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NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME (NICNAS)

FULL PUBLIC REPORT

2-Pyrrolidinone, 1-(2-hydroxyethyl)-

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Director
Chemicals Notification and Assessment

TABLE OF CONTENTS

It should also be noted that the total amount of notified chemical available for human exposure is limited by the maximum amount of ink in the small cartridge, although several colour ink cartridges may be used

FULL PUBLIC REPORT

2-Pyrrolidinone, 1-(2-hydroxyethyl)-

1. APPLICANT AND NOTIFICATION DETAILS

APPLICANT(S)
Hewlett-Packard Australia Pty Ltd

NOTIFICATION CATEGORY
Standard: Chemical other than polymer (more than 1 tonne per year).

EXEMPT INFORMATION (SECTION 75 OF THE ACT)
Data items and details claimed exempt from publication:
Identity and levels of impurities
Import volume
Details of formulation and use

VARIATION OF DATA REQUIREMENTS (SECTION 24 OF THE ACT)
Variation to the schedule of data requirements is claimed as follows:
Hydrolysis as a function of pH
Adsorption/Desorption
Dissociation Constant
Explosive properties
Acute dermal toxicity
Acute inhalation toxicity
Repeat dose toxicity
In vivo genotoxicity
Toxicity in fish

PREVIOUS NOTIFICATION IN AUSTRALIA BY APPLICANT(S)
None.

NOTIFICATION IN OTHER COUNTRIES
None known.

2. IDENTITY OF CHEMICAL

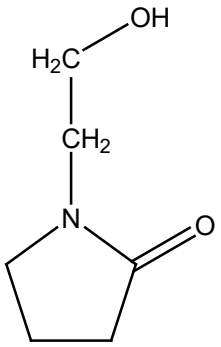
CHEMICAL NAME
2-Pyrrolidinone, 1-(2-hydroxyethyl)-

OTHER NAME(S)
1-(2-Hydroxyethyl)pyrrolidin-2-one
1-(2-Hydroxyethyl)-2-pyrrolidone;
N-(2-Hydroxyethyl)-2-pyrrolidone
N-2-Hydroxyethylpyrrolidin-2-one;
HEPD
HEP
Hydroxyethyl pyrrolidone

CAS NUMBER
3445-11-2

MOLECULAR FORMULA
$C_6H_{11}NO_2$

STRUCTURAL FORMULA



MOLECULAR WEIGHT
129.2

SPECTRAL DATA

METHOD	IR
Remarks	Condensed phase spectrum, peaks at: 3380, 2930, 2870, 2030, 1960, 1670, 1500, 1470, 1430, 1365, 1320, 1300, 1175, 1080, 1065, 1010, 930, 870, 750, 650 575, 520, 455 cm^{-1}
TEST FACILITY	NIST (2004a)

METHOD	Mass spectrum (no further details provided)
Remarks	M/Z reported. Peaks clustered at 15(8.7), 28(34), 41(48), 70(71), 98(>100), 111(15), 114(15), 129(14) (parent ion).
TEST FACILITY	NIST (2004b)

METHODS OF DETECTION AND DETERMINATION

METHOD	GC/MS, ^{1}H-NMR.
Remarks	2004 GC data also provided.
TEST FACILITY	BASF (2001a)

3. COMPOSITION

DEGREE OF PURITY
High

ADDITIVES/ADJUVANTS
None

4. INTRODUCTION AND USE INFORMATION

MODE OF INTRODUCTION OF NOTIFIED CHEMICAL (100%) OVER NEXT 5 YEARS
The notified chemical will be imported as a component (<14%) of small printing cartridges.

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

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

USE
Component of printing ink.

5. PROCESS AND RELEASE INFORMATION

5.1. Distribution, transport and storage

PORT OF ENTRY
Melbourne

IDENTITY OF MANUFACTURER/RECIPIENTS
Hewlett Packard Australia Pty Ltd, 31-41 Joseph Street, Blackburn Victoria 3130.
The product will be supplied to offices nationwide and office equipment retailers.

TRANSPORTATION AND PACKAGING
By ship in containers. Individual cartridges will be packed in sturdy cardboard boxes and would normally be transported by road. The cartridge containing the notified chemical is not a dangerous good, hazardous substance, or scheduled poison, and therefore no special transport or packaging requirements are necessary.

5.2. Operation description

No reformulation or repackaging of the imported product containing the notified chemical occurs in Australia. Sealed ink cartridges containing the notified chemical will be handled by service technicians or office workers or the public, who will replace spent cartridges in the printers as necessary. Office workers and the public will also use the printers for varied printing work. The ink cartridges containing the notified chemical are designed for a single use, and will not be refilled.

5.3. Occupational exposure

Number and Category of Workers

Category of Worker	*Number*	*Exposure Duration*	*Exposure Frequency*
Service technicians (estimate)	10	8 h/day	230 days/year
Office workers	1000	5 to 10 minutes/day	10 days/year

Exposure Details

Dermal or inhalation exposure of workers to the notified chemical could occur during replacement of cartridges in the printers, or during normal use of the printers. Both service technicians and office workers would experience the same types of exposure, but exposure duration is likely to be greater for the former.

Exposure while changing cartridges is expected to be limited to dermal exposure, occurring if the ink is inadvertently touched. However this would be avoided by users and would be evident if it occurred.

Similarly, occasional dermal exposure during use of the printer could occur if the printed pages were touched inadvertently before the ink dried, or if ink-stained parts of the printer were touched. Such exposure is expected to be low and to be avoided by workers.

Routine inhalation exposure to vapour or aerosol could occur during use of the printer. A proportion of the notified chemical is expected to evaporate from the printed page during the drying process. Although the boiling point is relatively high at 309^{o}C and vapour pressure expected to be corresponding low, some evaporation from thin films of print is considered likely. An additional small proportion of ink in aerosol form may escape from the printer enclosure into the atmosphere and be inhaled.

Controls on exposure are likely to be ventilation (either natural or mechanical) and PPE designed to prevent dermal or inhalation exposure. It is likely that the type of ventilation will reflect the other needs of the workplace eg temperature control. While PPE is recommended on the MSDS for inks containing the notified chemical, in most workplaces such controls may not be used for routine printing and cannot be depended on to reduce exposure.

Dermal or inhalation exposure of workers through handling printed paper is expected to be very low. While some continuing evaporation of the notified chemical would be expected after the main print drying process, this would occur at a very slow rate and produce very low concentrations in air.

5.4. Release

RELEASE OF CHEMICAL AT SITE

No release is expected as manufacture and reformulation of the notified chemical will not take place in Australia.

RELEASE OF CHEMICAL FROM USE

Release of the contents of the cartridge to the environment is not expected under normal use. The cartridge is installed inside of the machine or printer and designed to prevent leakage. Therefore, no environmental release is expected in the case that cartridge is replaced. However, if leakage or spill does occur, the ink will be contained with absorbent material which will be disposed of to landfill.

Once the notified chemical is released onto the paper, most of the notified chemical is expected to remain sorbed to the media and trapped within the print on the paper. A very small amount of the notified chemical, approximately 1% is likely to be volatized or emitted into air from printing application. The balance of HEPD is sorbed into the paper. Based on 0.4 grams of ink printed per page, the estimation of the notified chemical present in aerosol is calculated as follows:

<u>If 1% of the ejected ink goes into aerosol:</u>	
Amount of ink in aerosol generated inside the printer:	= 0.01 × 0.4 = 0.004 g
If amount of ink aerosol that escapes the printer enclosure is 20%:	= 0.2 × 0.004 = 0.0008 g
Amount of HEP in aerosol that escapes:	= mole fraction HEP × 0.0008 = 0.02 × 0.0008 = 0.000016 g.

Paper to which the notified chemical will be bound will eventually be buried in landfill or be incinerated, or the chemical may be released in effluent from de-inking processes. However, very little if any of the notified chemical is expected by the notifier to survive to this stage as the notified chemical is semi-volatile. Recent literature suggests that current paper recycling rates in Australia are 70-92% (Australian Environmental Review, 2001). Consequently, most of the paper containing the notified chemical could be recycled. Residues left in empty cartridges (estimated as <1%) will most likely be disposed of to landfill. Spent cartridges collected by the recovery system are recycled or reused along with all residual ink in the recycling process. Spent cartridges that are not recycled are likely to be sent to landfill.

5.5. Disposal

The notified chemical enclosed in cartridges can be disposed of directly by landfill. It can also be disposed of indirectly from waste paper containing the notified chemical via recycling, to landfill or by incineration.

5.6. Public exposure

The scenarios by which the public may be exposed to the notified chemical would involve home use of printers, and are similar to those for office workers (see section 5.3 above).

Using the same basic calculations as for exposure of workers, one factor that may cause higher exposure for the public is the lack of mechanical ventilation to disperse the vapour in private homes. On the other hand, it is less likely that the public would experience the higher range of exposures, through printing many pages at one time, or through routine daily printing.

Exposure expressed as mg/kg bw would be increased if the printers were used by children.

6. PHYSICAL AND CHEMICAL PROPERTIES

Appearance at 20ºC and 101.3 kPa Colourless to yellow liquid, odourless.
Technical data sheet (BASF, 1997a) states that the chemical has a weak amine odour.

Melting Point/Freezing Point 20°C

Remarks Stated on Technical Data Sheet (Chemical Intermediates) 1997 (BASF, 1997a).

< -4°C

Remarks Stated on Technical Leaflet, (BASF, 2001b).

Boiling Point 309ºC. Pressure not specified.

Remarks Report not provided.

Density 1144kg/m^3 at 20ºC

Remarks Report not provided

Vapour Pressure 0.1 kPa at 120ºC.

METHOD Dynamic method with argon atmosphere
Remarks Purity 99.7%, with the remainder being neighbouring constituents. The assay was distilled at 1 millibar (0.1 kPa) in a fractionating column with approximately 50 theoretical trays. It is not possible to confirm this result from the limited details provided.
TEST FACILITY BASF (1985)

<0.1 kPa at 20.0ºC
<0.1 kPa at 50.0ºC

METHOD Dynamic method based on the Cottrell pump principle.
Remarks Summary only provided. Based on the MPBPWIN (v1.40) program, the vapour pressures were calculated to be ranging from 0.019-0.64 Pa with the modified grain method result of 0.019 preferred. These values indicate that the notified chemical is considered to be moderately volatile (Mensink et al 1995).
TEST FACILITY BASF (1992)

Water Solubility Completely miscible.

Remarks Report not provided. It is assumed that the notified chemical is soluble in water, noting the low log Pow and that the MSDS states that the ink containing <14% of the notified chemical is soluble in water.
Based on the WSKOW v1.40 program, the water solubility was calculated to be 770 g/L, indicating that the notified chemical is readily soluble (Mensink et al 1995).

Hydrolysis as a Function of pH Not determined

Remarks It contains a cyclic amide which may undergo hydrolysis only under extreme conditions and will be stable under ambient environmental conditions in the pH

range of 4-9.

