

**Screening Assessment for the Challenge**

**Propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]-  
(Disperse Red 179)**

**Chemical Abstracts Service Registry Number  
16586-42-8**

**Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]-  
(DAPEP)**

**Chemical Abstracts Service Registry Number  
25176-89-0**

**Environment Canada  
Health Canada**

**March 2010**

## Synopsis

Pursuant to section 74 of the *Canadian Environmental Protection Act, 1999* (CEPA 1999), the Ministers of the Environment and of Health have conducted a screening assessment on Propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]- (Disperse Red 179), Chemical Abstracts Service Registry Number 16586-42-8; and Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]- (DAPEP), Chemical Abstracts Service Registry Number 25176-89-0. These substances were identified as high priorities for screening assessment and included in the Challenge because they had been found to meet the ecological categorization criteria for persistence, bioaccumulation potential and inherent toxicity to non-human organisms and are believed to be in commerce in Canada.

The substances Disperse Red 179 and DAPEP were not considered to be high priorities for assessment of potential risks to human health, based upon application of the simple exposure and hazard tools developed by Health Canada for categorization of substances on the Domestic Substances List. Therefore this assessment focuses principally on information relevant to the evaluation of ecological risks.

Disperse Red 179 and DAPEP are organic substances that are used in Canada primarily as red dyeing agents for synthetic fibres for clothing and home textile uses. Due to their similar structure and uses, Disperse Red 179 and DAPEP are being assessed together in this report. These substances are not naturally produced in the environment. They are not reported to be manufactured in Canada above the reporting threshold of 100 kg/year; however, 400 kg of Disperse Red 179 and 100 kg of DAPEP were imported into the country in 2006 for use in the textile industry.

Based on certain assumptions and reported use patterns in Canada, the greatest proportion of these substances is expected to end up in waste disposal sites. About 17% of Disperse Red 179 and DAPEP is estimated to be released to water, and no releases are predicted to air and soil. Disperse Red 179 and DAPEP present very low solubility in water and octanol (based on analogue and modelled data). Disperse Red 179 and DAPEP are present in the environment primarily as fine particulate matter that is not volatile, are chemically stable, and have a tendency to partition by gravity to sediments if released to surface waters, and would likely partition to soils if released to air.

Based on their physical and chemical properties and on experimental biodegradation test data, Disperse Red 179 and DAPEP are expected to be persistent in the environment in all media under aerobic conditions. Newly identified analogue experimental data and expert judgement indicate that these dyes have a low potential to accumulate in the lipid tissues of organisms. The substances therefore meet the persistence criteria but do not meet the bioaccumulation criteria as set out in the *Persistence and Bioaccumulation Regulations*. In addition, new experimental toxicity data for chemical analogues suggest that these substances have at most a low to moderate potential to cause acute harm to aquatic organisms.

For the environmental section of this screening assessment, two very conservative exposure scenarios representing releases from industrial and consumer use to the aquatic environment were applied. The first scenario simulated discharge of Disperse Red 179 or DAPEP to the aquatic environment following use of each dye by an industrial operation. The second scenario simulated the release of Disperse Red 179 or DAPEP to the aquatic environment from consumer use (such as washing laundry). The predicted environmental concentrations in water for each scenario were below the predicted no-effect concentrations calculated for pelagic organisms.

The potential for exposure of the general population in Canada to Disperse Red 179 and DAPEP from environmental media is expected to be negligible. Exposure to Disperse Red 179 and DAPEP from consumer products is expected to be low given the intended purpose of the product (dyes in synthetic textiles), taking into consideration potential for incidental exposures, such as mouthing by toddlers. Due to the lack of experimental data on these substances, upper-bounding exposure estimates were derived based on available data on the migration of disperse dyes from synthetic textiles.

The limited empirical data identified for Disperse Red 179 and DAPEP, potential metabolites, and analogues, together with mixed Quantitative Structure-Activity Relationship (QSAR) predictions, suggest these substances may pose a potential hazard to human health.

Although limited data may suggest a potential hazard associated with Disperse Red 179 and DAPEP, exposure of the general population in Canada to these substances based on their use in textiles is expected to be low, therefore the risk to human health is considered to be low.

Based on the information available, Disperse Red 179 and DAPEP do not meet any of the criteria set out in section 64 of the *Canadian Environmental Protection Act, 1999*.

Because these substances are listed on the *Domestic Substances List*, their import and manufacture in Canada are not subject to notification under subsection 81(1). Given the potential hazardous properties of these substances, there is concern that new activities that have not been identified or assessed could lead to these substances meeting the criteria set out in section 64 of the Act. Therefore, it is recommended to amend the *Domestic Substances List*, under subsection 87(3) of the Act, to indicate that subsection 81(3) of the Act applies with respect to these substances so that new manufacture, import or use of these substances is notified and undergoes ecological and human health risk assessments.

In addition and where relevant, research and monitoring will support verification of assumptions used during the screening assessment.

## Introduction

The *Canadian Environmental Protection Act, 1999* (CEPA 1999) (Canada 1999) requires the Minister of the Environment and the Minister of Health to conduct screening assessments of substances that have met the categorization criteria set out in the Act to determine whether these substances present or may present a risk to the environment or human health.

Based on the information obtained through the categorization process, the Ministers identified a number of substances as high priorities for action. These include substances that

- met all of the ecological categorization criteria, including persistence (P), bioaccumulation potential (B) and inherent toxicity to aquatic organisms (iT), and were believed to be in commerce in Canada; and/or
- met the categorization criteria for greatest potential for exposure (GPE) or presented an intermediate potential for exposure (IPE), and had been identified as posing a high hazard to human health based on classifications by other national or international agencies for carcinogenicity, genotoxicity, developmental toxicity or reproductive toxicity.

The Ministers therefore published a notice of intent in the *Canada Gazette*, Part I, on December 9, 2006 (Canada 2006a), that challenged industry and other interested stakeholders to submit, within specified timelines, specific information that may be used to inform risk assessment, and to develop and benchmark best practices for the risk management and product stewardship of those substances identified as high priorities.

The substances Propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]- (Disperse Red 179), and Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]- (DAPEP), were identified as high priorities for assessment of ecological risk as they had been found to be persistent, bioaccumulative and inherently toxic to aquatic organisms and are believed to be in commerce in Canada. The Challenge for these substances was published in the *Canada Gazette* on August 30, 2008 (Canada 2008). A substance profile for each substance was released at the same time. The substances profiles presented the technical information available prior to December 2005 that formed the basis for categorization of these substances. As a result of the Challenge, submissions of information pertaining to the persistence, hazard and uses of these substances were received.

Although Diperse Red 179 and DAPEP were determined to be a high priority for assessment with respect to the environment, they did not meet the criteria for GPE or IPE and high hazard to human health based on classifications by other national or international agencies for carcinogenicity, genotoxicity, developmental toxicity or reproductive toxicity.

Screening assessments focus on information critical to determining whether a substance meets the criteria set out in section 64 of CEPA 1999. Screening assessments examine scientific information and develop conclusions by incorporating a weight-of-evidence approach and precaution.

