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NATIONAL INDUSTRIAL CHEMICALS NOTIFICATION AND ASSESSMENT SCHEME

FULL PUBLIC REPORT

Dye for BJ Printer Ink 001

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Director
Chemicals Notification and Assessment

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FULL PUBLIC REPORT

Dye for BJ Printer Ink 001

1. APPLICANT

Canon Australia Pty Ltd of 1 Thomas Holt Drive North Ryde NSW 2113 (ACN 005 002 951) has submitted a standard notification statement in support of their application for an assessment certificate for the Dye for BJ Printer Ink 001.

2. IDENTITY OF THE CHEMICAL

The chemical name, CAS number, molecular and structural formulae, molecular weight, methods of detection and determination, spectral data, composition of the chemical, use and exact import volume have been exempted from publication in the Full Public Report and the Summary Report.

Marketing names: Dye for BJ Printer Ink 001

3. PHYSICAL AND CHEMICAL PROPERTIES

The physico-chemical parameters were determined in accordance with OECD Principles of Good Laboratory Practice, EC Commission Directive, and UK Good Laboratory Practice Regulations Test Guidelines (ASG, 1999).

Appearance at 20°C & 101.3 kPa: Black powder

Melting Point: >350°C

Specific Gravity: 1.52

Vapour Pressure: $<1 \times 10^{-6}$ kPa at 25°C

Water Solubility: 28.2% w/w at 25°C

Partition Co-efficient (n-octanol/water): log P_{ow} = <-2.95 at 25°C

Hydrolysis as a Function of pH:	$T_{1/2}$ at pH 4.0 = not determined due to low solubility $T_{1/2}$ at pH 7.0 = >115 hours $T_{1/2}$ at pH 9.0 = >115 hours
Adsorption/Desorption:	$\log_{10} K_{oc}$ = <1.5
Dissociation Constant:	Not determined, expected to dissociate at pH greater than 7
Flash Point:	Not applicable
Flammability Limits:	Not flammable
Autoignition Temperature:	230°C
Explosive Properties:	Not explosive
Reactivity/Stability:	Not oxidising
Surface tension:	71.37mNm^{-1}

Comments on Physico-Chemical Properties

Melting point was determined by the capillary tube method, using a block heater (ASG, 1999). The boiling point was unable to be determined because the notified chemical decomposed upon melting.

Relative density was determined in accordance with EC Method A3 using a dilatometric procedure in which a suitable liquid is used to determine the true volume of a given test mass of the test substance in a pyncnometer (ASG, 1999).

Vapour pressure was determined using an effusion manometry method, which is an approved method for EC Method A4 (ASG, 1999). The vapour pressure of the notified chemical was too low for measurement in the preferred measurement range of 0-50°C. Therefore, two measurements were made at 111.1 and 139.4°C respectively. The notified chemical is considered to be very slightly volatile (Mensink et al, 1995).

Water solubility was determined in accordance with EC Method A6 using the flask method (ASG, 1999). The notified chemical is considered to be very soluble in water.

The hydrolysis potential was determined in accordance with EC Method C7 (ASG, 1999). Hydrolysis of the notified chemical could not be determined at pH 4 because precipitation was observed at this pH and it proved impossible to produce a suitable solution for the determination of hydrolysis. At pH 7.0 and 9.0, the hydrolysis was found to be less than 1% after 115 hours at 50 ± 0.5°C. The hydrolysis potential of the notified chemical is considered to be limited and it is not expected to undergo significant abiotic degradation under environmental pH.

The partition coefficient was determined in accordance with EC Method A8 using the shaking flask method followed by UV/Vis spectrophotometric determination (ASG, 1999). The notified chemical is thus considered to be strongly hydrophilic and will preferentially partition into the aqueous phase.

Adsorption/desorption was determined in accordance with Draft OECD Guideline for the Testing of Chemicals (May 1997) using HPLC (ASG, 1999). The notified chemical is classified as mobile in soils.

The dissociation constant for the notified chemical was not determined. The notifier expects it to dissociate at pH >7 but as the sulfonate group is a strong acid, it is more likely to dissociate at ~ pH 4.

4. PURITY OF THE CHEMICAL

Degree of Purity: high

5. USE, VOLUME AND FORMULATION

The notified chemical will not be manufactured, formulated or repackaged in Australia. It will be imported in 15 mL pre-packed printing ink cartridge containing 5% notified chemical.

The import volume of the notified chemical is estimated at a maximum of 280 kg/annum for the first 5 years.

6. OCCUPATIONAL EXPOSURE

Office workers and printer maintenance workers may be intermittently exposed to the notified chemical contained in the ink cartridge when replacing the spent ink cartridge, and during repair maintenance and cleaning of ink jet printers. Maintenance workers for printers may potentially come in contact with the notified chemical more often than office workers. Exposure is expected to be controlled through the design of the ink cartridges and the printing machines. Printer maintenance personnel often wear cotton disposable gloves. Pre-packed ink cartridges are sealed and worker exposure to the ink is minimised by the use of the replacement procedures recommended by the manufacturer.

Waterside, warehouse and transport workers are unlikely to be exposed to the notified chemical unless the packaging is breached.

Contact with paper printed with printing inks containing the notified chemical is unlikely to result in dermal exposure, as it will be bound in the structure of the paper.