Partition Coefficient (n-octanol/water) Log Pow is -1.03 at 25ºC

METHOD The test was conducted in accordance with 92/69/EWG of the EU commission dated July 31, 1992, Part A8: Partition Coefficient; shake extraction method and following analysis of test substance in separated phases with gas chromatography.
Remarks The result correlates well with the theoretical log Pow calculation of N-(2-hydroxyethyl)-2-pyrrolidone according to the KOWWIN program; Syracuse Research Corp., Merrill Lane = C log P = -1.08. This value was used in the water solubility calculation above.
TEST FACILITY BASF (1997b)

Adsorption/Desorption Not determined

Remarks Based on the relatively low log P and the high water solubility, the adsorption of the notified chemical to organic matter in soil is expected to be low. This is confirmed by the Zahn-Wellens Test in Section 8.1.1 (b).

Dissociation Constant Not determined

Remarks The chemical is not expected to dissociate in water as it has no readily ionisable groups.

Flash Point 174^0C

METHOD Stated as DIN 51578
Remarks Summary only provided.
TEST FACILITY BASF (1992)

Flammability Limits Upper explosion limit: 9.90% by volume (219^0C)
Lower explosion limit: 1.50% by volume (168^0C)

METHOD Stated to be saturation method.
Remarks Summary only supplied.
TEST FACILITY BASF (1992)

Autoignition Temperature 275ºC

METHOD Stated as DIN 51794
Remarks Summary only provided
TEST FACILITY BASF (1992)

Explosive Properties Test not conducted.

Remarks Not expected to be explosive on structural grounds.

Reactivity Stable under normal conditions.

Remarks Avoid excessive heat, ignition sources. Esters may contribute to peroxide formation. Incompatible with acids and bases.
Hazardous decomposition products: carbon monoxide, carbon dioxide, nitrogen oxide.

7. TOXICOLOGICAL INVESTIGATIONS

The majority of the toxicity data was submitted for the notified chemical. Data for the following endpoints: Acute Dermal Toxicity, Repeat Dose Toxicity and *in vivo* Genotoxicity were taken from the IUCLID Data Set for 2-Pyrrolidone (Toxicology and Regulatory Affairs, 2003). 2-Pyrrolidone was considered to be a suitable analogue as its acute toxicity is similar to the notified chemical and it has similar physico-chemical properties.

Endpoint and Result	*Test Substance*	*Assessment Conclusion*
Rat, acute oral	Notified chemical	low toxicity, LD50 > 14,430 mg/kg bw
Rat, acute oral	Notified chemical	low toxicity, LD50 16,900 – 22,400 mg/kg bw
Rat, acute dermal	2-Pyrrolidone	low toxicity, LD50 > 2000 mg/kg bw
Rat, acute inhalation	2-Pyrrolidone	Not highly toxic, LC50 > 0.28 mg/L/8 h
Rabbit, skin irritation	Notified chemical	non-irritating
Rabbit, eye irritation	Notified chemical	slightly irritating
Guinea pig, skin sensitisation – non-adjuvant test.	Notified chemical	no evidence of sensitisation
Rat, repeat dose oral toxicity – 90 days.	2-Pyrrolidone	NOAEL 207 mg/kg bw/day
Genotoxicity – bacterial reverse mutation a)	Notified chemical	non mutagenic
Genotoxicity – bacterial reverse mutation b)	Notified chemical	mutagenic
Genotoxicity – bacterial reverse mutation c)	Notified chemical	mutagenic
Genotoxicity – bacterial reverse mutation d)	Notified chemical	mutagenic
Genotoxicity – in vitro Sister Chromatid Exchange Assay	Notified chemical	weakly clastogenic
Genotoxicity – in vitro Chromosome Aberration Test	Notified chemical	not clastogenic
Genotoxicity – in vitro Mammalian Cell Gene Mutation Test	Notified chemical	weakly clastogenic
Genotoxicity – in vitro Mammalian Cell Transformation Assay	Notified chemical	not clastogenic
Genotoxicity – in vivo Mammalian Erythrocyte Micronucleus Test	2-Pyrrolidone	non genotoxic

7.1 Acute Toxicity Oral

7.1.1 Acute toxicity – oral a)

TEST SUBSTANCE	Notified chemical
METHOD	
Species/Strain	Rat/Wistar
Vehicle	
Remarks – Method	A one page summary of the study was supplied. The rats were fasted for approximately 18 hours prior to administration of the test material. The material was given orally by intubation on a g/kg basis. Following treatment the animals were returned to their cages and food and water were freely available. The rats were observed for signs of toxicity and pharmacological effects for 14 days following treatment. Necropsies were inadvertently not performed.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
1	10 male	5000	2
2	10 male	7120	0
3	10 male	10,140	0
4	10 male	14,430	0

LD50	> 14,430 mg/kg bw
Signs of Toxicity	For each dose group the following signs of toxicity were noted: 5000 mg/kg bw: Lethargy, chromorhinorrhea: 7120 mg/kg bw: Diarrhoea; 10,140mg/kg bw: Lethargy, chromorhinorrhea; and 14,430 mg/kg bw: Diarrhoea, chromorhinorrhea, piloerection and lethargy.
Effects in Organs	Necropsies were not performed.
Remarks – Results	Two animals in the lowest dose group (5000 mg/kg) died on days 4 and 6 after dosing. No comment was made on these deaths in the test report. However, as no mortality was seen at significantly higher doses, the deaths are likely to be unrelated to the test substance.

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

TEST FACILITY MB Research Laboratories (1978)

7.1.2. Acute toxicity – oral b)

TEST SUBSTANCE Notified chemical

METHOD

Species/Strain	Rat/Sherman-Wistar
Vehicle	Test substance administered as supplied
Remarks – Method	One-page summary of study was provided. Five groups of albino rats, consisting of three males and two females, were set aside and observed for a period of one week to assure normalcy. The animals were then starved for 24 hours. Doses at levels as indicated in the table below were determined for each animal and administered directly into the stomach by means of a syringe and stomach tube. Following this, the animals were allowed food and water ad libitum during a fourteen-day observation period.

RESULTS

Group	*Number and Sex of Animals*	*Dose ml/kg bw*	*Mortality*
1	5 (3 males, 2 females)	10.00	0
2	5 (3 males, 2 females)	12.6	0
3	5 (3 males, 2 females)	16.0	2
4	5 (3 males, 2 females)	20.0	4
5	5 (3 males, 2 females)	25.1	5

LD50	14.8 – 19.6 ml/kg bw (~16,900 – 22,400 mg/kg bw)
Signs of Toxicity	Not reported
Effects in Organs	Not reported
Remarks – Results	The mortality data were evaluated according to the Thompson Moving Average Method as described by Carrol. S. Weil in his publication entitled “Tables for Convenient Calculation of Median-Effective Dose

(LD50 or ED50) and Instructions in Their Use", which appeared in Biometrics, Vol. 8, No. 3, pp 249-263, September 1952.

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

TEST FACILITY FDR Labs (1971a)

7.2. Acute toxicity – dermal

TEST SUBSTANCE 2-Pyrrolidone

METHOD OECD TG 402 Acute Dermal Toxicity – Limit Test.

Species/Strain	Rabbit/New Zealand White
Vehicle	Test substance administered as supplied
Type of dressing	Occlusive.
Remarks - Method	No significant protocol deviations. Full study report (1992) not reviewed.

RESULTS

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Mortality*
I	5 male	2000	0
II	5 female	2000	0

LD50	> 2000 mg/kg bw
Signs of Toxicity - Local	Dermal reactions were slight to well-defined on day 1 but were absent on days 7 and 14.
Signs of Toxicity - Systemic	There were no abnormal systemic signs noted in 9/10 animals. One male exhibited red staining of the nose/mouth area and an apparent cataract in the right eye on day 5. This was considered to result form a self-inflicted injury unrelated to the test substance. Body weight gains were normal at all weighing periods.
Effects in Organs	Necropsy did not reveal any treatment related changes.
Remarks - Results	None

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

TEST FACILITY Toxicology and Regulatory Affairs (2003).

Acute toxicity - Inhalation

7.3.1 Acute toxicity – inhalation of notified chemical

TEST SUBSTANCE Notified chemical

RESULTS Not determined. However, an MSDS (BASF, 2003) mentions that there were no deaths in an 8 h inhalation safety screen.

7.3.2 Acute toxicity – inhalation of analogue

TEST SUBSTANCE 2-Pyrrolidone, distilled, solid.

METHOD BASF in-house Inhalation Risk Test. Full study results not reviewed.

Species/Strain	Rat, strain not specified.
Vehicle	None
Method of Exposure	Not specified
Exposure Period	8 hours

Physical Form	vapour.
Remarks - Method	Rats were exposed to a saturated concentration in air of 2-pyrrolidone, stated to be approximately 80 ppm (280 mg/m^3). The test was part of toxicological pre-testing of the substance carried out in 1961.

RESULTS

Group	*Number and Sex of Animals*	*Concentration ppm*		*Mortality*
		Nominal	*Actual*	
1	6 (sex not specified)	80	-	None

LC50	> 0.28 mg/L/8 hours
Remarks - Results	None

CONCLUSION The notified chemical is not highly toxic via inhalation.

TEST FACILITY Toxicology and Regulatory Affairs (2003).

7.4. Irritation – skin

TEST SUBSTANCE Notified chemical

METHOD In accordance to Section 191.11 of the Final Order, Enforcement Regulations, Federal Register, Vol. 26, No. 155, P. 7336, 12 August 1961

Species/Strain	Rabbit/Albino
Number of Animals	6
Vehicle	Test substance administered as supplied
Observation Period	72 hours
Type of Dressing	Not provided
Remarks – Method	The test substance was applied to both abraded and intact sites. Observations were made 24 h and 72 h after dosing.

RESULTS

Lesion	*Mean Score**	*Maximum Value*	*Maximum Duration of Any Effect*	*Maximum Value at End of Observation Period*
Erythema/Eschar	0	0	N/A	0
Oedema	0	0	N/A	0

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

Remarks – Results	The test material produced no erythema or oedema in any of the animals.

CONCLUSION The notified chemical is non-irritating to skin.

TEST FACILITY FDR Labs (1971b)

7.5. Irritation – eye

TEST SUBSTANCE Notified chemical

METHOD In accordance to Section 191.12 (a) (1) and (2) of the Final Orders, Enforcement Regulations, Federal Register, Vol. 29, No. 182, P 13009, 17 September 1964.

Species/Strain	Rabbit/Albino
Number of Animals	6

Observation Period	72 hours
Remarks – Method	Ocular reactions were observed and recorded at 24, 48 and 72 hours.

RESULTS

Lesion	*Mean Score**	*Maximum Value*	*Maximum Duration of Any Effect*	*Maximum Value at End of Observation Period*
Conjunctiva: redness	0.33	1	24 hours	0
Conjunctiva: chemosis	0.06	1	24 hours	0
Conjunctiva:discharge	0	0	N/A	0
Corneal opacity	0	0	N/A	0
Iridial inflammation	0	0	N/A	0

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

Remarks – Results	The test material produced a very mild conjunctival effect in all of the animals (slight injection of the vessels) which cleared by the second day of observation.
CONCLUSION	The notified chemical is slightly irritating to the eye.
TEST FACILITY	FDR Labs (1971c)

7.6. Skin sensitisation

TEST SUBSTANCE	Notified chemical
METHOD	Guinea Pig sensitization study - Buehler Test. Study was conducted according to the regulations for Good Laboratory Practice as described by the FDA in 21 CFR Part 58, and FDRL Standard Operating Procedures.
Species/Strain	Hartley Derived Albino Guinea Pigs
PRELIMINARY STUDY	Not reported
MAIN STUDY	
Number of Animals	Test Group: 10 Control Group: 10
induction phase	topical application: test substance administered as supplied (100%)
Signs of Irritation	There were no reported signs of skin irritation following each induction application with the notified chemical. Mild to moderate erythema was observed in all positive control group animals.
CHALLENGE PHASE	
1st challenge	topical application (test group): test substance administered as supplied (100%)
Remarks – Method	Deviations from OECD TG 406 Skin Sensitisation – Buehler Test

- Only 10 test animals used.
- Positive rather than negative control group used. Positive control is 0.15% 1-chloro-2,4-dinitrobenzene in 80% ethanol.
- The three induction applications were made at different test areas.
- During induction, the sites were scored, using the Draize scale, at 24 and 48 hours following each application.
- Challenge patch applied for 24 hours.
- Skin reactions observed 1 hour and 24 hours after challenge patch removal.
- During challenge, positive control group animals also received 0.5mL of acetone.