This screening assessment includes consideration of information on chemical properties, hazards, uses and exposure, including the additional information submitted under the Challenge. Data relevant to the screening assessment of this substance were identified in original literature, review and assessment documents, stakeholder research reports and from recent literature searches, up to December 29<sup>th</sup>, 2009. Key studies were critically evaluated; modelling results may have been used to reach conclusions. When available and relevant, information presented in hazard assessments from other jurisdictions was considered. The screening assessment does not represent an exhaustive or critical review of all available data. Rather, it presents the most critical studies and lines of evidence pertinent to the conclusion.

Disperse Red 179 and DAPEP are being assessed together in this screening assessment report. Physical and chemical property data for these dyes are lacking, and given the similarities in their respective structures and uses, acceptable analogues have been identified that have relevant data to support the ecological assessment of these two dyes.

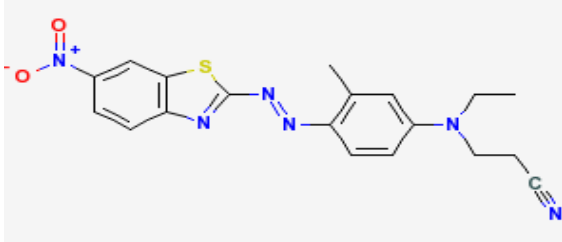
This screening assessment was prepared by staff in the Existing Substances Programs at Health Canada and Environment Canada and incorporates input from other programs within these departments. The ecological portion of this assessment has undergone external written peer review/consultation. Additionally, the draft of this screening assessment was subject to a 60-day public comment period. While external comments were taken into consideration, the final content and outcome of the screening risk assessment remain the responsibility of Health Canada and Environment Canada. The critical information and considerations upon which the assessment is based are summarized below.

## Substance Identity

For the purposes of this document, the substance Propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]- (CAS RN 16586-42-8), will be referred to as Disperse Red 179, instead of NBATP, which was used in the substance profile. “Disperse Red 179” is defined by the Colour Index (CII 2002–) as a combination of two CAS numbers (CAS RN 61951-64-2 and CAS RN 16195-64-8). The substances associated with CAS RN 16586-42-8 and CAS RN 61951-64-2 are identical since both have identical scientific names (NCI 2009), identical chemical structures (ChemID 2009; CII 2002–) and the same Colour Index number C.I. 11290 (CII 2002–). According to the Colour Index, a specific chemical can have more than one CAS RN, especially in the field of dyes, depending on whether the substance was submitted to registration to the Chemical Abstracts Services under its full chemical name or under its generic C.I. name (CII 2002–). Thus, in this particular instance, CAS RN 61951-64-2 refers to the C.I. name Disperse Red 179, while the CAS RN 16586-42-8 actually refers to the chemical name propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]-. The CAS RN 16195-64-8 however is not found on the Chemical Abstract Services inventory list (NCI 2009) and no substance is or was ever associated to this CAS RN (Chemical Abstract Services, personal communication, unreferenced). Therefore, Disperse Red 179 is not a mixture of two CAS numbers but a discrete chemical listed under the CAS RN 16586-42-8 (see Table 1a).

The substance Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]- (CAS RN 25176-89-0), is sometimes referred to as Disperse Red 153. However, Disperse Red 153, which is registered under the CAS RN 78564-87-1 (CI 111370), actually is a mixture of two structural isomers (Nakagawa 1996; CII 2002–). One isomer is registered under CAS RN 25176-89-0 (CI 111371), and the other does not have a registered CAS number but has a Colour Index registration number, CI 111372 (CII 2002–). These two substances are structural isomers (same chemical formula); therefore, it is anticipated that the properties of the chemical mixture closely resembles those of CAS RN 25176-89-0. For the purpose of this document, CAS RN 25176-89-0 will be referred to as DAPEP, an acronym derived from its inventory name (see Table 1b).

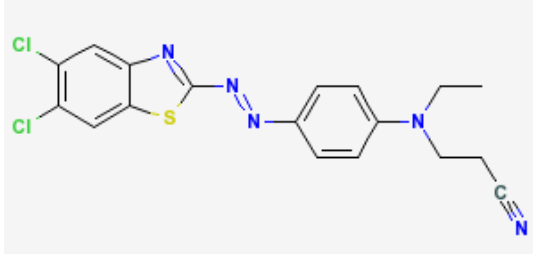
Table 1a. Substance identity for Disperse Red 179

<b>Chemical Abstracts Service Registry Number (CAS RN)</b>	<b>16586-42-8</b>
<b>DSL name</b>	<b>Propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]-</b>
<b>National Chemical Inventories (NCI) names<sup>1</sup></b>	<i>Propanenitrile, 3-[ethyl[3-methyl-4-[2-(6-nitro-2-benzothiazolyl)diazenyl]phenyl]amino]-</i> (TSCA) <i>Propanenitrile, 3-[ethyl[3-methyl-4-[(6-nitro-2-benzothiazolyl)azo]phenyl]amino]-</i> (AICS, PICCS, ASIA-PAC) <i>3-[Ethyl[3-methyl-4-[(6-nitrobenzothiazol-2-yl)azo]phenyl]amino]propiononitrile</i> (EINECS, ECL) <i>C.I. Disperse Violet 052</i> (ECL) <i>C.I. DISPERSE RED 179</i> (PICCS)
<b>Other names</b>	<i>3-[N-Ethyl-4-[(6-nitro-2-benzothiazolyl)azo]-m-toluidino]propionitrile</i> <i>C.I. 112290, C.I. Disperse Violet 52</i> <i>Disperse Red 179, Disperse Violet 52</i> <i>Kayalon Polyester Rubine BL-S</i> <i>Kayalon Polyester Rubine BL-S 200</i> <i>Propionitrile, 3-[N-ethyl-4-[(6-nitro-2-benzothiazolyl)azo]-m-toluidino]-</i> <i>3-(ethyl[3-methyl-4-[(6-nitrobenzothiazol-2-yl)azo]phenyl]amino)propiononitrile</i>
<b>Chemical group (DSL Stream)</b>	Discrete organics
<b>Major chemical class or use</b>	Organic disperse azo dye
<b>Major chemical sub-class</b>	Mono azo benzothiazole dye
<b>Chemical formula</b>	C <sub>19</sub> H <sub>18</sub> N <sub>6</sub> O <sub>2</sub> S
<b>Chemical structure</b>	
<b>SMILES<sup>2</sup></b>	<chem>N(=O)(=O)c1ccc(nc1=Nc2ccc(cc2N(CCC#N)CC)c1)C)c1s2)c23)c3</chem>
<b>Molecular mass</b>	394.45 g/mol

<sup>1</sup> National Chemical Inventories (NCI). 2009: AICS (Australian Inventory of Chemical Substances); ASIA-PAC (Asia-Pacific Substances Lists); ECL (Korean Existing Chemicals List); EINECS (European Inventory of Existing Commercial Chemical Substances); PICCS (Philippine Inventory of Chemicals and Chemical Substances); and TSCA (Toxic Substances Control Act Chemical Substance Inventory).

<sup>2</sup> Simplified Molecular Line Input Entry System.