7. PUBLIC EXPOSURE

Exposure of the public as a result of transport and disposal of the ink products containing the notified chemical is assessed as negligible. Ink products containing the notified chemical are fully contained within inkjet cartridges and are inserted directly into inkjet printers. Public exposure may result from contact with ink residues deposited within the printer particularly during cartridge replacement or printer maintenance. Trace amounts of the ink may be lifted off the printed page during handling; therefore, dermal contact with ink deposited onto paper is possible. However, public exposure via this route is expected to be low.

8. ENVIRONMENTAL EXPOSURE

8.1 Release

Baumann et al, (1999) have described the typical life cycles including discharge of substances used in the printing and publishing industries. Ink cartridges are not used within the major printing processes.

Transport
Losses during transport will be minimal as the notified chemical is housed in sealed cartridges. Accidental spillage of the chemical, either during transport or use should result in ink wastes being sent to either landfill or incineration facilities.

End User Site
Losses at end user sites are expected to be very low because the storage cartridges will remain sealed until they are placed inside printers. Under normal use, the ink is transferred onto a sheet of paper where it is fixed to the surface. Thus there will be limited release to the environment. Accidental spillage of the chemical, during replacement of cartridges, should result in ink wastes being sent to either landfill or incineration facilities. Additional release to the environment will result from the disposal of the used inkjet cartridge. Spent 15 mL capacity cartridges could contain as much as 5 mL residual ink. The amount of notified chemical within spent cartridges disposed to landfill is estimated to be as high as 56 kg per annum.

Recycling and Disposal
The majority of the notified chemical entering the environment will be bound to paper during the reprographic process. The waste paper generated will be disposed through landfill, recycling, or incineration. Current paper recycling rates in Australia are estimated to be in the order of 70-92% (Australian Environmental Review, 2001). In landfill, the ink (and the notified chemical) should remain fixed to the paper substrate, and remain immobile.

When the paper is recycled, waste sludge containing the notified chemical will be disposed to landfill. It is estimated that the removal of ink particles during the de-inking phase of paper recycling is 30-60% efficient for inkjet copying. It is likely that the same proportion of notified chemical retained in the paper fibre remains in the sludge when the waste paper is re-pulped. Recycling is carried out in paper mills where it is likely that at least primary sedimentation is carried out. Thus, it can be assumed that nearly 100% of easily soluble substances will be released to waste water after primary treatment while around 50% of poorly soluble substances will be removed. Sludges produced by flotation and clarification

will be de-watered and disposed to landfill (EC, 1994). Although the fugacity of the notified chemical in soil is not known, the high water solubility indicates the potential to mobilise to groundwater in landfill (even though the notifier has claimed that the notified chemical precipitates at pH 4). Incinerated paper/ink wastes will generate water and some oxides of carbon, nitrogen and sulphur. During recycling, waste paper is repulped using alkalis, dispersing and wetting agents, water emulsifiable organic solvents and bleaches. These agents enhance fibre separation, ink detachment from paper fibres, pulp brightness, and the whiteness of paper. De-inking wastes are ultimately expected to reside in the sewerage system.

Spent cartridges not recycled are likely to be sent to landfill. As a worst case, a maximum of 56 kg/year of the uncured notified chemical could be sent to landfill from this route if the maximum import quantity of 280 kg and a maximum loss of 20% from each cartridge are assumed. The disposal of cartridges would be widespread across Australia.

Overall Release
Virtually all of the notified chemical will ultimately be released to the environment. Over 90% of the chemical will be bound to printed paper which will either be buried in landfill, incinerated, or released in effluent generated from the de-inking process. Empty cartridges will be disposed of with normal office garbage. Up to 20% of the notified chemical will be disposed of to landfill in spent cartridges.

8.2 Fate

Paper recycling trends will dictate the fate of the majority of the notified chemical. It is anticipated that more than 90% of the notified chemical will be disposed of as paper-bound waste. Some waste paper may be disposed of directly to landfill with the notified chemical bound to the paper. Prolonged residence in an active landfill will eventually degrade the notified chemical. Incineration of the waste paper will destroy the compound with the generation of water vapours and oxides of carbon, nitrogen and sulphur. Recent literature suggests that current paper recycling rates are 70-92% (Australian Environmental Review, 2001). Given high water solubility of the notified chemical, up to 54% of the total import volume may ultimately reside in the sewerage system, where it will associate with the aquatic compartment. The remainder of notified chemical wastes generated from the recycling process will be bound to the paper pulp and become an integral part of the recycled paper.

Up to 20% of the notified chemical will be disposed of to landfill as residues in spent inkjet cartridges. The high water solubility and ionic nature of the notified chemical suggest significant leaching and ready association with the aquatic compartment if released from ruptured cartridges within landfill. This scenario is supported by modelling (Gustafson, 1989). Up to 56 kg per annum of the notified chemical could be leached to groundwater from this route.

The biodegradability of the notified chemical was determined using the 28 Day Ready Biodegradability Test, in accordance with EC Test Guidelines (Brixham Environmental Laboratory, 1999e). After 28 days there was little biodegradation (8%) and the chemical may therefore be classified as not readily biodegradable.