RESULTS

Animal	*Challenge test substance and concentration*	*Number of Animals Showing Skin Reactions after:*			
		1st challenge		*2nd challenge*	
		1 h	*24 h*	*24 h*	*48 h*
Test Group	100% 2-Pyrrolidinone, 1-(2-hydroxyethyl)-	0	0	-	-
Positive Control Group	0.15% 1-chloro-2,4-dinitrobenzene in 80% ethanol.	10	10	-	-
	acetone	0	0	-	-

Remarks – Results: The dermal sensitisation potential of the test article and positive control material was determined by statistically comparing erythema scores obtained 24 and 28 hours after the first sensitising dose with those obtained after the challenge application. A statistically significant difference was obtained in the positive control group (mean erythema score increased from 0.8 to 1.6) but not in the test article group.

CONCLUSION: There was no evidence of reactions indicative of skin sensitisation to the notified chemical under the conditions of the test.

TEST FACILITY: FDR Labs (1981)

7.7. Repeat dose toxicity

TEST SUBSTANCE: 2-Pyrrolidone (Purity 99.7%)

METHOD: OECD TG 408 Repeated Dose 90-Day Oral Toxicity Study in Rodents.
EC Directive 88/302/EEC B.26 Sub-Chronic Oral Toxicity Test: 90-Day Repeated Oral Dose Study using Rodent Species.

Species/Strain: Rat/Wistar (Chubb:THOM (SPF))
Route of Administration: Oral – drinking water.
Exposure Information: Total exposure days: 90 days.
Dose regimen: daily
Post-exposure observation period: None
Vehicle: Drinking water
Remarks – Method: No significant protocol deviations.

Test solutions were analysed at the start and end of the study to assure that the concentrations were correct and the 4-day stability was assessed as 97%.

Study report not reviewed.

RESULTS

Group	*Number and Sex of Animals*	*Dose/Concentration*		*Mortality*
		Nominal (ppm)	*Actual (mean) (mg/kg bw)*	
I (control)	10 per sex	0	0	0
II (low dose)	10 per sex	600	37	0
III (mid dose 1)	10 per sex	2400	207	0
III (mid dose 2)	10 per sex	7200	586	0
IV (high dose)	10 per sex	15000	1125	0

Mortality and Time to Death
No mortality was observed during the study.

Clinical Observations
Decreased food consumption (unspecified) was observed in both group IV animals (both sexes) and group III females. Decreased water consumption (unspecified) was noted in both Group IV and Group III males and females. Decreased body weight gains were noted in both Group IV animals (9% reduction (males), 8% reduction (females)) and Group III animals (7% reduction (males), 6% reduction (females)) compared to controls. No effects were recorded for Group I and II animals.

Laboratory Findings – Clinical Chemistry, Haematology, Urinalysis
Clinical Chemistry
Decreased creatinine levels (unspecified) were observed in both group IV and Group II males and females. Decreased total protein levels (unspecified) were observed in Group IV males and females and Group II females. Decreased globulin and triglyceride levels (unspecified) were observed in group IV males and females. No effects were recorded for Group I and II animals.

Haematology
A prolonged (unspecified) prothombin time was noted in Group IV males and females. No effects were recorded for Group I, II and III animals.

Urinalysis
Increased (unspecified) urinary specific gravity (reduced urinary volume) and a dark discolouration of urine specimens were noted in both group IV and Group III males. No effects were recorded for Group I and II animals.

Effects in Organs
Organ Weights
A statistically significant increase in the mean relative kidney weights was noted in both Group III males (7.3% increase) and Group IV males (13.2% increase) and females (12.6% increase). A 6.3% increase (but not significant) in the mean relative kidney weights was noted in Group III females.

Histopathology.
A finding of "altered cellular composition of the thymic cortex" was reported in all dosed groups of females.

Remarks – Results
Individual results not reviewed.

The kidney appears to be the target organ.

A second 90-day study was conducted at 0, 50 and 15,000 ppm in drinking water using groups of five female rats to investigate the significance of this altered thymic cortex. It this second study the identical finding was present; however, it also occurred in controls. In addition, retrieval and examination of thymus slides from controls animals in other studies were examined and were also found to have the same "pathology". Therefore, this was considered incidental and not compound related.

CONCLUSION
The No Observed Adverse Effect Level NOAEL was established as 2400ppm in drinking water (207 mg/kg bw/day) in this study, based on the absence of any treatment related effects.

TEST FACILITY Toxicology and Regulatory Affairs, (2003).

7.8 Genotoxicity - bacteria

7.8.1 Genotoxicity – bacteria (a)

TEST SUBSTANCE Notified chemical (99.8% Purity)

METHOD OECD TG 471 Bacterial Reverse Mutation Test.

	EC Directive 2000/32/EC B.13/14 Mutagenicity – Reverse Mutation Test using Bacteria. Plate incorporation procedure (Test 1) and Pre incubation procedure (Test 2)
Species/Strain	*S. typhimurium*: TA1535, TA1537, TA98, TA100 *E. coli*: WP2uvrA
Metabolic Activation System	S9 fraction from Aroclor 1254 induced rat liver.
Concentration Range in Main Test	a) With metabolic activation: 0 - 5000 µg/plate b) Without metabolic activation: 0 - 5000 µg/plate
Vehicle	Water
Remarks - Method	No significant protocol deviations. The following positive controls (not listed in the protocol) were used in the absence of metabolic activation N-methyl-N'-nitro-N-nitrosoguanidine (TA 1535 and TA 100) 4-nitro-o-phenylendiamine (TA98)

RESULTS

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

Remarks - Results	No toxicity or precipitation was observed. The test substance did not cause a marked increase in the number of revertants per plate of any of the tester strains either in the presence or absence of metabolic activation. Negative controls were within historical limits. Positive controls confirmed the sensitivity of the test system.
CONCLUSION	The notified chemical was not mutagenic to bacteria under the conditions of the test.
TEST FACILITY	BASF (2004)

7.8.2 Genotoxicity – bacteria (b)

TEST SUBSTANCE	Notified chemical, 98.1% purity
METHOD	OECD TG 471 Bacterial Reverse Mutation Test. EC Directive 2000/32/EC B.13/14 Mutagenicity – Reverse Mutation Test using Bacteria.
Species/Strain	*S. typhimurium*: TA1535, TA1537, TA98, TA100. *E. coli*: WP2 uvrA.
Metabolic Activation System	Standard plate test (SPT) both with and without metabolic activation (Aroclor-induced rat liver S-9 mix)
Concentration Range in Main Test	20 µg – 5000 µg/plate (TA 1535, TA1537, TA98, TA 100, E.coli WP2 uvrA) 20 µg- 7500 µg/plate (TA1535, TA100)
Vehicle	Water

Remarks – Method

No significant protocol deviations.
The positive controls used were as follows:
With activation: 2-aminoanthracene (2-AA)
Without activation:
N-methyl-N'-nitro-N-nitrosoguanidine (MMNG) – TA 1535, TA 100
4-nitro-o-phenylenediamine (NOPD) – TA 98
9-aminoacridine (AAC) – TA 1537
4-nitroquinoline-N-oxide (4-NQO) – E. coli WP2 uvrA.

RESULTS

Metabolic Activation	*Test Substance Concentration (µg/plate) Resulting in:*			
	Cytotoxicity in PreliminaryTest	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Present				
Test 1	-	> 5000	> 5000	Yes for TA 1535 and TA 100 only.
Test 2	-	> 7500	> 7500	Yes (for both strains tested)
Absent				
Test 1	-	> 5000	> 5000	Yes for TA 1535 and TA 100 only.
Test 2	-	> 7500	> 7500	Yes (for both strains tested)

Remarks – Results

Tests without S-9 mix
TA1535: Mutagenicty was observed from about 500 µg/plate (factor 2.2-2.5) onward up to 7,500 µg/plate (factor 15.6).
TA100: Slight increase in the number of his^+ revertants over a dose range of about 2,500 µg – 7,500 µg/plate (factor 1.7 - 2.0).
TA 98, TA 1537, E. coli WP2uvrA: No increase in the number of his^+ or trp^+ revertants up to 5000 µg/plate.

Tests with S-9 mix
TA1535: Positive reaction from about 500 µg/plate (factor 2.9 – 3.2) onward with an increase in the number of mutant colonies by a factor of 20.5 at 7,500 µg/plate.
TA100:Slight increase in the number of his^+ revertants over a dose range of about 2,500 µg – 7,500 µg/plate (factor 1.4 – 3.4)
TA 98, TA 1537, E. coli WP2uvrA: No increase in the number of his^+ or trp^+ revertants up to 5000 µg/plate.

Toxicity
No bacteriotoxic effect (reduced his^- or trp^- background growth, decrease in the number of his^+ or trp^+ revertants, reduction in the titer) was observed.

Solubility
No test substance precipitation was found.

Positive controls
All positive controls showed a significant increase in revertants, consistent with historical controls.

According to the results of the study, the test substance led to a dose-dependent increase in the number of his+ revertants with strains TA1535 and TA100 both with and without S-9 mix in two experiments carried out independently of each other.

CONCLUSION	The notified chemical was mutagenic to bacteria under the conditions of the test.
TEST FACILITY	BASF (2001d)

7.8.3 Genotoxicity – bacteria (c)

TEST SUBSTANCE	Notified chemical (experimental samples, no analyses carried out). 1) HEP-Crude, 8301-132 –described as brown viscous liquid. 2) HEP 1st OH, 8301-132 – described as clear liquid. 3) HEP 2nd OH, 8301-132 – described as light brown viscous liquid.
METHOD	Not Specified
Species/Strain	*S. typhimurium*: TA1538, TA1535, TA1537, TA98, TA100.
Metabolic Activation System	Aroclor-induced rat liver S-9
Concentration Range in Main Test	a) With metabolic activation: 0.32 - 200 µL/plate. b) Without metabolic activation: 0.32 - 200 µL/plate.
Vehicle	DMSO
Remarks – Method	Summary only of method provided. Test 1 was carried out on sample 1). Test 2 was carried out on samples 2) and 3). Positive controls used were as follows: With activation: 2-Aminoanthracene for all strains. Without activation: 2-Nitrofluorene – TA 98, TA 1538 Sodium Azide – TA 100, TA 1535 9-Aminoacridine – TA 1537 Two replicate plates were used for each strain and concentration tested.