Table 1b. Substance identity for DAPEP

<b>Chemical Abstracts Service Registry Number (CAS RN)</b>	<b>25176-89-0</b>
<b>DSL name</b>	<b>Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]-</b>
<b>National Chemical Inventories (NCI) names<sup>1</sup></b>	<i>Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]-</i> (AICS, PICCS, ASIA-PAC) <i>3-[[4-[(5,6-Dichlorobenzothiazol-2-yl)azo]phenyl]ethylamino]propiononitrile</i> (EINECS) <i>3-[[4-[(5,6-Dichlorobenzothiazol-2-yl)azo]phenyl]ethylamino]propiononitrile</i> (PICCS)
<b>Other names</b>	<i>Propionitrile, 3-[p-[(5,6-dichloro-2-benzothiazolyl)azo]-N-ethylanilino]-</i> <i>3-({4-[(5,6-Dichlorobenzothiazol-2-yl)azo]phenyl}ethylamino)propiononitrile</i>
<b>Chemical group (DSL Stream)</b>	Discrete organics
<b>Major chemical class or use</b>	Organic disperse azo dye
<b>Major chemical sub-class</b>	Mono azo benzothiazole dye
<b>Chemical formula</b>	C <sub>18</sub> H <sub>15</sub> Cl <sub>2</sub> N <sub>5</sub> S
<b>Chemical structure</b>	
<b>SMILES</b>	<chem>c12N=C(N=Nc3ccc(N(CC)CCC(#N))cc3)Sc1cc(Cl)c(Cl)c2</chem>
<b>Molecular mass</b>	404.32 g/mol

<sup>1</sup> National Chemical Inventories (NCI). 2009: AICS (Australian Inventory of Chemical Substances); ASIA-PAC (Asia-Pacific Substances Lists); EINECS (European Inventory of Existing Commercial Chemical Substances) and PICCS (Philippine Inventory of Chemicals and Chemical Substances).

<sup>2</sup> Simplified Molecular Line Input Entry System.



## Identification of Analogue Substances and Estimation of Physical and Chemical Properties

Few experimental data are available for Disperse Red 179 or DAPEP. At the Environment Canada-sponsored Quantitative Structure-Activity Relationship (QSAR) Workshop in 1999 (Environment Canada 2000), invited modelling experts identified many structural classes of pigment and dyes as “difficult to model” using QSARs. The physical and chemical properties of many of the structural classes of dyes and pigments (including acid and disperse dyes) are not amenable to model prediction because they are considered “out of the model domain of applicability” (e.g., structural and/or property parameter domains). Therefore, to determine potential utility, the domains of applicability of QSAR models to dyes and pigments are evaluated on a case-by-case basis.

Environment Canada has considered it inappropriate to use QSAR models to predict most of the physical and chemical properties of Disperse Red 179 or DAPEP and has consequently used a “read-across” approach to determine the approximate physical and chemical properties in Table 3. These properties were used for further modelling and lines of evidence in this assessment.

An analogue is a chemical which is structurally similar to the substance under assessment and is therefore expected to have similar physical and chemical properties, similar behaviour in the environment, and/or similar toxicity. Where there are experimental data for a given parameter for an analogue substance, these can be used directly or with adjustment to estimate that parameter value for the substance under assessment.

To find acceptable analogues, a review of data for several disperse azo dyes was performed (Anliker et al. 1981; Anliker and Moser 1987; Baughman and Perenich 1988; Savarino et al. 1989; Yen et al. 1989; Yen et al. 1991; Brown 1992; Peters and Gbadamosi 1992; Peters et al. 1992; ETAD 1995; Sijm et al. 1999; Maradiya 2004). These compounds have structural similarities to Disperse Red 179 and DAPEP but also share other important attributes that make them suitable analogues. These include properties affecting their fate in the environment, such as high molecular weights (generally >320 g/mol), similar cross-sectional diameters (1.31–2.11 nm), solid particulate structures, decomposition at greater than 110°C, and “dispersibility” in water (i.e., not truly “soluble”). In addition, they have a negligible vapour pressure and are designed to be stable under environmental conditions.

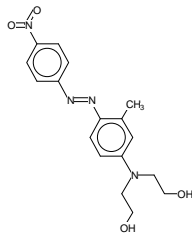
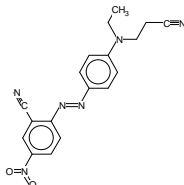
Disperse Red 179 and DAPEP are analogues because of the similarities in their chemical structure and molecular weights. Both substances contain the azo, benzothiazole and cyanide functional groups and both are used as textile dyes. However, slight differences in the physical and chemical properties and behaviour are to be expected. Disperse Red 179 is anticipated to have greater water solubility and a lower log  $K_{ow}$  than DAPEP because of the presence of a nitro group attached to its benzothiazole ring. Similarly, the

two chlorine atoms attached to the benzothiazole group of DAPEP will likely decrease its water solubility and increase its log  $K_{ow}$ .

Structural information on disperse azo analogues to Disperse Red 179 and DAPEP is presented in tables 2a and 2b. Some physical and chemical properties (see Table 3), empirical bioaccumulation data (Table 6), and empirical toxicity data (see Table 7) of these analogues were used in support of the weight of evidence and proposed decisions in this draft screening assessment.

**Table 2a. Information available for Disperse Red 179 and DAPEP, and several structural analogues.**

	CAS RN	Common Name	DSL name	Structure of analogue	Available empirical data
i	25176-89-0	DAPEP	Propanenitrile, 3-[[4-[(5,6-dichloro-2-benzothiazolyl)azo]phenyl]ethylamino]-		Persistence, aquatic toxicity
ii	16586-42-8	Disperse Red 179	Propanenitrile, 3-(ethyl(3-methyl-4-(2-(6-nitro-2-benzothiazolyl)diazenyl)phenyl)amino)-		Persistence, aquatic toxicity
iii	68133-69-7	n/a	Propanenitrile, 3-((2-(acetyloxy)ethyl)(4-(2-(6-nitro-2-benzothiazolyl)diazenyl)phenyl)amino)-		Melting point, solubility in octanol, solubility in water, log $K_{ow}$
iv	70198-17-3	n/a	Ethanol, 2-((4-(2-(6-chloro-2-benzothiazolyl)diazenyl)phenyl)ethylamino)-, 1-acetate		Aquatic toxicity
v	5261-31-4	Disperse Orange 30	Propanenitrile, 3-[[2-(acetyloxy)ethyl][4-[(2,6-dichloro-4-nitrophenyl)azo]phenyl]amino]-		Bioconcentration factor (BCF), aquatic toxicity
vi	31482-56-1	Disperse Orange 25	3-(Ethyl(4-((4-nitrophenyl)azo)phenyl)amino)propanenitrile		Aquatic toxicity

vii	3179-89-3	Disperse Red 17	Ethanol, 2,2'-((3-methyl-4-(2-(4-nitrophenyl)diazenyl)phenyl)amino)bis-		Aquatic toxicity
viii	16889-10-4	Disperse Red 73	2-((4-((2-Cyanoethyl)ethylamino)phenyl)azo)-5-nitrobenzonitrile		Aquatic toxicity

It should be noted that there are several uncertainties associated with the use of the physical and chemical, toxicological, and bioaccumulation data available for the substances presented in Table 2a. All these substances share the same chemical class, disperse azo dyes (with their characteristic azo bond) and are used for similar industrial purposes. However, there are differences between these substances associated with their differences in molecular size and their unique functional groups, notably the presence or absence of benzothiazole, cyano, nitro and/or ester functional groups, or the presence of halogen atoms, such as chlorine on one of the aromatic rings. Further, differences in results for substances may also be caused by analytical error during testing. As a result, these analogues have empirically determined water solubilities that range over four orders of magnitude from  $10^{-5}$  to 0.69 mg/L. It would be preferable to utilize empirical data (e.g., for water solubility and  $\log K_{ow}$ ) specific to the substances being assessed. However, because data are lacking in all areas for monoazo benzothiazole disperse dyes, the analogue data presented are considered the only relevant evidence for the evaluation of these two substances. The variability of the available data was taken into account.