The bioaccumulation of the notified chemical was determined using the Test for the Degree of Bioaccumulation in Fish in accordance with OECD Test Guidelines (Brixham

Environmental Laboratory, 1999f). The test species was mirror carp (*Cyprinus carpio*) of average weight 2.50 g and average length 4.6 cm. Based on the 96 hour LC_{50} and the sensitivity of the analysis of the test substance, the nominal test concentrations were 0.75 (Level 1) and 7.5 mg/L (Level 2). A concurrent blank vessel was used as a control. After the exposure period, the concentrations of the notified chemical in fish tissue were determined and were all below the method detection limit. Accordingly, the bioconcentration factor was also below the limit of detection at <2.3 (Level 1) and <0.24 (Level 2). Therefore, the notified chemical is not expected to bioaccumulate in fish. This is not surprising, given the high water solubility of the notified chemical.

9. EVALUATION OF TOXICOLOGICAL DATA

9.1 Acute Toxicity

Summary of the acute toxicity of the Dye for BJ Printer Ink 001

Test	***Species***	***Outcome***	Reference
acute oral toxicity	rat	LD_{50} >2000 mg/kg	(Noakes, 1999c)
acute dermal toxicity	rat	LD_{50} >2000 mg/kg	(Noakes, 1999a)
skin irritation	rabbit	Non irritant	(Noakes, 1999d)
eye irritation	rabbit	Slight irritant	(Noakes, 1999b)
skin sensitisation	guinea pig	Non sensitiser	(Noakes, 1999e)

9.1.1 Oral Toxicity (Noakes, 1999c)

Species/strain:	Rat/Alpk:AP_fSD
Number/sex of animals:	5/sex
Observation period:	14 days
Method of administration:	A single oral dose of 2000 mg/kg notified chemical in deionised water was given by gavage
Test method:	OECD TG 401 (limit test)
Mortality:	None
Clinical observations:	Blue urine, and blue or blue/black faeces.
	Fur and tail stained blue/black.

Morphological findings:	One male and one female had pelvic dilation of the kidney. Microscopic investigation was not conducted in the study.
Comment:	All animals showed bodyweight gain during the study. The pelvic dilation obaserved was a common spontaneous finding, therefore, is considered to be not treatment related.
LD_{50}:	>2000 mg/kg
Result:	the notified chemical was of low acute oral toxicity in rats

9.1.2 Dermal Toxicity (Noakes, 1999a)

Species/strain:	Rat/Alpk:AP_fSD
Number/sex of animals:	5/sex
Observation period:	14 days
Method of administration:	A single dermal dose of 2000 mg/kg (moistened with deionised water) was applied under an occlusive dressing for 24 hours.
Test method:	OECD TG 402 (limit test)
Mortality:	None
Clinical observations:	Black staining of application sites were noted in all animals which prevented accurate evaluation of erythema for up to 6 days. All female animals had black staining of the tail/skin and one had scabs at the application site between days 3 and 7.
Morphological findings:	Microscopic investigation was not conducted in the study.
Comment:	All but one animal gained weight during the study. Except for scabbing in one female, no sign of irritation was observed in all animals
LD_{50}:	>2000 mg/kg
Result:	the notified chemical was of low dermal toxicity in rats

9.1.3 Inhalation Toxicity

No report on inhalation toxicity was provided.

9.1.4 Skin Irritation (Noakes, 1999d)

Species/strain:	Rabbits/New Zealand White
Number/sex of animals:	3/female
Observation period:	3 days
Method of administration:	A single dermal dose of 500 mg notified chemical (moistened with 0.5mL deionised water) was applied under an occlusive dressing for 4 hours.
Test method:	OECD TG 404
Comment:	Histological examination was conducted on skin samples from the application sites of all animals due to black or black/green staining during the study. The staining prevented an accurate evaluation of irritation, as erythema could not be measured. There were no signs of oedema in any animals. Histological examination revealed no evidence of irritant effects.
Result:	the notified chemical was not irritating to the skin of rabbits

9.1.5 Eye Irritation (Noakes, 1999b)

Species/strain:	Rabbit/New Zealand White
Number/sex of animals:	3/female
Observation period:	3 days
Method of administration:	A single dose (approximately 100 mg notified chemical) was placed into the conjunctival sac of the left eye of each animal. The eyes remained unwashed. The untreated eye served as a control.
Test method:	OECD TG 405

Draize scores of unirrigated eyes:

	Time after instillation											
Animal	***1 day***			***2 days***			***3 days***			***4 days***		
Cornea	***o***		***a***	***o***		***a***	***o***		***a***	***o***		***a***
1	0[1]		0	0		0	0		0	0		0
2	0		0	0		0	0		0	ns		ns
3	0		0	0		0	0		0	ns		ns
Iris	All animals scored zero											
Conjunctiva	***r***	***c***	***d***	***r***	***c***	***d***	***r***	***c***	***d***	***r***	***c***	***d***
1	1	0	0	1	0	0	0	0	0	0	0	0
2	0	1	0	0	0	0	0	0	0	ns	ns	ns
3	1	1	0	0	0	0	0	0	0	ns	ns	ns

[1] see Attachment 1 for Draize scales
o = opacity a = area r = redness c = chemosis d = discharge
ns = not scored

Comment: Staining from the test substance hampered the assessment of irritation at one hour.

No corneal damage was evident in all animals when fluorescein dye was applied to the eyes.