RESULTS

Metabolic Activation	*Test Substance Concentration (µL/plate) Resulting in:*			
	Cytotoxicity in Preliminary Test	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Present				
Test 1	-	> 200	> 200	Yes for TA 100 and TA 1535. Slight mutagenicity indicated for TA 98.
Test 2	-	≥ 8 for TA 1535 ≥ 200 for other strains.	> 200	Yes for sample 3) with TA 155. Slight mutagenicity indicated for sample 3) with TA 100.
Absent				
Test 1	-	> 200	> 200	Yes for TA 100 and TA 1535. Slight mutagenicity indicated for TA 98.
Test 2	-	≥ 1.6 with TA 1535. ≥ 200 with other strains.	> 200	Yes for sample 3) with TA 155. Slight mutagenicity indicated for sample 3) with TA 100.

Remarks – Results	In both assays all positive control chemicals elicited a positive response;

DMSO control backgrounds were acceptable.

In the assay of Sample 1) (HEP-Crude, 8301-132), mutagenic activity was evident in strains TA-100 and TA-1535 with and without metabolic activation. At the concentrations of 200, 40 and 8 µL/plate, the number of revertants observed exceeded double the control background in both strains. Only in TA-1535 did a doubling of the revertants occur at 1.6 µL/plate in both conditions and in the presence of metabolic activation at 0.32 µL/plate. Strain TA-98 showed slight mutagenic activity only at the two highest concentration tested (200 and 40 µL/plate). A dose response was obtained.

Mutagenic activity was not evident for Sample 2) (HEP 1st OH, 8301-132). The sample showed a toxic effect (sparse lawn) in all strains with and without metabolic activation at 200 µL/plate. Strain Ta 1535 showed slightly more toxicity at concentrations of 40, 8 and 1.6 µL/plate.

Sample 3 (HEP 2nd OH, 8301-132), tested in the same experiment as Sample 2 (HEP 1st OH, 8301-132), was toxic in all strains at the highest concentrations tested (200 µL/plate). A mutagenic response and some toxicity were seen in strain TA 1535 with metabolic activity at 40, 8 and 1.6 µL/plate and without metabolic activation at 40 and 8 µL/plate.

Lower than average numbers of revertants were seen in several strains and concentrations in Test 2 (samples 2 and 3), suggesting that some toxicity may have occurred at concentrations <200 µL/plate. This effect could have masked any further indications of slight mutagenicity.

CONCLUSION: Samples 1 and 3 were mutagenic to bacteria under the conditions of the test.

TEST FACILITY: Arthur D Little (1983)

7.8.4 Genotoxicity – bacteria (d)

TEST SUBSTANCE: Notified chemical (three samples). No analyses performed.
1) Sample received 22/12/81, accompanied by MSDS, which describes the material as an odourless, light yellow liquid.
2) Sample received 4/2/82, production lot sample, described as heavy brown liquid.
3) Sample received 1/3/82, purified sample (GAF 8522-10), white solid with melting point 26.8ºC.

METHOD: *Salmonella*/Mammalian-Microsomal Mutagenicity Testing (Ames Assay)
Method not specified.
Positive controls used were as follows:
2-Anthramine – all strains.
2-Nitrofluorene – TA 98, TA 1538.
Sodium azide – TA 100, TA 1535.
9-Aminoacridine – TA 1537.

Species/Strain	*S. typhimurium*: TA1538, TA1535, TA1537, TA98, TA100.
Metabolic Activation System	S-9
Concentration Range in Main Test	a) With metabolic activation: 0.32 - 200µL/plate. b) Without metabolic activation: 0.32 - 200 µL/plate.
Vehicle	DMSO
Remarks – Method	Three separate assays were conducted: Sample 1 – Test 1 Sample 2 – Test 2

Sample 3 – Test 3.

RESULTS

Metabolic Activation	*Test Substance Concentration (µL/plate) Resulting in:*			
	Cytotoxicity in PreliminaryTest	*Cytotoxicity in Main Test*	*Precipitation*	*Genotoxic Effect*
Present				
Test 1	-	≥ 40 for TA 100 ≥ 200 for other strains	-	Yes for TA 100 and TA 1535 at 40 µL/plate
Test 2	-	≥ 200	-	Yes for TA 100 at 40 µL/plate and TA 1535 at ≥ 1.6 µL/plate.
Test 3	-	> 200	-	Yes for TA 1535 at 200 µL/plate.
Absent				
Test 1	-	≥ 40 for TA 100 ≥ 200 for other strains	-	Yes for TA 100 and TA 1535 at 40 µL/plate
Test 2	-	≥ 200	-	Yes for TA 100 at 40 µL/plate and TA 1535 at ≥ 8 µL/plate.
Test 3	-	> 200	-	Yes for TA 1535 at 200 µL/plate.

Remarks – Results

Sample 1

In the first assay all positive control chemicals elicited a positive response. Slight toxicity was evident in one plate of TA-1537 and in one plate of TA-1535 treated with 2-anthramine in the presence of S-9. DMSO control backgrounds were acceptable.

N-(2-hydroxyethyl)-2-pyrrolidone gave a positive (mutagenic) response in TA-100 and TA-1535 even in the absence of metabolic activation. Toxicity (a sparse lawn) was evident at 200 µl/plate in all strains, and slight toxicity was noted in TA-100 at 40 µl/plate.

Sample 2

In the second assay slight toxicity was evident with 2-anthramine in strain TA-1535 in the presence of metabolic activation. All positive control chemicals elicited a positive response and DMSO controls were acceptable.

The second production lot of N-(2-hydroxyethyl)-2-pyrrolidone gave a positive (mutagenic) response in TA-100 and TA-1535 even in the absence of metabolic activation, confirming results obtained with the first lot of this material. Toxicity (a sparse lawn) was evident at 200 µl/plate in all strains and was also evident to some extent at all concentrations in TA-98 (i.e. there was a reduced number of revertants in contrast to the DMSO control plates).

Sample 3

In the third assay, all positive control chemicals elicited a positive response. Slight toxicity was evident with 2-anthramine in all strains in the presence of metabolic activation. DMSO control backgrounds were acceptable.

Mutagenic activity was evident for GAF 8522-10 (purified N-(2-

hydroxy-ethyl)-2-pyrrolidone) in TA-1535 both in the absence and presence of metabolic activation. A doubling of the number of revertants over the control background was observed only at the highest concentration tested (200 µl/plate). Toxicity was evident in TA-100 at 200 µl/plate (lawn effect) and to some extent at all the lower concentrations tested (number of revertants lower than the number obtained in the DMSO control backgrounds). It appears that GAF 8522-10 was not potent a mutagen as the production lots of N-(2-hydroxyethyl)-2-pyrrolidone, based on mutagenicity being evident in one strain rather than two, and on a lesser increase in the number of revertants over the controls. However it should be noted that toxicity may have masked slight mutagenic effects.

CONCLUSION The notified chemical was mutagenic to bacteria under the conditions of the test.

TEST FACILITY Arthur D Little (1982a).

7.9 Genotoxicity in vitro

7.9.1 Genotoxicity – in vitro Sister Chromatid Exchange Assay

TEST SUBSTANCE Notified chemical, technical grade, no analysis provided. Sample received from GAF Corporation 8/3/82, described in accompanying MSDS as odourless, light yellow liquid.

METHOD In vitro Sister Chromatid Exchange Assay in Mammalian Cells – in house method.

Species/Strain	Chinese Hamster
Cell Type/Cell Line	Ovary (CHO)/ K_1 subclone BH_4 cells
Metabolic Activation System	Not applicable
Vehicle	Not specified
Remarks - Method	Significant deviations from OECD TG 479 Genetic Toxicology: In vitro Sister Chromatid Exchange (SCE) Assay in Mammalian Cells:

- Cells exposed to the test substance only in the absence of metabolic activation
- Cells exposed for 24 hours

Cytotoxicity testing was carried out prior to this assay. As the chemical was not toxic, the highest concentration used in the assay was 1,000 µg/mL.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	125*, 250*, 500*, 1000*	24h	24h
Test 2	-	-	-

*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	>250	>1000	not reported	weak positive

Test 2	-	-	-	-

Remarks - Results

A slight, but statistically significant increase in the number of SCEs was observed in cells treated with 500ug/mL (11% increase, $p<0.05$) and 1000 ug/mL (13.5% increase, $p<0.025$) when compared with controls. The dose dependent effect observed suggests the result is biologically significant. The positive control (EMS) gave a >3-fold increase. The mitotic index was similar to the control, confirming the absence of cytotoxicity.

The mutagenicity of the test substance may be enhanced in the presence of metabolic activation.

CONCLUSION

The notified chemical was weakly clastogenic to Chinese Hamster Ovary (CHO) cells treated in vitro under the conditions of the test.

TEST FACILITY

Arthur D Little (1982b)

7.9.2 Genotoxicity – in vitro Chromosome Aberration Test

TEST SUBSTANCE

Notified chemical, technical grade, no analysis provided. Sample received from GAF Corporation 8/3/82, described in accompanying MSDS as odourless, light yellow liquid.

METHOD

In vitro Mammalian Chromosome Aberration Test – In house method

Species/Strain	Chinese Hamster
Cell Type/Cell Line	Ovary (CHO)/ K_1 subclone BH_4 cells
Metabolic Activation System	Not applicable
Vehicle	Not specified
Remarks - Method	Significant deviations from OECD TG 473 In vitro Mammalian Chromosome Aberration Test:

- Cells exposed to the test substance only in the absence of metabolic activation.
- Cells exposed only for 24 hours
- 100 metaphases (rather than 200) scored per concentration

Gaps were scored but were not included in the final calculations.

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	125*, 250*, 500*, 1000*	24h	24h
Test 2	-	-	-

*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	>250	>1000	not specified	negative
Test 2	-	-	-	-

Remarks - Results

Three chromosome aberrations (excluding gaps) were found in CHO cells treated with 125 ug/mL. Although this was a statistically significant ($p<0.05$) increase compared with controls, no aberrations were found in cells treated with higher concentrations, therefore this is not considered to be biologically significant. As gaps primarily occurred also at 125 ug/mL,

their inclusion in the calculations does not change the situation.
The positive control confirmed the sensitivity of the test system.
The mitotic indices were 54% to 81% controls, indicating low toxicity.

CONCLUSION The notified chemical was not clastogenic to Chinese Hamster Ovary (CHO) cells treated in vitro under the conditions of the test.

TEST FACILITY Arthur D Little (1982b)

7.9.3 Genotoxicity – in vitro Mammalian Cell Gene Mutation Test.

TEST SUBSTANCE Notified chemical, technical grade, no analysis provided. Sample received from GAF Corporation 8/3/82, described in accompanying MSDS as odourless, light yellow liquid.

METHOD In vitro Mammalian Cell Gene Mutation Test – in house

Species/Strain	Chinese Hamster
Cell Type/Cell Line	Ovary (CHO)/ K_1 subclone BH_4 cells
Metabolic Activation System	Not applicable
Vehicle	Not specified
Remarks - Method	Significant deviation from OECD TG 476 In vitro Mammalian Cell Gene Mutation Test:

- Cells exposed to the test substance only in the absence of metabolic activation.
- Cells exposed for 16 hours

Metabolic Activation	*Test Substance Concentration (µg/mL)*	*Exposure Period*	*Expression Time*	*Selection Time*
Absent				
Test 1	125*, 250*, 500*, 1000*	16h	24h	9 days
Test 2	-	-	-	-

*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	>250	>1000	Not reported	weak positive
Test 2	-	-	-	-

Remarks - Results The test substance induced a 2-3 fold increase in the number of mutations at the HGRPT locus compared with the untreated control. This increase, with the exception of cells treated with 500ug/mL was statistically significant ($p<0.05$). No dose dependent effect was observed. The positive control confirmed the sensitivity of the test system.