**Table 2b. Comparisons of structural analogues with Disperse Red 179 and DAPEP<sup>1</sup>**

	CAS RN	Common Name	Molecular mass (g/mol)	Structure similarity		Minimum-maximum cross-sectional diameter ( $D_{Max}$ ) in (nm)
				to Disperse Red 179 (%)	to DAPEP (%)	
i	16586-42-9	Disperse Red 179	394.45	100	82.33	1.31–2.11
ii	25176-89-0	DAPEP	404.32	82.33	100	1.41–2.08
iii	68133-69-7	n/a	438.5	89.76	78.47	1.96–2.32
iv	70198-17-3	n/a	402.90	73.71	84.2	1.87–2.31
v	5261-31-4	Disperse Orange 30	450.28	< 60	< 60	1.40–2.10
vi	31482-56-1	Disperse Orange 25	323.35	< 60	< 60	1.37–1.95
vii	3179-89-3	Disperse Red 17	344.36	< 60	< 60	1.41–1.86
viii	16889-10-4	Disperse Red 73	348.36	< 60	< 60	1.31–1.93

<sup>1</sup> ChemID (2009); value presented if > 60%.

Table 3 contains experimental and modelled physical and chemical properties for Disperse Red 179 and DAPEP and structural analogues that are relevant to their environmental fate.

**Table 3. Physical and chemical properties for Disperse Red 179 and DAPEP and available analogues**

Chemical	Type <sup>1</sup>	Value	Temperature (°C)	Reference
Physical State				
Disperse Red 179		Powder		Environment Canada 2009a
		Granular powder		Sarex Overseas 1995
DAPEP		Powder		Environment Canada 2009a
Disperse Red 153 (CAS RN 78564-87-1)	Analogue	Reddish powder or granule		S.M.S Technology, not dated
Melting point <sup>2</sup> (°C)				
DAPEP	Experimental	219–220		Peters et al. 1992
		177–180 <sup>3</sup>		Peters and Gbadamosi 1992
CAS RN unknown (Structural isomer to DAPEP)	Experimental (analogue)	181–182 <sup>4</sup>		Peters et al. 1992
CAS RN 68133-69-7	Experimental (analogue)	172		Yen et al. 1989
		167		Sijm et al. 1999
CAS RN 3771-31-1	Experimental (analogue)	228–230		Maradiya 2004
CAS RN 68083-97-6	Experimental (analogue)	242–243		Maradiya 2004
Disperse Orange 30	Experimental (analogue)	126.9–128.5		ETAD 2005
Benzothiazole azo disperse dyes	Read-across	114–230		Peters et al. 1992
		141–269		Peters and Gbadamosi 1992
		119–243		Savarino et al. 1989
Boiling point <sup>5</sup> (°C)				
Not applicable				

Density (kg/m <sup>3</sup> )				
Disperse Red 153 (CAS RN 78564-87-1)	Not available	950		S.M.S Technology, not dated
Vapour pressure (Pa)				
Azo disperse dyes	Read-across	$5.33 \times 10^{-12}$ to $5.33 \times 10^{-5}$ ( $4 \times 10^{-14}$ to $4 \times 10^{-7}$ mm Hg)	25	Baughman and Perenich 1988
Henry's Law constant (Pa·m <sup>3</sup> /mol)				
Azo disperse dyes	Read-across <sup>6</sup>	$10^{-8}$ to 0.1 ( $10^{-13}$ to $10^{-6}$ atm m <sup>3</sup> /mol)		Baughman and Perenich 1988
Log K <sub>ow</sub> (Octanol-water partition coefficient) (dimensionless)				
Disperse Red 179	Modelled <sup>7</sup>	5.09		KOWWIN 2000
DAPEP	Modelled <sup>7</sup>	6.01		KOWWIN 2000
CAS RN 68133-69-7	Experimental (analogue)	$4.6 (\pm 3.35)^8$		Yen et al. 1989
		4.08 <sup>9</sup>		Sijm et al. 1999
Disperse Orange 30	Experimental (analogue)	4.2		Brown 1992
Azo disperse dyes	Read-across	1.79–5.07		Baughman and Perenich 1988
		> 2–5.1		Anliker et al. 1981; Anliker and Moser 1987
		3.74 to > 5.8		Sijm et al. 1999
Log K <sub>oc</sub> (Organic carbon partition coefficient) (dimensionless)				
Azo disperse dyes	Read-across, calculated <sup>10</sup>	3.4–4.2		Baughman and Perenich 1988
Water solubility (mg/L)				
Disperse Red 179	Modelled <sup>11</sup>	0.012		WATERNT 2002
DAPEP	Modelled <sup>11</sup>	0.004		WATERNT 2002
CAS RN 68133-69-7	Experimental (analogue)	$0.021 \pm 0.004$ $0.690 \pm$ $0.170^{12}$		Sijm et al. 1999
		$0.0079 \pm$ $0.0014$		Yen et al. 1989
Azo disperse dyes	Read-across	< 0.01		Anliker and Moser 1987
		$1.19 \times 10^{-5}$ to 35.46		Baughman and Perenich 1988
n-octanol solubility (mg/L)				
CAS RN 68133-69-7	Experimental (analogue)	$66 \pm 6$		Sijm et al. 1999

Azo disperse dyes	Read-across	81–2430	20	Anliker and Moser 1987
		14.1–3000	20	Sijm et al. 1999
pK <sub>a</sub> (Acid dissociation constant) (dimensionless)				
Disperse Red 179	Modelled	1.9		ACD/pK <sub>a</sub> DB 2005
DAPEP	Modelled	2.05		ACD/pK <sub>a</sub> DB 2005

<sup>1</sup> The extrapolated values used for Disperse Red 179 and DAPEP are based on evidence on disperse dyes submitted to Environment Canada under the *New Substances Notification Regulations (Chemicals and Polymers)* (ETAD 1995) and evidence available from other disperse dye analogues found in literature.

<sup>2</sup> The phrase “melting point” is used but this could be better referred to as a decomposition point because disperse dyes are known to char at high temperatures (greater than 200°C) rather than melt.

<sup>3</sup> The lower melting point value measured by Peters and Gbadamosi (1992) may have been caused by analytical error or variation in measurements.

<sup>4</sup> This melting point value refers to the structural isomer of CAS RN 25176-89-0, which, together with DAPEP, makes up the mixture CAS RN 78564-87-1.

<sup>5</sup> Boiling point is generally not applicable to disperse dyes. For powder dyes, charring or decomposition occurs at high temperatures instead of boiling. For liquids and pastes, boiling will occur only for the solvent component while the unevaporated solid will decompose or char (ETAD 1995).

<sup>6</sup> Solubilities of several disperse dyes at 25 and 80°C were used by Baughman and Perenich (1988) to calculate Henry’s Law constants for these dyes. These values are presented here as a range to illustrate the expected Henry’s Law constant for Disperse Red 179 and DAPEP.