No overt corneal or iridial effects observed in all animals.

All signs of irritation had completely resolved by day 4.

Slight mucous discharge was observed in one animal and grey staining by the test substance in all animals.

Result: the notified chemical was slightly irritating to the eyes of rabbits

9.1.6 Skin Sensitisation (Noakes, 1999e)

Species/strain: Guinea pigs/Dunkin Hartley

Number of animals: Control group: 10
Test group: 20

Induction procedure:

test group: day 1	Intradermal Induction: Three pairs of intradermal injections (0.05-0.1mL) across the scapular region of the animals: - Freund's Complete Adjuvant (FCA) 1:1 in deionised water - 0.3% w/v preparation of the test substance in deionised water - 0.3% w/v preparation of the test substance in 1:1 mixture of FCA and deionised water
day 7	Local Irritation: Animals pre-treated with 10% w/v sodium lauryl sulphate in paraffin wax at the induction site.
day 8	Topical Induction: A 48-hour occluded application of 102% w/v test substance in deionised water to the test area.
control group:	Treated similarly to the test animals using deionised water in intradermal injections and topical application instead of the notified chemical.

Challenge procedure:

day 22	Test and Control Animals: Occluded applications of patches containing 102% w/v, 75% w/v, 50% w/v and 25% w/v notified chemical in deionised water on the right and left flank of each animal for 24 hours.

Test method: OECD TG 406, Magnusson and Kligman Maximisation Test

Challenge outcome:

Challenge concentration	• *Test animals* • *Control animals*			
	• *24 hours**	• *48 hours**	• *24 hours*	• *48 hours*
25%	**0/20	0/20	0/10	0/10
50%	0/20	0/20	0/10	0/10
75%	0/20	0/20	0/10	0/10
102%	0/20	0/20	0/10	0/10

* time after patch removal
** number of animals exhibiting positive response

Comment: The test substance stained the skin black of most test and control animals preventing full assessment of erythema. Therefore, histological examination was conducted on skin samples from the application sites of all animals treated with 102% and 75% w/v preparations.

Minimal to slight changes (acanthosis, inflammatory cell infiltration or parakeratosis) on application site were observed in test animals treated with 102% or 75% w/v preparations and in control animals.

A positive control using hexylcinnamaldehyde demonstrated the sensitivity of the test system.

Result: the notified chemical was not sensitising to the skin of guinea pigs under the conditions of the test

9.2 28 Day Repeated Dose Oral Toxicity (Rattray, 1999)

Species/strain: Rats/Alpk:AP$_f$SD

Number/sex of animals: 5/sex/group

Method of administration: Oral (gavage)

Dose/Study duration:
Test Group Control: 0 mg/kg/day
Low dose: 25 mg/kg/day
Mid dose: 250 mg/kg/day
High dose: 1000 or 500 mg/kg/day

Recovery group Control: 0 mg/kg/day
Dose: 1000 or 500 mg/kg/day

(vehicle: deionised water)

Test animals were treated with the notified chemical for 28 consecutive days. Animals treated with high dose were retained for further 14 days after treatment to monitor recovery from any effects observed (recovery phase).

After 1 week of dosing at 1000 mg/kg/day, a number of animals in high and recovery groups showed clinical changes ranging from slight to extreme loss of appetite and bodyweight. Therefore, the dose level for the high dose and recovery groups was reduced to 500 mg/kg/day.

Test method: OECD TG 407

Clinical observations:
Some animals were terminated or had shedding of bloody tears (chromodacryorrhea), dehydration, piloerection, salivation, sides pinched in, thin, tiptoe gait and upward curvature of the spine after 7 days of dosing at 1000 mg/kg/day. Animals with clinical signs at the end of first week, which persisted to the second week, showed evidence of recovery when dosing was discontinued. No mortalities were observed in the high or recovery group

after the dose level was reduced to 500 mg/kg/day.

In high and recovery groups, body weight gain were reduced in both sexes during the first week, but were either comparable or higher than controls by the end of the study. Food consumption also improved when dosing was reduced to 500 mg/kg/day.

Other non-treatment related observations in the high dose group include stained tail and bulging eyes.

No effects on any of the functional observational battery tested were reported in all animals.

Haematology
Minimal decrease in red blood cell count was confined to high dose males and was not present following recovery period.

Clinical chemistry
Increase in albumin/globulin ratio was seen in all treated female groups; however, the increase was non-dose related and the control value was lower than the historical control data. Increase in albumin and cholesterol values in high dose males was also observed but was not present following recovery period.

Increase in alanine aminotransferase (ALT) was seen in mid dose males and in both sexes in high dose groups. Alkaline phosphatase (ALP) was increased in mid dose females and high dose group, but not statistically significant in high dose group. For both enzymes, no effects were present following the recovery period. There is a decreased in aspartate aminotransferase activity seen in low, mid and recovery groups, but was considered to be attributed to unusually high values in some control animals.

No effects on electrolyte or mineral parameters were reported in all dose groups. Other variations in clinical chemistry parameters were confined to single sexes at mid dose group and were not considered to be treatment-related.