The mutagenicity of the test substance may be enhanced in the presence of metabolic activation.

CONCLUSION The notified chemical was weakly clastogenic to Chinese Hamster Ovary (CHO) cells treated in vitro under the conditions of the test.

TEST FACILITY Arthur D Little (1982b)

7.9.4 Genotoxicity – in vitro Mammalian Cell Transformation Assay

TEST SUBSTANCE	Notified chemical, technical grade, no analysis provided. Sample received from GAF Corporation 8/3/82, described in accompanying MSDS as odourless, light yellow liquid.
METHOD	In vitro Mammalian Cell Transformation Assay – in house.
Species/Strain	Mouse
Cell Type/Cell Line	Fibroblast/BALB/c-3T3
Metabolic Activation System	Not applicable
Vehicle	Not specified
Remarks - Method	Deviations from EC Directive 88/303/EEC B.21 Mutagenicity - In vitro Mammalian Cell Transformation Tests. • Cells exposed to the test substance only in the absence of metabolic activation. • Concentrations used should yield a concentration-related toxic effect. This was not possible as the notified chemical did not show cytotoxicity.

Metabolic Activation	*Test Substance Concentration (μg/mL)*	*Exposure Period*	*Harvest Time*
Absent			
Test 1	2*, 10*, 50*, 250*	3 days	4 weeks
Test 2	-	-	-

*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	>250	>250	not reported	negative
Test 2	-	-	-	-

Remarks - Results	No dose related increase in the number of type III foci/plate was observed. The positive control confirmed the sensitivity of the test system.
CONCLUSION	The notified chemical was not clastogenic to mouse fibroblast cells treated in vitro under the conditions of the test.
TEST FACILITY	Arthur D Little (1982b)

7.10. Genotoxicity – in vivo

TEST SUBSTANCE	2-Pyrrolidone (Purity 99.5%)
METHOD	OECD TG 474 Mammalian Erythrocyte Micronucleus Test.
Species/Strain	Mice/NMRI
Route of Administration	Intraperitoneal injection
Vehicle	Distilled water
Remarks - Method	Study report not reviewed. Deviations from OECD protocol. Samples of bone marrow were taken from animals dosed with 2000

mg/kg bw at 16, 24 an 48 hours. Only one sample of bone marrow was taken from animals dosed with 1000 mg/kg bw, 500 mg/kg bw and positive controls.

Two positive control substances were used, Cyclophosphamide and vincristine

Group	*Number and Sex of Animals*	*Dose mg/kg bw*	*Sacrifice Time hours*
I (vehicle control)	5 per sex	0	16, 24 and 48
II (low dose)	5 per sex	500	24
III (mid dose)	5 per sex	1000	24
IV (high dose)	5 per sex	2000	24
V (positive control, CP)	5 (2/3 per sex)	20	24
VI (positive control VC)	5 (2/3 per sex)	0.15	24

CP=cyclophosphamide. CV=vincristine

RESULTS

Doses Producing Toxicity

Irregular respiration, piloerection, abdominal position and apathy was noted in animals dosed with 2000 mg/kg bw. The general state of some of these animals was poor.

Animals treated with 10 and 500 mg/kg bw showed signs of irregular respiration and piloerection, 30 minutes after treatment. All clinical signs of toxicity had reversed 1 – 2 hours after administration.

Genotoxic Effects

Administration of test substance did not lead to an increase in the rate of micronuclei. The number of normochromatic erythrocytes (NCE) or polychromatic erythrocytes (PCE) containing small or large micronuclei did not deviate from the solvent control value at any sacrifice interval. No inhibition of erythropoiesis induced by the treatment of mice with the test substance was detected. The ration of PCE/NCE was always in the same range as that of the control values in all dose groups.

Remarks - Results

Individual results not reviewed.

The frequency of micronucleated PCE in the positive controls were higher (9 fold increase (CP), 55 fold increase (CV)) than the vehicle control.

CONCLUSION

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

TEST FACILITY

Toxicology and Regulatory Affairs (2003).

8. ENVIRONMENT

8.1. Environmental fate

8.1.1 (a) Ready biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	DOC-Reduction (Die-Away) Test in accordance with OECD Guideline 301 A, EU Guideline 92/69/EEC and ISO Standard 7827
Inoculum	Activated sludge from laboratory sewage treatment facility of pilot plant Z 570; 50% from units 1 and 3 (in 80:20 ratio) and 50% from unit 2. These units are run with municipal and synthetic wastewater.
Exposure Period	21 days
Auxiliary Solvent	none
Analytical Monitoring	DOC
Remarks – Method	The test substance at a test concentration of 17.5 mg/L (10.0 mg/L of DOC), a defined inorganic medium and the inoculum were incubated and aerated for up to 21 days. Duplicate samples on days 0, 1, 3, 6, 10, 14, 17, 20 and 21 were taken and analysed for DOC reduction. Aniline was used as the reference substance at 10.0 mg/L DOC. Elimination from the water was expressed as percent of DOC reduction at the end of the test compared to the initial concentration measured.

RESULTS

Test substance		*Reference substance (Aniline)*	
Day	*Mean % elimination*	*Day*	*Mean % elimination*
1	-12	1	1
3	-10	3	9
6	-9	6	87
10	13	10	96
14	41	14	98
21	105	21	99

Remarks – Results	The degree of degradation of the test substance was >70% at 21 days and the graph included shows the test met the criteria for >60% degradation within 10 days of 10% of degradation having been reached. Thus the notified chemical is considered to be ready biodegradable. The degradation of the reference control was >70% after 14 days, confirming the validity of the test.
CONCLUSION	The notified chemical is considered to be readily biodegradable.
TEST FACILITY	BASF (1995a).

8.1.1 (b) Inherent biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	Zahn-Wellens Static Test in accordance with OECD Guideline 302 B, EU Guideline 88/302/EEC, ISO Standard 9888 and German Industry Standard DIN EN 29 888.
Inoculum	Activated sludge from laboratory sewage treatment facility of pilot plant Z 570; 50% from units 1 and 3 (in 80:20 ratio) and 50% from unit 2. These units are run with municipal and synthetic wastewater.
Exposure Period	14 days

Auxiliary Solvent	none
Analytical Monitoring	DOC
Remarks – Method	The test substance at a test concentration of 179 mg/L (100 mg/L of DOC), a defined inorganic medium and the inoculum were stirred and aerated for up to 14 days. Duplicate samples on days 0, 0.125, 1, 3, 6, 7, 10, 13 and 14 were taken and analysed for DOC reduction. Aniline was used as the reference substance at 200 mg/L. Elimination from the water was expressed as percent of DOC reduction compared to the initial concentration measured. Biodegradability was expressed as percent reduction of DOC based on the value measured after 3 hours (absorbed part). The total organic carbon (TOC) was 555 mg/g and the dissolved organic carbon (DOC) was 558 mg/g.

RESULTS

Test substance			*Reference substance (Aniline)*	
Day	*Mean % elimination*		*Day*	*Mean % elimination*
	T1	*T2*		
1	0	0	1	-3
3	-1	1	3	1
6	82	65	6	98
10	98	98	10	98
14	98	97	14	99

Remarks – Results	In the short-term respiration test, there was no significant respiration inhibition up to 1000 mg/L. Degree of degradation (DOC Reduction) after 14 days was 90-100%. The degree of DOC elimination after 3 h: was 0-10%. The degradation of the reference control was >80% after 14 days, confirming the validity of the test.
CONCLUSION	The notified chemical is considered to be inherently biodegradable.
TEST FACILITY	BASF (1995b).

8.1.1. (c) Inherent biodegradability

TEST SUBSTANCE	Notified chemical
METHOD	Zahn-Wellens Static Test in accordance with EU Guidelines 88/32/EEC and OECD TG 302B ISO 9888-1991
Inoculum	Activated sludge from the BASF sewage treatment facility, not adapted, 0.5 g/L, dry basis.
Exposure Period	15 days
Auxiliary Solvent	None
Analytical Monitoring	DOC
Remarks – Method	The test substance at a test concentration of 179 mg/L (100 mg/L of DOC) and the inoculum were stirred and aerated in a static arrangement under standard conditions for up to 15 days. Duplicate samples on days 0, 0.125, 1, 3, 7, 14, 15 were taken and analysed for DOC reduction. Diethylene glycol was used as the reference substance. The reduction in DOC compared to the initial value was used as a measure of complete elimination from water.

RESULTS

Day	*Mean % elimination*

1	0
3	53
7	96
14	94
15	99

Remarks – Results	The DOC % elimination of the notified chemical after 15 days was 90-100%. DOC % elimination after 3 h (adsorption) was <10%. No results were obtained for the control substance and thus the validity of the test was not confirmed.
CONCLUSION	While the test is considered invalid in the absence of the control test results, it confirms the results above.
TEST FACILITY	BASF (1995c).

8.1.2. Bioaccumulation

No study was provided. The low Log Pow indicates that the bioaccumulation potential is likely to be low.

8.2. Ecotoxicological investigations

8.2.1. Acute toxicity to fish

TEST SUBSTANCE	2-Pyrrolidone (analogue)
METHOD	OECD TG 203 Fish, Acute Toxicity Test – 96 hour, static.
Species	*Brachydanio rerio* (Zebra fish; freshwater fish)
Exposure Period	96 hours
Auxiliary Solvent	None
Water Hardness	250 mg $CaCO_3$/L
Analytical Monitoring	Not reported
Remarks – Method	10 fish per treatment group was performed at nominal concentrations of 0, 50, 100, 1000, 2150, 4640 or 10,000 mg/L. Measured concentrations were close to the nominal concentrations (within 5%) at 1 and 96 h. Sub-lethal effects were observed daily for an exposure period of 96 h. Temperatures, dissolved oxygen and pH remained within acceptable limits during the test.
RESULTS	
LC50	6800 mg/L at 96 hours (nominal)
NOEC	4600 mg/L at 96 hours (nominal)
Remarks – Results	There was no mortality except at the high concentration of 10,000 mg/L where the cumulative mortality at 24 hours was 6/10, at 48 hours was 8/10 at 72 and 96 hours was 10/10. The effects observed were apathy and tumbling in surviving fish.
CONCLUSION	The analogue is considered to be very slightly toxic to *Brachydanio rerio* (Mensink et al 1995).
TEST FACILITY	Toxicology and Regulatory Affairs (2003)

The above result is from the robust summary in the IUCLID database. Consideration of the properties, particularly water solubility, indicate this is probably an acceptable analogue, despite the lack of alcohol functionality. ECOSAR v0.99g estimates a 96 h LC50 to fish of 75 g/L for the notified chemical.

The notifier has also provided aquatic toxicity test results conducted on the ink formulations. The ink formulation is a complex mixture of materials but typically consists of >70% water. The aquatic toxicity is primarily a function of the identity and amount of surfactant materials (typically <2%) present in formulations. The maximum concentration of the notified chemical present in the formulation is <14%. No test report was provided for the formulations tested, but pH, dissolved oxygen, temperature, hardness and alkalinity were summarised. The test was conducted for fathead minnows under the guideline "Static acute bioassay procedures for hazardous waste samples", California Department of Fish and Game, 1988. The test for rainbow trout was conducted under the guideline "Biological testing methods, static acute fish toxicity test" Hazardous waste Section, Washington State Dept of Ecology 1991.