<sup>7</sup> These values were modelled using the “Experimental value adjustment method” of KOWWIN (2000), which estimated the log K<sub>ow</sub> of the substances based on the experimental log K<sub>ow</sub> value of 4.08 for the analogue CAS RN 68133-69-7 (Sijm et al. 1999).

<sup>8</sup> The experimental K<sub>ow</sub> values were measured by Yen et al. (1989) at the dye saturation point using the batch equilibration method. This value is of low confidence, since batch systems are not ideal for determination of large partition coefficients (Yen et al. 1989).

<sup>9</sup> This experimental log K<sub>ow</sub> value (which represents a low-end estimate) was determined using the slow stirring method (De Bruijn et al. 1989).

<sup>10</sup> Log K<sub>oc</sub> values are based on calculations by Baughman and Perenich (1988) using a range of measured solubility for commercial dyes and an assumed melting point of 200°C.

<sup>11</sup> These values were modelled using the “Experimental value adjustment method” of WATERNT (2002), which estimated the water solubility of the substances based on the water solubility values of the analogue CAS 68133-69-7. The water solubility of the analogue (0.0485 55 mg/L) is a geometric average of CAS 68133-69-7 experimental solubility values (Sijm et al. 1999).

<sup>12</sup> The variation in water solubility value is explained by the polymorphic form of the crystal structure of the dyes. Each morphologic form has its own melting point and enthalpy of melting, and these result in different solubility (Sijm et al. 1999).

## Sources

Disperse Red 179 and DAPEP are not naturally produced in the environment.

Recent information was collected through industry surveys conducted for the years 2005 and 2006 under *Canada Gazette* notices issued pursuant to section 71 of CEPA 1999 (Canada 2006b, 2008). These notices requested data on the Canadian manufacture and quantities of the substances imported into Canada. In the notice for 2006, data was also requested on use quantities of Disperse Red 179 and DAPEP.

In response to the CEPA 1999 section 71 survey notice for the 2006 calendar year, no manufacture of Disperse Red 179 or DAPEP was reported above the threshold of 100 kg/year. However, one company reported importing 400 kg of Disperse Red 179 and 100 kg of DAPEP into Canada in 2006 (the exporter country was not identified) (Environment Canada 2009a). Moreover, four companies reported using 400 kg of Disperse Red 179 in 2006, while four additional companies reported using 180 kg of DAPEP in 2006. The larger quantity of 180 kg/yr of DAPEP compared to the import quantity of 100kg for the year 2006 is likely due to unused stocks from previous years. Two other stakeholders were also identified as having an interest in these substances.

Information received in response to the CEPA 1999 section 71 survey notice for the 2005 calendar year determined that between 100 and 1000 kg of Disperse Red 179 were in commerce in Canada (Environment Canada 2006). No reports of manufacture in Canada or import into Canada of DAPEP at or above the reporting threshold of 100 kg in the 2005 calendar year were received in response to the same notice (Environment Canada 2006). However, one stakeholder was identified as having an interest in these substances.

The quantities reported under the Domestic Substances List (DSL) as manufactured in, imported into, or in commerce in Canada during the 1986 calendar year for Disperse Red 179 were between 1000 and 10 000 kg. The quantities reported under the DSL as manufactured in, imported into, or in commerce in Canada during the 1986 calendar year for DAPEP were between 100 and 1000 kg.

Production of Disperse Red 179 in the United States has been estimated to be between 10 000 and 500 000 pounds (approximately 4500-230 000 kg) in each of the following years: 1986, 1990, 1994 and 1998 (US EPA 2009). However, no quantity was reported for 2002 (US EPA 2009). DAPEP was not produced in the United States during this period (US EPA 2009).

## Uses

Information on uses in the 2005 and 2006 calendar years was gathered in response to the CEPA 1999 section 71 notices (Canada 2006b, 2008).

In 2006, the company importing Disperse Red 179 and DAPEP identified its business activity as “Chemical (except Agricultural) and allied product wholesaler distributor.” Disperse Red 179 is reportedly used as a dye in the chemical colourant Foron Rubine RD-S, while DAPEP is used in the chemical colourant Foron Scarlet RD-S (Environment Canada 2009a). Disperse Red 179 was reported to be in commerce in Canada in 2005 under the same business activity group, namely “Chemical (except Agricultural) and allied product wholesaler distributor” (Environment Canada 2006).

The following DSL use codes have been identified for Disperse Red 179 during the DSL nomination (1984-1986): “Colourant - pigment/stain/dye/ink” and “Textile, Product.” Only the DSL use code “Colourant – pigment/stain/dye/ink” was identified for DAPEP.

Review of the available technical information indicates that Disperse Red 179 and DAPEP are red disperse dyes used in the textile industry to colour synthetic fabrics such as polyesters and polyamides (Danish EPA 1998). They may be used as dyeing agents for synthetic fibres for clothing and home textile uses (CII 2002–; Choi et al. 2007; Environment Canada 2009a).

## **Releases to the Environment**

According to information received in response to the CEPA 1999 section 71 survey notice for the year 2006, the most important direct release of the dye to the environment occurs in the textile industry following the dyeing process when the unfixed dye is washed off of the fibres and discharged with wastewater. Most textile mills in Canada discharge their wastewater to treatment plants with primary or secondary capabilities, either municipal or located at the facility (Environment Canada 2009a).

### ***Mass Flow***

To estimate potential releases of substances to the environment at different stages of their life cycle, a Mass Flow Tool was developed (Environment Canada 2009b). Empirical data concerning releases of specific substances to the environment are seldom available. Therefore, for each identified type of use of the substance, the proportion and quantity released to the various environmental media are estimated, as is the proportion of the substance chemically transformed or sent for waste disposal. Unless specific information on the rate or potential for release of the substance from landfills and incinerators is available, the Mass Flow Tool does not quantitatively account for off-site releases to the environment from waste disposal sites.

Assumptions and input parameters used in making the release estimates are based on information obtained from a variety of sources including responses to regulatory surveys, Statistics Canada, manufacturers’ websites, technical databases and documents, and professional knowledge and assumptions. Of particular relevance are emission factors,



which are generally expressed as the fraction of a substance released to the environment, particularly during its manufacture, processing, and use associated with industrial processes. Sources of such information include emission scenario documents, often developed under the auspices of the Organisation for Economic Co-operation and Development (OECD), and default assumptions used by different international chemical regulatory agencies. It is noted that the level of uncertainty in the mass of substance and quantity released to the environment generally increases toward the end of the life cycle.

**Table 4. Estimated releases and losses of Disperse Red 179 and DAPEP to environmental media, chemical transformation during life cycle and transfer to waste disposal sites, based on the Mass Flow Tool**

<b>Fate</b>	<b>Proportion of the mass (%)<sup>1</sup></b>	<b>Major life cycle stage involved<sup>2</sup></b>
<b>Released to receiving media:</b>		
To soil	0	n/a
To air	0	n/a
To wastewater <sup>3</sup>	17.1	Manufacturing of products, Consumer use
<b>Chemically transformed (incineration)</b>	2.5	Waste disposal
<b>Transferred to waste disposal sites (e.g., landfill, incineration)</b>	80.5	Waste disposal

<sup>1</sup> For each substance, information from the following OECD emission scenario documents was used to estimate releases to the environment and the distribution of the substance as summarized in this table: Adhesive formulation (OECD 2004), and Textile manufacturing wool mills (OECD 2007). Specific assumptions used in the derivation of these estimates are summarized in Environment Canada (2009c) and Environment Canada (2009d).