Histopathology:
Blue discolouration of gastrointestinal contents and black/grey discolouration of the tail due to staining by the test material was observed in a number of animals dosed at 1000 mg/kg/day and terminated on day 7. Histopathological changes in the kidney indicative of a slight nephrotoxicity in the cortical and medullary collecting ducts were also observed. The absence of hepatic glycogen in the liver was considered to be secondary to anorexia associated with poor clinical condition at the time of termination.

None of the above changes were observed in any of the high and recovery groups, which survived to termination after reducing the dose to 500 mg/kg/day. There were increases in kidney weight in surviving high dose (2/4 females and 2/3 males) and female mid dose groups. These increases were not seen in the recovery group, and in the absence of histopathological change in the kidney at these dose levels, the effect is considered to be an adaptive response to the treatment.

Liver weights were increased in female high dose and in male recovery groups. No histopathological changes in the liver were observed.

Increases in both heart and ovary weights were observed in female high and recovery groups. All weights were within the control range for this type of study and no histopathological changes in these organs were observed.

At scheduled termination, no compound related changes were observed except from staining of the tail and gastrointestinal contents in some animals in the mid and low dose groups. Staining of the ingesta and stomach lining was observed in some animals but was not associated with histological changes in the gastrointestinal tract.

There was no evidence for neuro- or immunotoxicity in the study.

Comment:
At a dose level of 250 mg/kg/day notified chemical, there were increases in female kidney weights and increase in ALT and ALP. No histopathological changes in the kidney and liver were observed.

Oral administration of 500 mg/kg/day notified chemical for 28 consecutive days produced increases in kidney (both sexes) and liver (females only) weights, however, no histopathological effects were seen in these organs. Increase in ALT and ALP were also observed in both sexes. Similar changes were not seen in the recovery group animals dosed at 500 mg/kg/day, therefore these findings were considered not of toxicological importance.

Result:
The no observed effect level (NOEL) is considered to be 25 mg/kg/day, the lowest dose tested, based on increases in liver and kidney weights seen at higher doses.

9.3 Genotoxicity

9.3.1 *Salmonella typhimurium* and *Escherichia coli* Reverse Mutation Assay (Callander, 1999)

Strains:	*Salmonella typhimurium*: TA1535, TA1537; TA98 and TA100 *Escherichia coli*: WP2P and WP2P *uvrA*
Metabolic activation:	Liver S9 fraction from rats pretreated with combined phenobarbital and β-naphthoflavone in corn oil solution.
Concentration range:	0 – 5184 μg/plate of test substance in deionised water Each concentration was tested in triplicate, with or without metabolic activation with S9, in two independent experiments. Appropriate strain specific positive control reference substances were used.
Test method:	OECD TG 471 and 472

Comment:	No toxicity was observed in any of the tested strains of *Salmonella* and *E. coli*. There were no significant, reproducible increases in the number of revertant colonies in the presence or absence of metabolic activation at any test concentration. Concurrent positive controls induced marked increases in the number of revertant colonies and the activity of the S9 fraction was found to be satisfactory.
Result:	The notified chemical was non mutagenic under the conditions of the test

9.3.2 Chromosomal Aberration Assay in Human Lymphocytes *In Vitro* (Fox, 1999)

Cells:	Human lymphocytes
Metabolic activation system:	Liver S9 fraction from rats pretreated with combined phenobarbital and β-naphthoflavone corn oil preparation
Dosing schedule:	Each concentration was tested in duplicate, with or without metabolic activation (S9), in two independent experiments.

• *Metabolic Activation*	• ***Experiment Number***	• *Test concentration (μg/mL)*	• *Controls*
-S9	I	treatment time =3 hours (20 hours harvest) 0, 10, 50, 100, 200, 500*, 1000, 2500* and 5187* μg/mL	Positive: CP Negative: Mitomycin C
	II	treatment time = 20 hours 0, 100, 250*, 500, 1000*, 1750, 2500*, 3800 and 5187 μg/mL	
+S9	I	treatment time = 3 hours (20 hours harvest) 0, 10, 50, 100, 200, 500*, 1000, 2500* and 5187* μg/mL	Positive: CP Negative: Mitomycin C
	II	treatment time = 3 hours (20 hours harvest) 0, 10, 50, 100, 200, 500*, 1000, 2500* and 5187* μg/mL	

CP - cyclophosphamide * - cultures selected for metaphase analysis

Test method:	OECD TG 473
Comment:	No statistically significant increases in the incidence of aberrant cells in either Experiment 1 or Experiment 2 in the presence or absence of metabolic activation. Cultures treated for 20 hours (Experiment 2, without S9) had reduced mean mitotic activity at the highest concentration selected for chromosomal aberration analysis (2500 µg/mL). Concentrations higher than 2500 µg/mL showed excessive toxicity. No significant reductions in mitotic activity were observed following 3 hour treatment periods (Experiment 1, with or without S9 and Experiment 2, with S9) Positive controls used in the test caused marked increases in the incidence of aberrant cells and the activity of S9 fraction was found to be satisfactory.
Result:	The notified chemical was non clastogenic under the conditions of the test

9.3.3 Micronucleus Assay in the Bone Marrow Cells of the Mouse

No report of *in vivo* micronucleus assay was provided.

9.4 Overall Assessment of Toxicological Data

The notified chemical was of low acute oral (LD_{50} >2000 mg/kg) and dermal (LD_{50} >2000 mg/kg) toxicity in rats. No report on acute inhalation toxicity was provided.