For formulations at 100 mg/L, 100% of the rainbow trout survived the tests, indicating that the LC50 of the ink is likely to be >100 mg/L. It is not possible to clearly define the aquatic toxicity of the notified chemical from these tests, considering the presence of surfactants etc.

8.2.2. Acute toxicity to aquatic invertebrates

TEST SUBSTANCE	Notified chemical
METHOD	EEC Directive 79/831/EEC, Annex V, Part C: Methods for the determination of ecotoxicity, C2. Acute toxicity for *Daphnia*; updated Nov, 1989.
Species	*Daphnia magna*

Exposure Period	48 hours
Auxiliary Solvent	None
Water Hardness	220 – 320 mg $CaCO_3$/L
Analytical Monitoring	GC
Remarks – Method	Animals were considered to be immobile when unable to swim within 15 seconds after agitation of the test vessel. The toxic effects were observed visually after 0, 3, 6, 24 and 48 h. Temperatures, dissolved oxygen and pH remained within acceptable limits during the test.

RESULTS

Concentration mg/L Nominal	*Number of D. magna*	*Number Immobilised*	
		24 h	*48 h*
6.25	20	0	0
12.5	20	0	0
25	20	0	0
50	20	0	0
100	20	0	0

EC50	>100 mg/L at 48 hours
Remarks – Results	The test criterion of the acute toxicity is immobilization of the daphnids after 48 hours of exposure which is expressed as effect concentration (EC). None of the test organisms was immobilised after 48 h of exposure at concentration up to 100 mg/L. In the control, immobilisation was ≤10%. Thus, the validity criteria were fulfilled by the test.
CONCLUSION	The notified chemical is considered to be very slightly toxic to *Daphnia magna* (Mensink et al 1995).
TEST FACILITY	BASF (1995d)

8.2.3. Algal growth inhibition test

TEST SUBSTANCE	Notified chemical
METHOD	EEC Directive 79/831/EEC, Annex V, Part C; Algae: Growth inhibition test, updated May 1998.
Species	*Scenedesmus subspicatus* (green algae)
Exposure Period	72 hours
Concentration Range	
Nominal	0.39 mg/L – 100 mg/L
Auxiliary Solvent	None
Water Hardness	Not reported
Analytical Monitoring	GC with mass selective detection
Remarks – Method	Three replicates per each test concentration and three replicates for the control were used. In vivo chlorophyll-a fluorescence was measured after 0, 24, 48 and 72 h. Cell counts were performed after 72 h in a counting chamber. The EC values were calculated from the concentration-response curve.

RESULTS

Biomass		*Growth*	
Nominal EbC50 (mg/L) at 72 h	*Nominal NOEbC (mg/L) at 72 h*	*Nominal ErC50 (mg/L) at 72 h*	*Nominal NOErC (mg/L) at 72 h*
>100 mg/L	≥100 mg/L	>100 mg/L	≥100 mg/L

Remarks – Results	The 72 h LOEC of >100 mg/L is determined by comparing the means of the fluorescence measurement of the various concentrations with the control. The NOEC (72 h) is the test concentration immediately below the LOEC and is determined to be ≥100 mg/L. The validity criteria were maintained.
CONCLUSION	The notified chemical is considered to be very slightly toxic to algae (Mensink et al 1995).
TEST FACILITY	BASF (1995e)

8.2.4. (a) Inhibition of microbial activity

TEST SUBSTANCE	Notified chemical
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test EC Directive 88/302/EEC C.11 Biodegradation: Activated Sludge Respiration Inhibition Test ISO 8192-1986 (E) (Method B) German Industrial Standard DIN ISO 8192 (1994)
Inoculum	Active sludge from the laboratory sewage treatment facilities 1, 2 and 3 in the Z 570 pilot plant run with municipal and synthetic wastewater.
Exposure Period	30 min
Concentration Range Nominal	1-1000 mg/L
Remarks – Method	The respiration inhibition of activated sludge by the notified chemical was determined in the short-term respiration test (30 min). The test results gave the EC at which the respiration is inhibited at 20, 50 and 80% compared to a control value. EC20 is the limit concentration at which inhibition of the respiration of activated sludge can be expected in biological sewage treatment plants. The reference substance used in the test was 3, 5-dichlorophenol.
RESULTS	
Remarks – Results	EC50 >1000 mg/L. No significant respiration inhibition on the respiration of municipal activated sludge was observed up to the highest tested concentration of 1000 mg/L. As the deviation of reference values was <15% and the EC50 of 3, 5-dichlorophenol was in the range of 5-30 mg/L, the test was considered valid.
CONCLUSION	The notified chemical is considered not toxic to sewage micro-organisms. Note, however, the short 30 minutes period compared with the 3 h over which the test is normally conducted.
TEST FACILITY	BASF (1995f).

8.2.4. (b) Inhibition of microbial activity

TEST SUBSTANCE	1-(2-Hydroxyethyl) pyrrolidine-2-on EG (It is not clear what does EG stand for)
METHOD	OECD TG 209 Activated Sludge, Respiration Inhibition Test. EC Directive 88/302/EEC C.11 Biodegradation: Activated Sludge Respiration Inhibition Test. ISO 8192-1986 (E) (Method B)
Inoculum	Activated sludge from laboratory wastewater plant treating municipal sewage. Concentration of dry substance 1 g/L.
Exposure Period	30 min
Concentration Range	1-1000 mg/L

Nominal	
Remarks – Method	The respiration inhibition by the notified chemical was measured by the oxygen consumption rate of the activated sludge in the short-term respiration test (30 min). The test results gave the EC at which the respiration was inhibited at 20, 50 and 80% compared to a control value. EC20 is the limit concentration at which inhibition of the respiration of activated sludge can be expected in biological sewage treatment plants. The reference substance used in the test was 3, 5-dichlorophenol.
RESULTS	
EC50 (30 min)	>1000 mg/L (nominal)
Remarks – Results	The EC values are based on nominal concentrations. The respiration inhibition of 20, 50 and 80% were taken from the graph of the inhibition curve. Deviation of the blank control was <15% and the EC50 of 3, 5-dichlorophenol was within the range of 5-30 mg/L, confirming the validity of the test.
CONCLUSION	The notified chemical is considered to be not toxic sewage micro-organisms (but again a short test).
TEST FACILITY	BASF (2001c).

8.2.4.(c) Inhibition of microbial activity

TEST SUBSTANCE	Notified chemical
METHOD	DIN 38412 part 8 (March 1991)
Inoculum	The test strain of *Pseudomonas putida* DSM 50026 used is obtained in regular intervals from DSM (German Collection of Micro-organisms/Deutsche Sammlung von Mikroorgnanismen in Braunschweig) and kept on agar slants for further cultivation at BASF AG Ludwigshafen.
Exposure Period	16 hours
Concentration Range	39.06 – 10,000 mg/L
Nominal	
Remarks – Method	The inhibitory effect of the notified chemical on the cell multiplication of the bacterium *Pseudomonas putida* was investigated in a 16-hour static test. For the preparation of the stock solution (12,500 mg/L) the test substance was diluted in deionised water by stirring for about 5 min at 23°C. The bacteria were cultured under specific conditions. After the incubation time the optical density of the bacterial suspension was measured in a photometer at 436 nm. The measured optical density of the treated samples were compared to the untreated samples to show possible toxic effects.
RESULTS	
EC50 (16 h)	>10,000 mg/L
Remarks – Results	The test substance caused a slightly increased bacterial cell multiplication over the whole tested concentration range (max. 12.1% compared to an untreated control).
CONCLUSION	The notified chemical is not toxic to *Pseudomonas putida*.
TEST FACILITY	BASF (1995g)

9. RISK ASSESSMENT

9.1. Environment

9.1.1. Environment – exposure assessment

Environmental exposure of the notified chemical will result from the disposal of cartridges, printed paper and any leaked ink containing the chemical during the use of the cartridges. The total import volume of the notified chemical will ultimately be either disposed of to landfill, incinerated or recycled with paper.

It was not possible to confirm the vapour pressure results from the limited details provided. A calculation based on the MPBPWIN program yielded vapour pressures ranging from 0.019-0.64 Pa, indicating that the notified chemical is moderately volatile. It is water soluble and is expected to remain within the aquatic environment where loss to the atmosphere is unlikely to be significant. However, volatilization of the notified chemical from paper is possible and an OECD based calculation (OECD Environment Monographs 1992) was undertaken to determine the stability of the notified chemical in air as a result of its moderate volatility. The calculation indicates that the notified chemical will have a half life of 1.52 h with respect to its reaction with OH radicals in the air. Therefore, it is relatively unstable in air in the case of its release from printed paper. It will not readily hydrolyse in natural waters at environmental pH values. The low log Pow is consistent with the high water solubility indicating a low affinity for the organic phase and component of soils and sediments. It can be highly mobile in soil and although not expected to adsorb to organic matter in soil, will adhere to cellulose fibres on paper.

It is considered to be readily biodegradable. Thus it is anticipated that in an active landfill environment the notified chemical will degrade in the biotic processes. Incineration of waste paper and sludge will destroy the chemical with the generation of water vapour and oxides of carbon and nitrogen.

Recycling may take place in a number of centres throughout Australia. During the paper recycling process, waste paper is repulped using a variety of alkaline, dispersing and wetting agents, water emulsifiable organic solvents and bleaches. Trade sources estimate the washing process will recover 30-60% of the total amount of ink and therefore, at least 30% of the notified chemical in the recycled paper will be disposed of with sludge in landfill. However, a greater proportion can be expected to remain in the aqueous phase due to the high water solubility of the notified chemical.

A predicted environmental concentration (PEC) in the aquatic environment is estimated below using a worst-case scenario where the entire import volume (the maximum of 10 tonnes) of the notified chemical will be used on paper and 50% of the printed paper will be recycled with 60% of the chemical remaining in the aqueous phase during the recycling process. Under this scenario 3000 kg of the notified chemical per year will be discharged to sewer and if it is assumed based on high water solubility that none is attenuated within the sewage treatment plants (STP), the daily release on a nationwide basis to receiving waters is estimated to be 8.2 kg/day (Environment Australia 2003).

Assuming a national population of 20 million and that each person contributes an average 200 L/day to overall sewage flows, the worst-case predicted environmental concentration (PEC) in sewage effluent on a nationwide basis is estimated as 2.1 µg/L (Environment Australia 2003). Based on the respective dilution factors of 1 and 10 for inland and ocean discharges of effluents, the PECs of the notified chemical in freshwater and marine water may approximate 2.1 and 0.21 µg/L, respectively.

Due to the low log P_{ow} and the high water solubility of the notified chemical, its potential for bioaccumulation is low in exposed aquatic organisms.

9.1.2. Environment – effects assessment

The results of the aquatic toxicity tests are summarised below.