<sup>2</sup> Applicable stage(s): production, formulation, industrial use, consumer use, service life of article/product, waste disposal.

<sup>3</sup> Wastewater before any form of treatment, either on-site industrial or off-site municipal wastewater treatment.

Based on Statistics Canada information and an analysis by both Environment Canada and Industry Canada, it is recognized that dyes may be imported in manufactured articles. Following the Statistics Canada proposal, a ratio of 30:70 (textiles manufactured in Canada versus imported) was used to estimate the amount of dye imported in coloured products (Environment Canada 2009a). The import quantity of 400 kg for Disperse Red 179 and the use quantity of 180 kg for DAPEP for the year 2006 were included in the Mass Flow Tool calculations, yielding approximate total quantities of Disperse Red 179 and DAPEP in commerce in Canada of 1 128 kg and 508 kg respectively.

Results indicate that Disperse Red 179 and DAPEP can be expected to be found largely in waste disposal sites (80.5% or 908 kg/year of Disperse Red 179 and 408 kg/year of DAPEP), due to the eventual disposal of manufactured items containing them. A small fraction of solid waste is incinerated, which is expected to result in chemical transformation of the substance. Based largely on information contained in OECD emission scenario documents for processing and uses associated with this type of substance (OECD 2004, 2007), it is estimated that 17.1% (192 kg/year of Disperse Red 179 and 87 kg/year of DAPEP) may be released to wastewater, mainly resulting from activities associated with their industrial use (7.9 %) but also from the service life of

products containing the substances such as releases associated with laundry washing (9.2%). Although not considered by the Mass Flow Tool, it should be noted that these dyes may be applied to agricultural soils and pasture lands in Canada as a component of wastewater treatment biosludge, which is commonly used for soil enrichment. The potential loss to groundwater from the portion of substances finding their way into landfill sites (through the disposal of manufactured items) is anticipated to be limited. For the ecological assessment, the aquatic environment is considered the critical medium (based on releases from wastewater treatment plants).

## Environmental Fate

As indicated by the results of the Mass Flow Tool (Table 4), the substances Disperse Red 179 and DAPEP are expected to be released to wastewater during industrial processing and consumer use (Environment Canada 2009c, 2009d). The high  $\log K_{ow}$  (analogues 4.08–4.6, read across > 4 and modelled values 5.09–6.01) and high  $\log K_{oc}$  (read across 3.4 to 4.2) values (see Table 2) indicate that these substances may have affinity for solids. However, the  $\log K_{oc}$  is a calculated value (see footnote 3 below Table 2) for azo disperse dyes without a benzothiazole functional group, and the adsorption potential of solid particulate dye structures is generally not well understood. Therefore, the degree of this particular behaviour for the two substances being assessed is uncertain.

Disperse Red 179 and DAPEP do not biodegrade rapidly under aerobic conditions (see Table 5). Disperse Red 179 and DAPEP are used in the form of powders with limited water solubility (see Table 3). In solution, Disperse Red 179 and DAPEP behave as bases with very low estimated  $pK_a$  (2.05 and 1.9, respectively; see Table 3). Consequently, dissolved forms of either substance are not expected to ionize in water at environmentally relevant pHs (6–8 for surface waters). Because of their low solubility, these substances are expected to behave as colloidal dispersions when released into water (Yen et al. 1991). They will therefore mostly be present as solids or adsorbed to suspended particles and will eventually sink to bed sediments, where they are expected to remain in a relatively biologically unavailable form. Yen et al. (1989) concluded that disperse dyes tend to accumulate extensively in sediments and biota unless they are degraded at rates comparable to uptake. Razo-Flores et al. (1997) have stated that, due to the recalcitrant nature of azo dyes in the aerobic environment, they eventually end up in anaerobic sediments due to sediment burial, in shallow aquifers or in groundwater. Yen et al. (1991) observed that an azo benzothiazole analogue was transformed under anaerobic conditions in sediment via hydrolysis and reduction, and concluded that most azo dyes would likely not persist in anaerobic sediment systems.

The rate of volatilization from the surface of water is proportional to the Henry's Law constant (Baughman and Perenich 1988). Baughman and Perenich (1988) also state that volatilization from aquatic systems will not be an important loss process for disperse dyes. This statement agrees with the low to negligible read-across Henry's Law constant values ( $10^{-8}$  to  $0.1 \text{ Pa}\cdot\text{m}^3/\text{mol}$ ; Table 3) as well as the low analogue vapour pressure ( $5.33 \times 10^{-12}$  to  $5.33 \times 10^{-5}$ ; Table 3). Based on these analogue and read-across data for disperse

azo dyes, transport in air due to the loss of this substance from moist and dry soil surfaces is not likely to be significant. These data are consistent with the physical state (solid particulate structure) of Disperse Red 179 and DAPEP; this state does not make them likely candidates for volatilization.

## **Persistence and Bioaccumulation Potential**

### **Environmental Persistence**

No environmental monitoring data have been identified relating to the presence of Disperse Red 179 or DAPEP in the Canadian environment (air, water, soil or sediment).

According to the Ecological and Toxicological Association of Dyes and Organic Pigments Manufacturers, dyes are, with some exceptions, considered essentially non-biodegradable under aerobic conditions (ETAD 1995). Repeated evaluation of ready and inherent biodegradability using accepted screening tests (see OECD Guidelines for Testing Chemicals) have confirmed this assumption (Pagga and Brown 1986; ETAD 1992). Based on the chemical structure of Disperse Red 179 and DAPEP, there is no reason to suspect that biodegradation will be other than that described for dyes generally (ETAD 1995).

Some disperse azo dyes, including benzothiazole compounds; have been shown to undergo relatively rapid anaerobic degradation in sediment at depth, where anoxic conditions prevail (Yen et al. 1991; Baughman and Weber 1994; Weber and Adams 1995). Disperse dyes enter the aquatic system mostly as a dispersion of fine suspended particles and eventually settle to the aerobic layers of surface sediment, where they will persist until sediment burial creates reducing conditions. The rate of sediment deposition and the extent of bioturbation varies from site to site and it is thus very difficult to ascertain the residence time of dyes in aerobic sediment layers. It is likely, however, that in many cases this is greater than 365 days. Once under anaerobic or reducing conditions, azo dyes may undergo rapid degradation to substituted aromatic amine constituents, as demonstrated by Yen et al., (1991) who measured reduction half-life values in compacted sediments at room temperature of 1.9–2.0 days for an azo benzothiazole dye (CAS 68133-69-7). However, most aquatic organisms are not expected to be exposed to these biodegradation transformation products in deep anoxic sediments, in part because contact with anoxic sediment is likely to be limited and in part because the amine degradation products are expected to be tightly bound to sediments, so that they would have very low bioavailability (Weber et al. 2001; Colon et al. 2002). Therefore, the degradation products are not likely to present an ecological concern.