It was not a skin irritant but slight eye irritant to rabbits. The irritation studies were hampered by colouring by the test substance. Evidence of skin sensitisation potential was not observed in guinea pigs in an adjuvant study.

In a repeat dose oral toxicity study in rats, a NOEL was established as 25 mg/kg/day, based on increased kidney and liver weights seen at higher doses. Histopathological changes were not observed at doses 500 mg/kg/day or less.

The notified chemical was not mutagenic in the bacterial strains tested, and not clastogenic in *in vitro* chromosome aberration study using human lymphocytes.

Jones (1999) assessed the toxicokinetic potential of the notified chemical using the above toxicity studies. He concluded that the notified chemical is absorbed following oral administration as evidenced by the excretion of coloured urine, some histopathological changes, organ weight changes and some perturbations in clinical chemistry parameters. However, there was no evidence of any dermal penetration of the notified chemical.

The high molecular weight of the notified chemical would restrict absorption across the

gastrointestinal mucosa following oral administration. However, based on its aqueous solubility, the absorbed chemical could be excreted in the urine. Alternatively, it could be eliminated through the bile for subsequent excretion in the faeces, together with the unabsorbed dose.

Cleavage of two azo bonds in the notified chemical may occur by the intestinal microflora. The resultant cleavage products would be expected to be more highly absorbed than the parent molecule. Cleavage products of the notified chemical would be subject to conjugation of the hydroxy and carboxylic acid moieties with potential of acetylation of amine groups to promote excretion.

Based on the toxicological data provided, the notified chemical, Dye for BJ Printer Ink 001, is not classified as a hazardous substance according to the NOHSC *Approved Criteria for Classifying Hazardous Substances* (NOHSC, 1999).

10. ASSESSMENT OF ENVIRONMENTAL EFFECTS

The following ecotoxicity studies have been provided and the results are summarised in the following table. The tests were performed in compliance with OECD/EEC Test Methods and in accordance with OECD Principles of Good Laboratory Practice.

Test	***Species***	***Results***
acute toxicity to fish	Mirror carp *Cyprinus carpio*	LC_{50} (96 h) = 750 mg/L NOEC (96 h) = 375 mg/L
acute toxicity to marine invertebrates	Water flea *Daphnia magna*	EC_{50} (48 h) = 130 mg/L NOEC (48 h) = 56 mg/L
algal growth inhibition	Algae *Selenastrum capricornutum*	E_bC_{50} (72 h) = 0.81 mg/L E_rC_{50} (72 h) = 4.4 mg/L NOEC (biomass) = <0.2 mg/L NOEC (growth) = 0.2 mg/L See below
inhibition of microbial activity	Activated sludge	EC_{50} (3 h) >200 mg/L NOEC (3 h) = 200 mg/L

- NOEC - no observable effect concentration
- LC_{50} - median lethal concentration
- EC_{50} - median environmental concentration
- E_bC_{50} - median effective concentration, biomass
- E_rC_{50} - median effective concentration, growth rate

Acute Toxicity to Fish (Brixham Environmental Laboratory, 1999a)

The test organism was the mirror carp (*Cyprinus carpio*), a sensitive freshwater fish. Less than 1% mortality was observed in the fish 7 days prior to the test. Prior to the test, the fish were acclimatised to the test temperature (22 ± 1°C) for a minimum period of 7 days. Food was withheld from the fish for at least 24 hours prior to the commencement of the test. At the end of the exposure period, the fish from the dilution water control were weighed and measured. The range in weight was 1.84 to 3.54 g with a mean of 2.50 g, and the range in

length was 42 to 51 mm with a mean of 46 mm. The fish exceeded the guideline recommended total length but the fish loading of 0.71 g/L was below the maximum loading of 1.0 g/L. This deviation from the guideline is not considered to have affected the validity of the test.

The test procedure employed was a static system. The apparatus used were glass vessels with a working volume of 35 L, with single vessels used for the dilution water control and the exposure solutions. Ten fish were used in both the test concentrations and the dilution water control. The test was undertaken in a temperature-controlled room which was set at the nominal test temperature of 22 ± 1°C and the light regime was fluorescent lighting for 16 hours and darkness hours with 20 minute transition periods. Nominal exposure concentrations of 0, 375, 750, 1500 and 6000 mg/L were employed. The test solutions were gently aerated during the 96 hour test period and the fish were not fed during the course of the test. Due to the intensity of colouration of the test solutions, it was not possible to observe symptoms of toxicity. Therefore, the NOEC is based on mortalities only.

The 96 hour LC_{50} for the notified chemical was 750 mg/L (96 hour LC_{100} was 1500 mg/L), the 96 hour NOEC was 375 mg/L and the chemical is considered to be practically non-toxic to carp. All reported values are nominal concentrations.

Acute Toxicity to Aquatic Invertebrates (Brixham Environmental Laboratory, 1999b)

The test organism was the freshwater crustacean, *Daphnia magna* Straus, obtained from continuous laboratory cultures. The stock cultures of daphnia were maintained in a reconstituted water medium, identical to the test dilution water, at 20 ± 2°C with a photoperiod of 16 hours light:8 hours dark. The cultures were fed a defined diet of algae *Chlorella vulgaris* and "Frippak Booster®" (a commercially available microencapsulated diet). Nominal test solution concentrations of 0, 18, 32, 56, 100 and 180 mg/L were employed. Due to the intensity of colouration in the test solutions, observations could not be made at 24 hours. At 48 hours, test solutions were diluted until observation of the daphnids became possible.