Organism	*Duration*	*End Point*	*mg/L*
Zebra fish (*Brachydanio rerio*)	96 h	LC50	>100 (based on the analogue)
Daphnia magna	48 h	EC50	>100
Algae (*Scenedesmus subspicatus*):	72 h	EbC50	>100
		NOEC	≥100
Sewage micro-organisms	30 min	EC50	>1000

A predicted no effect concentration (PNEC - aquatic ecosystems) of >1000 µg/L has been derived by dividing the end point value of >100 mg/L for *Daphnia magna* by a worst-case scenario uncertainty (safety) factor of 100 as toxicity data are available for the three trophic levels.

9.1.3. Environment – risk characterisation

Location	PEC µg/L	PNEC µg/L	Risk Quotient (RQ)
Australia-wide STPs			
Ocean outfall	0.21	>100	$<2.1 \times 10^{-4}$
Inland River	2.1	>100	$<2.1 \times 10^{-3}$

The RQ values (PEC/PNEC) derived for the aquatic environment (assuming nationwide use, only 50% of the printed paper recycled and 60% of the notified chemical partitioned to water in STP) are considerably below 1 for both freshwater and marine water, indicating no immediate concern to the aquatic compartment. Based on the proposed use pattern the notified chemical is not expected to pose an unacceptable risk to the health of aquatic life. Bioaccumulation is not expected from the diffuse use pattern and low import volume.

9.2. Human health

9.2.1. Occupational health and safety – exposure assessment

Exposure to the notified chemical during transport and storage is expected to be very low, as the cartridges carrying ink containing the notified chemical are imported as sealed units. Accidental exposure through breach of packaging is possible but expected to be infrequent.

Workers may be exposed to the notified chemical through dermal contact and inhalation while changing spent cartridges, repairing printers or during normal printing processes. Service technicians are expected to have the highest occupational exposure, because they are likely to work with the printers for up to 8 h/day.

For all workers, dermal exposure is likely to occur only occasionally, and to be avoided because it would stain the skin and/or smudge the printed page.

Inhalation exposure can occur during normal printing processes to vapour or aerosol of the notified chemical, one of the solvents in the ink. The notifier has advised that 0.4 g (400 mg) of ink is used for each printed page, an estimate that may be at the high end of the usage range. As the ink contains approximately 10% of the notified chemical, there is potential exposure to 40 mg of the notified chemical when each page is printed. The actual exposure is likely to be considerably lower than this, as normal air circulation would disperse such small quantities quickly, and some of the chemical would be trapped permanently or temporarily in the printed paper. Exposure of workers can vary depending on the type of ventilation, the amount of printing carried out in a short time, and the tendency of the chemical to be trapped in the paper. Two different estimates of exposure are below:

1) The notifier has estimated that 0.37 mg of the notified chemical in total may be released from one printed page in the first 15 minutes after printing, as vapour or aerosol, with the remainder trapped in the dried ink and paper. This estimate of 0.36 mg evaporated is based on the total

amount of ink evaporated in 15 minutes and the partial pressures of the volatile solvents in the ink. The estimation of 0.016 mg released as aerosol is based on 1% of the ink being aerosolised, and 20% of this 1% escaping the printer enclosure.

2) If it is assumed that 10% of the notified chemical becomes airborne after a page is printed, a higher estimate of 4 mg released per page is reached. If this quantity was released into $1m^3$ of air as vapour and aerosol, the concentration in air would be close to 1 ppm ($5.3 mg/m^3$).
This concentration would be increased if:

- Evaporation from multiple printed pages was occurring at the same time.
- More than 10% of the notified chemical evaporated from the ink immediately after printing.

This concentration would be decreased if:

- The ink evaporated into a larger area than 1 m^3.

Less than 10% of the notified chemical evaporates during the drying of the printed page (as suggested by the notifier)

It should also be noted that the total amount of notified chemical available for human exposure is limited by the maximum amount of ink in the small cartridge, although several colour ink cartridges may be used in the one printer. The use of one cartridge is likely to occur over a period of time. Therefore exposure is likely to be episodic rather than continuous. Overall the exposure from the notified chemical in air immediately after printing could vary with a number of factors.

If further volatile components of the ink (including the notified chemical) continue to be released slowly into the air after the ink has dried, this could lead to further inhalation exposure, albeit at a low level. However the notifier states that after drying the notified chemical is captured within the paper.

As a scenario tending towards the worst case, the proportion of notified chemical released during printing could be estimated at 20% (rather than 10% as above or 1% as estimated by the notifier) ie 8 mg/page, and exposure estimated to be 20 pages a day. Therefore exposure/day would be 160 mg/day or 2.3 mg/kg/day for a 70 kg worker.

Good ventilation and PPE are recommended on the MSDS for inks containing the notified chemical, but PPE may not be used for routine printing.

9.2.2. Public health – exposure assessment

Similarly to office workers, the public may be intermittently exposed to the notified chemical when replacing spent cartridges, and during use of printers. Dermal exposure to ink containing the notified chemical could occur accidentally but would be avoided because skin staining and/or smudging of the printed page. Inhalation exposure could also occur, but is expected to be episodic and limited by the small number of pages printed in a day.

Exposure to very low levels of the notified chemical from printed paper could occur, because some evaporation would be expected after the initial drying process.

Overall, exposure of the public is likely to be limited by the small quantity of notified chemical in each cartridge, the controlled release during printing, relatively low vapour pressure and intermittent nature of exposure.

9.2.3. Human health – effects assessment

General comments
Test results on the notified chemical were available for several endpoints, although most tests were carried out more than twenty years ago and do not conform to current testing protocols. The analogue 2-pyrrolidone (CAS 616-45-5) was considered suitable for the remaining endpoints on the basis of similarly low acute oral toxicity and similarity in physicochemical properties such as the partition coefficient. The results on this analogue for acute dermal and

inhalation toxicity, repeated dose toxicity and in vivo genotoxicity are included in this report, and it has been used as the basis for the NOAEL calculation.

However the possibility of similar toxicity to other analogues cannot be ruled out for endpoints where the notified chemical itself has not been tested. N-methyl pyrrolidone (CAS 872-50-4) and N-vinyl pyrrolidone (CAS 88-12-0) are more toxic analogues of the notified chemical for which substantial toxicological data is available [IPCS (2001), NICNAS (2000)]. Both are classified as hazardous substances under the NOHSC criteria, and show toxicological effects not seen in 2-pyrrolidone.

Ansell and Fowler (1988) compared the acute oral toxicity, dermal irritation and eye irritation properties of a range of N-alkyl-2-pyrrolidones, noting that the toxicity varied according to the alkyl substituent. Acute oral toxicity decreased with increasing hydrophilicity, with a high LD_{50} for the notified chemical. The authors also noted that effects on the central nervous system were found at high but sublethal doses, and suggested that this effect is a property of the class of chemical.

There is a strong suggestion that the toxicity of the notified chemical may be partially based on the identity and concentration of impurities. Analytical data was not available for each of the studies submitted on the notified chemical, but was available for some studies. The identity and concentration of impurities varied in these samples. Of four reverse mutation bacterial tests carried out, only the most recent, using material of 99.8% purity, gave negative results (BASF, 2004). Positive mutagenicity results were obtained on a sample of 98.1% purity (BASF, 2001d), and non-analysed samples that are not expected to be of high purity (Arthur D Little, 1983, 1982a).

Individual endpoints
No information on toxicokinetics, metabolism and distribution was supplied for the notified chemical. However both the notified chemical and some analogues have been investigated as penetration enhancers for topically applied drugs, and may therefore be absorbed dermally.

The notified chemical is of low acute oral toxicity. Acute dermal toxicity is expected to be low, based on the absence of reported effects in the skin irritation test, and low toxicity of the analogue 2-pyrrolidone. Limited information on acute inhalation toxicity of the notified chemical and 2-pyrrolidone suggests low toxicity via this route also, however only a low concentration was tested. The notified chemical is non-irritant to skin and a slight eye irritant in rabbits. It was not a skin sensitiser in a Buehler test.

A NOAEL of 207 mg/kg bw/day was determined for the analogue 2-pyrrolidone on the basis of a 90-day drinking water study in rats. No mortality occurred in the study, which used doses up to 1125 mg/kg. The target organ was the kidney, with alterations in urinary parameters and increase in kidney weight at the two top dosages (586 and 1125 mg/kg bw/day).

Mutagenic effects in some strains of bacteria were found in 3 of 4 reverse mutation tests. The most recent study on high purity material gave negative results. A battery of in vitro genotoxicity tests conducted on the notified chemical in the absence of metabolic activation produced some weakly positive results and some negative results. An in vivo micronucleus test on the analogue 2-pyrrolidone was negative.

No testing was carried out for reproductive toxicity or carcinogenicity.

Based on the available data, the notified chemical is not classified as a hazardous substance in accordance with the NOHSC *Approved Criteria for Classifying Hazardous Substances* (NOHSC 2004).

9.2.4. Occupational health and safety – risk characterisation

The notified chemical is not classified as a hazardous substance, based on animal studies on the notified chemical and on the analogue 2-pyrrolidone. Mutagenicity of the notified chemical appears to be linked to its purity, with a negative response on a sample of 99.8% purity, and

positive responses on a sample of 98.1% purity and earlier samples expected to be of lower purity. The current specification for the notified chemical's purity is ≥ 99.0%, therefore it cannot be assumed that all production material will have the same purity as that which was negative in the mutagenicity test.

Other areas of uncertainty in characterisation of the health effects of the chemical result from the fact that test reports for several endpoints do not meet current test protocols, and that analogue data has been used for acute dermal and inhalation toxicity, repeat dose toxicity, and in vivo genotoxicity endpoints.

The MSDS for inks containing the notified chemical recommends use of PPE in case of accidental release, and if needed for normal use, and recommends that the ink be used in a well ventilated area. If implemented, these recommendations would reduce exposure and potential risk.

The NOAEL established for the analogue via the oral route is 207 mg/kg/day for repeated exposure. From the toxicological testing carried out on the analogue and the notified chemical, it is likely that this testing would also be relevant for dermal and inhalation exposure. As estimated worker exposure is up to 2.3 mg/kg/day, there is a margin of safety of approximately 100. Acute effects are not expected to occur. Occupational risk is therefore estimated to be low, assuming that production controls are sufficient to rule out mutagenicity from low purity material.

9.2.5. Public health – risk characterisation

On the available data the notified chemical is not classified as a hazardous substance. Areas of uncertainty are the relationship between purity and mutagenicity, use of analogue data, and outdated test protocols used for some endpoints.

It is noted that the label for the ink cartridges recommends that it should be kept out of reach of children, and that the cartridge is intended for single use only. Both these warnings should improve public safety. An additional precaution would be to recommend on the label that the printer be used with good ventilation (as the MSDS would not be available to the public).

It is possible that home printing may be carried out by children, with their lower body weight leading to a lower margin of safety for repeated exposure than that calculated for occupational exposure (100). However the use pattern of the cartridges by the public is expected to be intermittent rather than continuous, and overall exposure is likely to be lower than for occupational use. Based on the expected low exposure to the public, and the known health effects, the risk to the public through use of printing cartridges is expected 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 not classified as hazardous under the NOHSC *Approved Criteria for Classifying Hazardous Substances*.

and

As a comparison only, the classification of 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.
The notified chemical is not classified under the Globally Harmonised System of Classification and Labelling of Chemicals (GHS) for both health and environmental hazards.

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.

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 in printing cartridges as described.