Empirical biodegradation data were submitted by industry in response to the CEPA 1999 section 71 survey notice for the 2006 calendar year (Environment Canada 2009a). Inherent biodegradability studies evaluating the aerobic biodegradability in an aqueous medium of Foron Rubin RD-S (a commercial product that contains CAS RN 16586-42-8) and Disperse Red 153 (a commercial product that contains CAS RN 25176-89-0)

determined that neither compound was biodegradable (BMG 2001, 2003a). These tests were performed according to OECD Guidelines for Testing of Chemicals, Test No. 302B-1992, “Inherent Biodegradability: Zahn-Wellens / EMPA Test.” Although the protocol used in these two studies was acceptable, there is a general lack of information on the substances used in each test. In neither study is the solubility of the compounds being tested reported. In the first study, neither the proportion of Disperse Red 179 (CAS RN 16586-42-8) in the commercial product Foron Rubin RD-S nor the other components of this commercial product were reported. In the second study, the proportion of DAPEP present in Disperse Red 153 (CAS RN 78564-87-1), which is actually a mixture of DAPEP and another structural isomer (Nakagawa 1996; CII 2002— ), is not reported.

The absence of degradation could be explained by bacterial inhibition caused by Disperse Red 179 and DAPEP toxicity. However, respiration inhibition test studies performed on the same compounds according to OECD Guidelines for Testing of Chemicals, Test No. 209-1984, determined that activated sludge showed no significant toxic effects from either of the tested substances (BMG 2000a, 2003b). EC<sub>20</sub> and EC<sub>80</sub> for Foron Rubin RD-S (CAS RN 16586-42-8) and Disperse Red 153 (which contains CAS RN 25176-89-0) were estimated to be above 1000 mg/L and 4000 mg/L, respectively (BMG 2000a, 2003b). Based on this additional information, the two inherent biodegradability studies are deemed acceptable, despite the lack of clarity regarding the composition of the test substances (see Appendix 1).

Table 5a presents the empirical biodegradation data (BMG 2001, 2003a) that show no biodegradation over 28 days in an inherent-biodegradation test for Foron Rubin RD-S and Disperse Red 153. These tests indicate that the half-life in an oxic aqueous medium is likely to be longer than 182 days (6 months) and that the substances are therefore likely to persist under aerobic conditions in that environmental compartment.

**Table 5a. Empirical data for inherent degradability of Disperse Red 179 and DAPEP**

Substance	Medium	Fate process	Degradation value	Degradation endpoint / units	Reference
Foron Rubin RD-S (Disperse Red 179)	Water/activated sludge	Biodegradation	0	28-day biodegradation / %	BMG 2003a
Disperse Red 153 (DAPEP)	Water/activated sludge	Biodegradation	0	28-day biodegradation / %	BMG 2001

Since few experimental data on the degradation of Disperse Red 179 and DAPEP are available, a QSAR-based weight-of-evidence approach was also applied using the degradation models shown in Table 5b. Although the expected release of Disperse Red 179 and DAPEP will be to wastewater, their residence time in the water column, due to

their low solubility and their behaviour as colloidal dispersions, may be short before they sink to the sediment bed. However, given the lack of data regarding this issue, persistence in water was examined using predictive QSAR models for biodegradation. The following analysis applies primarily to the portion of this substance that is present in the environment in the dissolved form, recognizing that a significant proportion would also likely exist in a dispersed form as solid particles. Table 5b summarizes the results of available QSAR models for biodegradation in water for Disperse Red 179 and DAPEP.

**Table 5b. Modelled data for degradation of Disperse Red 179 and DAPEP**

Fate process	Model and model basis	Substance	Model result and prediction	Extrapolated half-life (days)
<b>WATER</b>				
Biodegradation (aerobic)	BIOWIN 2000 Sub-model 3: Expert Survey (ultimate biodegradation)	Disperse Red 179	1.445 <sup>1</sup> “biodegrades very slowly”	> 182
		DAPEP	1.2549 <sup>1</sup> “biodegrades very slowly”	> 182
Biodegradation (aerobic)	BIOWIN 2000 Sub-model 5: MITI linear probability	Disperse Red 179	-0.4686 <sup>2</sup> “biodegrades very slowly”	> 18 <sup>2</sup>
		DAPEP	-0.3395 <sup>2</sup> “biodegrades very slowly”	> 182
Biodegradation (aerobic)	BIOWIN 2000 Sub-model 6: MITI non-linear probability	Disperse Red 179	0 <sup>2</sup> “biodegrades very slowly”	> 182
		DAPEP	0 <sup>2</sup> “biodegrades very slowly”	> 182
Biodegradation (aerobic)	TOPKAT 2004 Probability	Disperse Red 179	0.0 <sup>2</sup> “biodegrades very slowly”	> 182
		DAPEP	0.0 <sup>2</sup> “biodegrades very slowly”	> 182
Biodegradation (aerobic)	CATABOL 2008 % BOD (biological oxygen demand)	Disperse Red 179	% BOD = 3.3 “Biodegrades very slowly”	> 182
		DAPEP	% BOD = 3.1 “Biodegrades very slowly”	> 182

<sup>1</sup> Output is a numerical score.

<sup>2</sup> Output is a probability score.

Results from Table 5b show that the the two BIOWIN probability models (5 and 6) suggest these substances biodegrade slowly and that their half-life in water would be >182 days. In fact, both probability results are much less than 0.3, the cut-off suggested by Aronson et al. (2006) for identifying substances as having a half-life > 60 days (based on the MITI probability models). The ultimate survey model (BIOWIN 3) result of “biodegrades very slowly” is suggested to mean 180 to 240 days (US EPA 2002a;

Aronson et al. 2006). The overall conclusion from BIOWIN (2000) is that these substances are not readily biodegradable.

Other ultimate degradation models (CATABOL and TOPKAT) predict that Disperse Red 179 and DAPEP do not undergo mineralization in a 28-day timeframe with probability or extent of biodegradation in the range of very persistent chemicals. TOPKAT, which simulates the Japanese MITI 28-day biodegradation test, produced a probability of 0 for both substances. This is much less than the suggested cut-off for persistent substances in this model ( $< 0.3$ ) (0.7 is suggested for non-persistent chemicals) (TOPKAT 2004). CATABOL predicted only 3.3% and 3.1% biodegradation for Disperse Red 179 and DAPEP, respectively, based on the OECD 301 ready biodegradation test (% BOD). This has been suggested as meaning “likely persistent” (Aronson and Howard 1999) and having a half-life in water of  $>182$  days. The modeled values in Table 5b are considered reliable as several chemicals of structural comparability are contained in their training sets.

When the results of the empirical inherent biodegradation tests as well as the predictive models are considered together, there is a consensus suggesting that the ultimate degradation half-life in water is  $> 182$  days, which is consistent with what would be expected for chemicals used as a disperse dye (i.e., manufactured to be relatively insoluble and durable). Using a 1:1:4 ratio for a water:soil:sediment half-life extrapolation (Boethling et al. 1995), the half-life in soil is also  $>182$  days and the half-life in oxic sediments is  $> 365$  days.

Based on the results of experimental data, predictive modelling and expert judgement (ETAD 2005), Disperse Red 179 and DAPEP meet the persistence criteria in water and soil (half-lives in soil and water  $\geq 182$  days) and half-life in sediment  $\geq 365$  days), as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

### Potential for Bioaccumulation

No experimental bioaccumulation factor (BAF) and/or bioconcentration factor (BCF) data for Disperse Red 179 or DAPEP were available; therefore, empirical bioconcentration test data for fish using the analogue substance Disperse Orange 30 (Shen and Hu 2008) were used to determine the bioaccumulation potential of the substances subject to this assessment.