The 48 hour EC_{50} was 130 mg/L (48 hour EC_{100} was >180 mg/L), and the 48 hour NOEC was 56 mg/L, indicating that the notified chemical is practically non-toxic to daphnids. All reported values are nominal concentrations.

Algal Inhibition Test (Brixham Environmental Laboratory, 1999c)

The test organism was the unicellular green alga *Selenastrum capricornutum* Printz (Strain ATCC 22662) from laboratory cultures maintained under axenic conditions. A culture of the alga in the exponential growth phase was used as inoculum for the test.

Four replicate cultures of the control and each test substance concentration were employed. For each test substance concentration there were two replicates of the exposure and shaded test vessels. Exposure vessels incubated alga in test substance solutions, shaded by excess control culture medium; shaded vessels incubated alga in control culture medium shaded by the test substance solutions. One blank (no algal inoculum) was incubated concurrently for each control and test concentration. Nominal test solution concentrations of 0, 0.20, 0.45, 1.0, 2.3, 5.0, 11 and 25 mg/L were employed.

The 72 hour E_bC_{50} was 0.81 mg/L, the NOEC (biomass) was <0.20 mg/L, the 72 hour E_rC_{50} was 4.4 mg/L, and the NOEC (growth) was 0.20 mg/L. All reported values pertain to nominal concentrations. Superficially, these results indicate that the notified chemical is highly toxic to algae. However, it is recommended that growth rates should be used for the calculations of EC_{50} for coloured substances. Consequently, those data are used for all subsequent comparisons. From data obtained for exposure in shaded vessels, it can be surmised that the inhibition curves are essentially identical. Therefore, the notified chemical satisfies the Annex V1 (Dir. 93/21/EEC) exemption clause and the 72 hour EC_{50} should not be used as a basis for classification. Thus, the notified chemical is not considered to be biochemically toxic to algae, although may cause algal growth inhibition through light exclusion effects.

Activated Sludge Respiration Inhibition (Brixham Environmental Laboratory, 1999d)

This test measures the respiration rate of an activated sludge 3 hours after feeding an excess, but standard amount of an OECD synthetic sewage and compares this with the respiration rate of the same activated sludge in the presence of the test chemical.

Nominal test solution concentrations of 0, 2.0, 6.4, 20, 64 and 200 mg/L were employed. 3,5-Dichlorophenol was used as the reference material, at nominal concentrations of 1.0, 3.2, 10, 32 and 100 mg/L.

The 3 hour EC_{50} for the notified chemical was determined to be >200 mg/L (3 hour EC_{10} was >200 mg/L), and the 3 hour EC_{50} of the reference material was 5 mg/L. The 3 hour NOEC of the notified chemical 200 mg/L. All reported values are nominal concentrations. The notified chemical is not considered to significantly affect the respiration rate of activated sludge.

11. ASSESSMENT OF ENVIRONMENTAL HAZARD

The notified chemical will enter environmental compartments indirectly by disposal of waste paper (for recycling, to landfill, or for incineration) and by direct release from discarded spent cartridges at landfill sites. Based on the import volume, method of packaging and low concentration of the notified chemical in ink, release of the notified chemical to the environment is expected to be low and widespread. Waste from the recycling process includes sludge, which is dried and disposed of to landfill, but most of the notified chemical will partition to the supernatant water that is released to the sewer. If it is assumed that up to 92% of waste paper is recycled, up to 60% of the notified chemical will be removed during the de-inking phase of the paper recycling process. This water-soluble fraction will be released to the sewerage system.

The Predicted Environmental Concentration (PEC) is calculated below:

Maximum Annual Volume of Imported Chemical	280 kg
Maximum Volume of Chemical Transferred to Paper (95%)	323 kg
Maximum Volume of Chemical Recycled Paper (92%)	297 kg
Maximum Volume of Chemical in Sewerage System (assuming a 60% de-inking rate)	178 kg (0.49 kg/day)
National Population	18,000,000
Daily Water Usage/Person	150 L
PEC	0.18 µg/L

The PEC of 0.18 µg/L is well below the toxicity threshold of the most sensitive aquatic species tested, namely the EC_{50} of 130 mg/L for daphnids. The notified chemical may, however, exert phototoxic effects on organisms which depend upon light to survive and reproduce. Even so, the E_bC_{50} for algae is more than three orders of magnitude greater than PEC of the notified chemical.

Abiotic or slow biotic processes would be largely responsible for the degradation of the notified chemical as it was not readily biodegradable. The low octanol-water partition coefficient and high water solubility indicate the notified chemical will be predominantly distributed in water, where it will become diluted and dispersed.

Any released chemical is not expected to adversely affect aquatic organisms, since it is practically non toxic to carp, daphnia and algae. In addition, bioaccumulation is not expected due to its low log P_{ow}, indicating low lipid solubility, and large molecular weight (~700) which inhibits passage through cell membranes.

Overall, the environmental risk presented by the introduction of the notified chemical is predicted to be low.