11. MATERIAL SAFETY DATA SHEET

11.1. Material Safety Data Sheet

The MSDS of a product 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 a product 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

CONTROL MEASURES

Occupational Health and Safety

- The manufacturer of the ink products should monitor the purity of the notified chemical used, to ensure that mutagenic effects do not occur.
- Employers should implement the following engineering controls to minimise occupational exposure to the notified chemical in the ink products:
  - Use of good natural or mechanical ventilation in the vicinity of printers.
- Employers should ensure that the following personal protective equipment is used by workers to minimise occupational exposure to the notified chemical in the ink products:
  - Gloves, if dermal exposure is likely
  - Respiratory protection, if significant inhalation exposure is likely.

  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.

Public Health

- The following measure should be taken by the supplier of the inks containing the notified chemical, to minimise public exposure:
  - Inclusion of the following statement on the ink cartridge labels: "Use in a well ventilated area".

Disposal

- The notified chemical should be disposed of by incineration or to landfill.

Emergency procedures

- Spills/release of the notified chemical should be handled by soaking up with absorbent material. Slowly vacuum or sweep the material into bag or other sealed container. Dispose of in compliance with federal, state and local 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 Section 64(1) of the Act; if
  - Any additional use is proposed, other than as a component of printing ink in cartridges containing a maximum of 100 g.
  - The concentration of notified chemical in printing inks exceeds 20%

or

(2) 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.

13. BIBLIOGRAPHY

Ansell J M & Fowler J A (1988) The acute oral toxicity and primary ocular and dermal irritation of selected *N*-alkyl –2-pyrrolidones. Fd. Chem. Toxic. Vol 26, No. 5, pp 475-479.

Arthur D. Little (1982a) Mutagenicity testing (Ames Assays) of three samples of N-(2-Hydroxyethyl)-2-pyrrolidone. ADL Reference: 73260-65. Report for GAF Corporation, New Jersey 19/7/82. Arthur D Little, Inc, Acorn Park, Cambridge, Massachusetts (Unpublished report provided by notifier).

Arthur D. Little (1982b) Evaluation of N-(2-Hydroxyethyl)-2-pyrrolidone in four mammalian short-term assays. ADL Reference: 87738. Report to GAF Corporation, New Jersey 26/5/82. Arthur D Little, Inc, Acorn Park, Cambridge, Massachusetts. (Report was submitted to USEPA).

Arthur D. Little (1983) Mutagenicity Testing (Ames Assays) of Three HEP Samples (HEP-Crude, 8301-132, HEP 1st OH, 8301-132, HEP 2nd OH, 8301-132). ADL Reference: 73260-66. Report for GAF Corporation, New Jersey 23/2/83. Arthur D Little, Inc, Cambridge, Massachusetts. (Unpublished report provided by notifier).

Australian Environmental Review (2001). On track for best ever recycling rates.

BASF (1985) Vapour pressure: N-(2-Hydroxyethyl)-pyrrolidone-2. Report BRU 85.160 Sept 25, 185 (2pp). BASF ZAM/P. (Unpublished report provided by notifier).

BASF (1992) Safety engineering characteristics: N-(2-Hydroxyethyl)-pyrrolidone (HEP) Enclosure to memorandum BASF ZET/ES 91/1624. Jan 2, 1992. BASF Technical Development /Process Engineering (Unpublished report provided by notifier).

BASF (1995a) Final report — Determination of the biodegradability of Hydroxyethylpyrrolidon by the DOC reduction (Die-Away) test. Project Number: 95/0601/21/1. BASF Aktiengesellschaft, Emission Surveillance and Ecology, Laboratory for Microbiology, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1995b) Final report — Determination of the biodegradability of hydroxyethylpyrrolidone by the Zahn-Wellens Static Test. Project Number: 95/0601/10/1. BASF Aktiengesellschaft, Emission Surveillance and Ecology, Laboratory for Microbiology, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1995c) Test report — Determination of the biodegradability or of the elimination of hydroxyethylpyrrolidone by the Zahn-Wellens Static Test. Project Number: 95/0601/10/2. BASF Aktiengesellschaft, Emission Surveillance and Ecology, Laboratory for Microbiology, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1995d) Determination of the acute toxicity of Hydroxyethylpyrrolidon to the water flea *Daphnia magna* STRAUS. Project Number: 95/0601/50/1. BASF Aktiengesellschaft, Ökologie und Umweltanalytik, Labor für Ökotoxikologie, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1995e) Determination of the inhibitory effect of Hydroxyethylpyrrolidon on the cell division of the green alga *Scenedesmus subspicatus*. Project Number: 95/0601/60/1. BASF Aktiengesellschaft, Ökologie und Umweltanalytik, Labor für Ökotoxikologie, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1995f) Final report — Determination of respiration inhibition of activated sludge by hydroxyethylpyrrolidone in the short-term respiration test. Project Number: 95/0601/08/1. BASF Aktiengesellschaft, Emission Surveillance and Ecology, Laboratory for Microbiology, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1995g) Determination of the inhibitory effect of Hydroxyethylpyrrolidon on the cell multiplication of the bacterium *Pseudomonas putida* . Project Number: 95/0601/70/1. BASF Aktiengesellschaft, Ökologie und Umweltanalytik, Labor für Ökotoxikologie, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (1997a) Technical Data Sheet: 1-(2-Hydroxyethyl)-2-Pyrrolidone. Chemical Intermediates BASF Corporation, Chemicals Division, New Jersey.

BASF (1997b) N-(2-hydroxyethyl)-2-pyrrolidone: n-octanol/water partition coefficient. Report CZA/B-O 619. 20/1/97. BASF Co. Ludwigshafen. (Unpublished report provided by notifier).

BASF (2001a) Charakteriserung von Hydroxyethylpyrrolidon AL.Nr. 13-0963 für toxikologische Studien ZH/T Nr.: 01/0108/1. BASF Aktiengesellschaft, Ludwigshafen. Report dated 12/3/2001 (Unpublished report provided by notifier).

BASF (2001b) Technical Leaflet (Hydroxyethyl-pyrrolidone) Electronic Grade. BASF Aktiengesellschaft, Global Business Management ,Electronic Chemicals, Ludwigshafen, Germany. Accessed at http://www.basf.de/en/produkte/chemikalien/anorganika/elektronik/products/solvents/?id=hzo_I6.0Zbcp.1c 8/12/04.

BASF (2001c) Report 1-(2-Hydroxyethyl) pyrrolidine-2-on EG – Determination of the inhibition of oxygen consumption by activated sludge in the activated sludge respiration inhibition test. Project Number: 01/0109/08/1. BASF Aktiengesellschaft, Product Safety, Regulations, Toxicology and Ecology, D-67056 Ludwigshafen, FRG. (Unpublished report provided by notifier).

BASF (2001d) *Salmonella typhimurium* / *Escherichia coli* Reverse Mutation Assay (Standard Plate Test) with Hydroxyethylpyrrolidon AL Nr. 13-0963. Laboratory Project No. 40M0109/014008, study completed 16/8/01. BASF Aktiengesellschaft, Product Safety, Regulations, Toxicology and Ecology, D-67056 Ludwigshafen. (Unpublished report provided by notifier).

BASF (2003) Material Safety Data Sheet: N-(2-hydroxyethyl)-2-pyrrolidone. Original date 4/12/92. Revised 2/5/03. BASF Corporation, Mount Olive, NJ. Accessed from internet 20/10/04

BASF (2004) *Salmonella typhimurium* / *Escherichia coli* Reverse Mutation Assay (Standard plate test and Preincubation test) with N-(2-Hydroxyethyl)-2-pyrrolidon. Test substance batch 55805068E0. Laboratory Project No. 40M0443/044101, study completed 21/10/04. BASF Aktiengesellschaft, Experimental Toxicology and Ecology, Ludwigshafen. (Unpublished report provided by notifier).

Environment Australia (2003) Model and Guidance for Estimating Predicted Environmental Concentrations to Surface Water and Soil from Chemicals Released to the Environment Through a Sewage Treatment Plant. Chemical Assessment Section, Environment Australia, Canberra Australia.

FDR Labs (1971a) Acute Oral Toxicity Study (Test Substance: Hydroxyethyl pyrrolidone 6818-228-4). I.B.L. No: 9063. Report dated 15/7/71. Study for GAF Corporation, New York. Food and Drug Research Laboratories Incorporated, New York. (Unpublished report provided by notifier).

FDR Labs (1971b) Primary skin irritation study (Test substance: Hydroxyethyl pyrrolidone 6818-228-4). I.B.L. No: 9063. Report dated 15/7//71, Study for GAF Corporation, New York. Food and Drug Research Laboratories Incorporated, New York. (Unpublished report provided by notifier).

FDR Labs (1971c) Rabbit Eye Irritation Study (Test Substance: Hydroxyethyl pyrrolidone 6818-228-4). I.B.L. No: 9063. Report dated 15/7/71, Study for GAF Corporation, New York. Food and Drug Research Laboratories Incorporated, New York. (Unpublished report provided by notifier).

FDR Labs (1981) Guinea Pig Sensitization Study: Buehler Test for GAF Corporation: Test Article Hydroxyethyl Pyrrolidone. FDRL Study No: 7003. Report dated 14/9/81. Food and Drug Research Laboratories Incorporated, New York. (Unpublished report provided by notifier).

IPCS (2001) Concise International Assessment Document No. 35: *N*-Methyl-2-pyrrolidone. World Health Organization, Geneva.

M B Research Laboratories (1978) Report on Oral LD 50 in Rats: Sample number 7701-62-6. Project No: MB 78-2478. Study for GAF Corporation. Report 29/3/78. MB Research Laboratories, Inc., Spinnerstown, Pennsylvania. (Unpublished report provided by notifier).

Mensink BJWG, Montforts M, Wijkhuizen-Maslankiewicz L, Tibosch H and Linders JBHJ (1995) Report no. 679101022: Manual for Summarising and Evaluating the Environmental Aspects of Pesticides. National Institute of Public Health and Environmental Protection, Bilthoven, The Netherlands.

NICNAS (2000) N-vinyl-2-pyrrolidone (NVP): Priority Existing Chemical Assessment Report No. 11. Commonwealth of Australia: Canberra.

NIST (2004a) National Institute of Standards and Technology (NIST) Chemistry Webbook. N-(2-Hydroxyethyl)-2-pyrrolidone. IR spectrum – Gas Phase. Coblentz Society No. 06702. Accessed at http://webbook.nist.gov./cgi 22/7/04

NIST (2004b) National Institute of Standards and Technology (NIST) Chemistry Webbook. N-(2-Hydroxyethyl)-2-pyrrolidone. Mass spectrum. Data compiled by NIST Mass Spec Data Center. Accessed at http://webbook.nist.gov./cgi 22/7/04

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

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

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

OECD Environment Monographs (1992) The rate of photochemical transformation of gaseous organic compounds in air under tropospheric conditions. OECD Environment Monographs No. 61. OCDE/GD (92)172.

Toxicology and Regulatory Affairs (2003) IUCLID dataset for 2-Pyrrolidone (CAS No. 616-45-5), August 2003, prepared by Toxicology and Regulatory Affairs, Freeburg, Illinois. Participating Members of Consortium: BASF Corporation and International Specialty Products. (IUCLID supplied by notifier).

United Nations (2003) Globally Harmonised System of Classification and Labelling of Chemicals (GHS). United Nations Economic Commission for Europe (UN/ECE), New York and Geneva.