receiving waters.

We have also calculated a daily PEC using the worst-case scenario of 100% of the import volume being discharged to sewer each year in a diffuse manner with no attenuation within the sewage systems. Based on dilution factors of 0 and 10 for inland and ocean discharges of STP-treated effluents, the predicted daily PEC of the notified chemical in fresh water is approximately 0.07 μg/L and in marine surface waters, approximately 0.007 μg/L. We assume an Australian population of 19.5 million people and an average value for water consumption of 200 L/person/day (3900 ML/day for total population).

The notified chemical is readily water soluble and is expected to remain predominantly in the water compartment, unless ionized, when it should partition to sediment. The chemical is not expected to volatilise from water, or to partition into sludge or sediment. The notifier provided modelled data of biodegradation using BIOWIN, which indicated the notified chemical is not readily biodegradable, but may ultimately degrade with a time frame of weeks to months. On the basis of log Kow, there is a low potential for bioconcentration of the notified chemical in exposed aquatic organisms.

9.1.2. Environment – effects assessment

One measured toxicity endpoint was provided for aquatic organisms. The data indicate an LC50 of 45 mg/L for *Daphnia magna* in an acute toxicity test. Using the EC50 for *Daphnia magna*, and assuming a safety factor of 1000 (since measured toxicity data are available for only one trophic level), the predicted no effect concentration (PNEC) is 0.045 mg/L.

ECOSAR modelling was provided based on the measured log Pow (0.11) and using neutral organics SAR. These data predict acute and chronic toxicity endpoints for fish, Daphnia, green algae, mysid shrimps, and earthworms. The results are listed below:

Species	Duration/parameter	Endpoint
Fish	96 h LC50	9319 mg/L
Daphnia	48 h LC50	8763 mg/L
Green algae	96 h EC50	4914 mg/L
Mysid shrimp	96 h LC50	10,358 mg/L
Earthworms	14 day LC50	4942 mg/kg dry soil weight

However, these data should be viewed with caution, noting that the measured EC50 for Daphnia indicates a 195 fold higher toxicity than is indicated by the modelled data. This is a direct result of estimation for the neutral form and demonstrates the caution needed with QSAR estimates of ionizable organic substances (Clements *et al*. 1993). As indicated under dissociation constant, in water, the more toxic cationic forms are likely to predominate.

9.1.3. Environment – risk characterisation

Location	PEC (μg/L)	PNEC (μg/L)	Risk Quotient (RQ)[(a)]
Australia-wide STPs			
Ocean outfall	0.007	45	1.6×10^{-4}
Inland River	0.07[b]	45	1.6×10^{-3}

a. RQ = PEC ÷ PNEC. b. PEC values calculated assuming no attenuation of notified chemical in biosolids and no loss through volatilisation during STP process

On the basis of the RQ values provided in the table above, the low volumes used, and nationwide and diffuse use of the notified chemical, it is not considered to pose an unacceptable risk to the health of aquatic life based on its reported use pattern.

9.2. Human health

9.2.1. Occupational health and safety – exposure assessment

Exposure of hair salon workers to the notified chemical during mixing of the imported product with developer and application of the mixture to hair is expected to be low given its low concentration in the product and the use of gloves. Some accidental dermal exposure can be expected but this would be expected to be intermittent.

9.2.2. Public health – exposure assessment

The public may be exposed to the notified chemical following a transport accident but this exposure is limited by the low concentration in the imported product and the rarity of accidents.

The public will be exposed to the notified chemical during mixing and application in a similar way to salon workers. Again exposure is likely to be low given the concentration of chemical in the imported product and the likely use of gloves.

9.2.3. Human health - effects assessment

The notified chemical exhibited low acute toxicity via the oral route in rats. It was not a skin or eye irritant in rabbits at the concentration in the imported product and was not a skin sensitiser in guinea pigs. The NOAEL in a 28-day subchronic study in rats was 100 mg/kg/day bw based on effects on liver and kidneys. The only histopathological changes of importance were renal tubular basophilia at 316 mg/kg bw/day. Therefore, the notified chemical would not be classified as a hazardous substance according to NOHSC (NOHSC, 1999) or GHS (UN, 2003) criteria.

The notified chemical is related to aromatic amines known to be carcinogenic but was negative in a number of in vitro and in vivo short term genotoxicity tests. However, the chemical was mutagenic in bacteria. Toxicokinetic data suggested rapid elimination in rats. Given the fact that the chemical is directly applied to the scalp, it was important to be sure the probability of carcinogenicity was low. There are good QSAR descriptors for aromatic amines and an expert opinion was submitted. Using these descriptors, the probability of the notified chemical being a carcinogen was assessed as low.

9.2.4. Occupational health and safety – risk characterisation

The likely low hazard of the notified chemical (particularly with respect to mutagenicity testing and QSAR modelling of carcinogenicity), its low concentration in the imported product (< 1%) coupled with standard use of impervious gloves by hair salon workers suggests a low risk of adverse health effects from normal use. The risk to workers involved in transport, storage and disposal of the notified chemical is also low on the basis of low hazard and limited exposure.

9.2.5. Public health – risk characterisation

The public may come in contact with the notified chemical in the imported product for approximately 6 hours per year. This low exposure coupled with the low hazard suggests a low risk of adverse health effects. Even in the event of a transport accident public exposure to large spills is unlikely given the nature of the packaging of the imported product and the public health risk is low.

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

10.1. Hazard classification

Under the Global Harmonised System for Classification and Labelling of Chemicals (United Nations, 2003), the notified chemical is classified as harmful to *Daphnia magna*.

Based on the available data the notified chemical is not classified as hazardous under the NOHSC *Approved Criteria for Classifying Hazardous Substances* (NOHSC, 1999).

10.2. Environmental risk assessment

On the basis of the PEC/PNEC ratio: the chemical is not considered to pose a risk to the environment based on its reported use pattern. However, if import volumes are increased, further information will be required, including the full suite of ecotoxicity and physico-chemical data so that a more comprehensive risk assessment can be performed.

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 Negligible Concern to public health when used as described.

11. MATERIAL SAFETY DATA SHEET

11.1. Material Safety Data Sheet

The MSDS of a product containing the chemical provided by the notifier was in accordance with the NOHSC *National Code of Practice for the Preparation of Material Safety Data Sheets* (NOHSC, 1994a). 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 a product containing the chemical provided by the notifier was in accordance with the NOHSC *National Code of Practice for the Labelling of Workplace Substances* (NOHSC, 1994b). The accuracy of the information on the label remains the responsibility of the applicant.

12. RECOMMENDATIONS

CONTROL MEASURES

Occupational Health and Safety

- 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.

Environment

Disposal

- The notified chemical should be disposed of in landfill.

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 Section 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.

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