12. ASSESSMENT OF PUBLIC AND OCCUPATIONAL HEALTH AND SAFETY EFFECTS

Hazard Assessment

Based on the toxicological data provided, the notified chemical would not be acutely toxic via oral or dermal routes. It is not likely to be a skin irritant or skin sensitiser. However, it is likely to be a slight eye irritant. In a repeat dose oral toxicity study in rats, a NOEL was established as 25 mg/kg/day, the lowest dose tested, based on increases in liver and kidney weights seen at higher doses. In genetoxicity studies, the notified chemical was not mutagenic in the bacterial strains tested, and not clastogenic in *in vitro* chromosome aberration study using human lymphocytes.

The notified chemical would not be classified as a hazardous substance according to NOHSC *Approved Criteria for Classifying Hazardous Substances* (NOHSC, 1999) in terms of the toxicological data provided.

Occupational Health and Safety

Exposure to printing inks containing the notified chemical during transport of pre-packed

cartridges should not result in exposure except in the event of accidental spillage.

The notified chemical will be in imported inkjet cartridges at a maximum of 3.5%. Dermal exposure of office workers to the notified chemical will potentially occur when replacing spent cartridges and clearing paper jams from the printer. However, the design of the cartridges is such that exposure and risk to the notified chemical should be negligible.

Dermal exposure of maintenance workers to the notified chemical is possible during routine maintenance but is expected to be low due to the low concentration of the notified chemical in the ink. However, due to their frequent exposure to inks, maintenance personnel should wear cotton or disposable gloves.

It is concluded that the risk of eye irritation or other topical or systemic health effects in workers involved in transport, storage, use and disposal of the notified chemical in this application is low.

Public Health
There will be no significant public exposure to the notified chemical given the low concentration in the ink product and the design of the cartridges. Contact with residue on the printer's internal workings will be very small. Contact with printed paper is unlikely to lead to significant dermal exposure, as the chemical will be bound to the paper. There is unlikely to be any public health risk posed by the notified chemical.

There is a very slight chance of ingestion of the chemical due to accidental rupture of a cartridge. A 10 kg child ingesting 5 mL of a 3.5% solution would receive a dose of approximately 17.5 mg/kg which is significantly below the lethal dose (LD_{50} > 2000 mg/kg). The chemical has a low acute oral toxicity and the quantities consumed would be minimal, so the chemical is unlikely to pose a significant risk to human health.

13. MATERIAL SAFETY DATA SHEET (MSDS) AND LABEL ASSESSMENT

13.1 MSDS

The MSDS for the notified chemical was provided in a format consistent with the *National Code of Practice for the Preparation of Material Safety Data Sheets* (NOHSC, 1994a).

This MSDS was provided by the applicant as part of the notification statement. It is reproduced here as a matter of public record. The accuracy of this information remains the responsibility of the applicant.

13.2 Label

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

13. RECOMMENDATIONS

Control measures (end-user)

No special precautions are required for the notified chemical when used at low quantities in inkjet printer cartridges. However, in the interests of good occupational health and safety, the following guidelines and precautions should be observed:

- Service personnel should wear cotton or disposable gloves when servicing printers.

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.

Secondary notification

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

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

The Director will then decide whether secondary notification is required.

No additional secondary notification conditions are stipulated.

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Attachment 1

The Draize Scale (Draize, 1959) for evaluation of skin reactions is as follows:

Erythema Formation	*Rating*	*Oedema Formation*	*Rating*
No erythema	0	No oedema	0
Very slight erythema (barely perceptible)	1	Very slight oedema (barely perceptible)	1
Well-defined erythema	2	Slight oedema (edges of area well-defined by definite raising	2
Moderate to severe erythema	3	Moderate oedema (raised approx. 1 mm)	3
Severe erythema (beet redness)	4	Severe oedema (raised more than 1 mm and extending beyond area of exposure)	4

The Draize scale (Draize *et al*., 1944) for evaluation of eye reactions is as follows:

CORNEA

Opacity	*Rating*	*Area of Cornea involved*	*Rating*
No opacity	0 none	25% or less (not zero)	1
Diffuse area, details of iris clearly visible	1 slight	25% to 50%	2
Easily visible translucent areas, details of iris slightly obscure	2 mild	50% to 75%	3
Opalescent areas, no details of iris visible, size of pupil barely discernible	3 moderate	Greater than 75%	4
Opaque, iris invisible	4 severe		

CONJUNCTIVAE

Redness	*Rating*	*Chemosis*	*Rating*	*Discharge*	*Rating*
Vessels normal	0 none	No swelling	0 none	No discharge	0 none
Vessels definitely injected above normal	1 slight	Any swelling above normal	1 slight	Any amount different from normal	1 slight
More diffuse, deeper crimson red with individual vessels not easily discernible	2 mod.	Obvious swelling with partial eversion of lids	2 mild	Discharge with moistening of lids and adjacent hairs	2 mod.
		Swelling with lids half-closed	3 mod.	Discharge with moistening of lids and hairs and considerable area around eye	3 severe
Diffuse beefy red	3 severe	Swelling with lids half-closed to completely closed	4 severe		

IRIS

Values	*Rating*
Normal	0 none
Folds above normal, congestion, swelling, circumcorneal injection, iris reacts to light	1 slight
No reaction to light, haemorrhage, gross destruction	2 severe