The chemical structure and molecular weight of Disperse Orange 30 are similar to those of DAPEP and especially Disperse Red 179, with the greatest differences being that Disperse Orange 30 has an ester group but is lacking a benzothiazole functional group. The bioavailability of most disperse dyes is generally considered to be very low (and this limits bioaccumulation potential); however, based only on structural considerations, it is possible that the bioaccumulation potential of DAPEP will be slightly greater than the bioaccumulation potential of Disperse Red 179 because of the presence of the two chlorine atoms on its benzothiazole group.

A bioconcentration study of Disperse Orange 30 found that it is unlikely to accumulate in fish (Shen and Hu 2008). This study was performed according to OECD Guidelines for Testing of Chemicals, Test No. 305B-1996, Bioconcentration: Semi-Static Fish Test. The bioconcentration of Disperse Orange 30 in zebra fish (*Brachydanio rerio*) was determined in a 28-day semi-static test with test medium renewal every two days. An exposure test at a nominal concentration of 20 mg/L (mean measured concentration 0.028 ~ 0.28 mg/L) was performed (in accordance with the result of a fish acute toxicity test) to check the bioconcentration potential of the test substance. Samples from both test solutions and test organisms were taken daily from Day 26 to Day 28 of the 28-day exposure test period. Samples were prepared by extracting the lipid component from the test fish. The measured concentration of test substance, fish lipid content and BCF calculation are reported in Table 6a.

**Table 6a. Measured concentration of Disperse Orange 30, fish lipid content and BCF calculation**

		Sampling Time		
		Day 26	Day 27	Day 28
<b>Treatments (20 mg/L)</b>	Measured concentration of the test substance in extracted solutions (mg/L)	< 0.028	< 0.028	< 0.028
	Content of the test substance in the fish lipids (mg)	< 1.68	< 1.68	< 1.68
	Fish total weight (g)	2.07	2.13	2.53
	Concentration of the test substance in the fish $C_f$ (mg/kg)	< 0.81	< 0.79	< 0.66
	Measured concentration of the test substance in the water $C_w$ (mg/L)	0.028 ~ 0.28	0.028 ~ 0.28	0.028 ~ 0.28
	Fish lipid content (%)	0.81	0.57	1.25
	BCF	< 100	< 100	< 100
	Average BCF	< 100		

The Shen and Hu (2008) study has been reviewed and considered acceptable (see Appendix 1). Lack of detection in fish extracts (< 0.028 mg/L) suggests a limited solubility in lipids and/or limited potential to partition into fish tissue from aqueous systems (more likely both). However, there is some uncertainty associated with limit-bounded values in any study because the “true” value is not known.

Given the structure of the substance and the likely behaviour of this class of disperse dye in aqueous systems, a low BCF result would be expected. Most disperse dyes, as their name would suggest, exist as fine dispersible particles with limited truly soluble fractions. Solubility, however, can be increased by adding polar functional groups to the molecule. Disperse Orange 30 contains some of these solubilizing groups (nitroso), so some degree of water solubility would be expected. Assuming that the concentration in

solution in the test was equal to the lowest water solubility value of 0.028 mg/L, and using the fish concentration of 0.81 mg/kg as a worst-case estimate, the BCF may be calculated to be < 100.

The above study serves as primary evidence to support the lack of bioaccumulation potential of Disperse Red 179 and DAPEP, and other research supports this conclusion. Anliker et al. (1981) reported experimental fish bioaccumulation values for 18 disperse monoazo dyes, performed according to test methods specified by the Japanese Ministry of International Trade and Industry (MITI). Expressed on the basis of wet body weight of the fish, these log bioaccumulation factors ranged from 0.00 to 1.76 (Anliker et al. 1981). A lack of reporting of chemical registry numbers and chemical structures limited the utility of this study for read-across purposes to Disperse Red 179 and DAPEP. However, follow-up studies, which provided the chemical structures for the disperse dyes tested, confirmed low bioaccumulation potential for 10 nitroazo dyes, with reported log bioaccumulation factors ranging from 0.3 to 1.76 (Anliker and Moser 1987; Anliker et al. 1988). Studies available from MITI also support low bioaccumulation potential for azo disperse dyes. Reported BCFs for three azo disperse dyes (CAS RN 40690-89-9, 61968-52-3 and 71767-67-4) tested at a concentration of 0.01 mg/L were in the range of < 0.3 to 47 (MITI 1992). An accumulation study by Brown (1987) also showed that none of the 12 disperse dyes tested accumulated during an eight-week study with carp.

Although lack of significant bioavailability in water and food is expected to significantly mitigate the uptake potential of most disperse dyes, the empirical log  $K_{ow}$  for a close analogue, based on the data from Sijm et al. (1999), suggests that Disperse Red 179 and DAPEP could be soluble in lipids should environmental conditions promote the bioavailability of these substances to fish. Corrected log  $K_{ow}$  values of 5.09 and 6.01 were estimated for Disperse Red 179 and DAPEP from the known and acceptable log  $K_{ow}$  value of 4.08 (Sijm et al. 1999) for the close analogue CAS RN 68133-69-7 using the Expert Value Adjustment method of KOWWIN (2000). In the EVA approach, the estimate begins with the experimental log  $K_{ow}$  of the similar compound. The similar structure is then modified by subtracting and adding fragments to “build” the compound being estimated. The estimate then becomes the sum of the experimental value and the value of the fragment modifications.

Therefore, a log  $K_{ow}$  value of 4.08 for the analogue CAS RN 68133-69-7 and elevated corrected log  $K_{ow}$  values of 5.09 and 6.01 for Disperse Red 179 and DAPEP, respectively (Table 2), are the only lines of evidence to suggest that these substances may have a potential for significant bioaccumulation. In spite of their high  $K_{ow}$  values, evidence for bioaccumulation of disperse azo dyes is lacking (Anliker et al. 1981; Anliker and Moser 1987; MITI 1992). Authors who have found high log  $K_{ow}$  values and concomitant low bioaccumulation factors for azo disperse dyes suggest the low accumulation factors may be due in some cases to the low absolute fat solubility of these substances (Brown 1987) or to the relatively high molecular weight (typically 450–550 g/mol). Low fat solubility and high molecular weight may make transport across fish membranes difficult (Anliker et al. 1981; Anliker and Moser 1987). It is also likely that the lack of bioavailability and



the limited capacity to partition under BCF test conditions, as well as in vivo metabolic degradation, limit accumulation in fish lipids.

It has been stated by ETAD (1995) that the molecular characteristics indicating the absence of bioaccumulation are a molecular weight of  $> 450$  g/mol and a cross-sectional diameter of  $> 1.05$  nm. Recent investigation by Dimitrov et al. (2002), Dimitrov et al. (2005) and the BBM (2008) suggests that the probability of a molecule crossing cell membranes as a result of passive diffusion declines significantly with increasing maximum cross-sectional diameter ( $D_{\max}$ ). The probability of passive diffusion falls appreciably when cross-sectional diameter is greater than  $\sim 1.5$  nm and falls more significantly when molecules have a cross-sectional diameter of  $> 1.7$  nm. Sakuratani et al. (2008) have also investigated the effect of cross-sectional diameter on passive diffusion in a test set of about 1200 new and existing chemicals. They observed that substances that do not have a very high bioconcentration potential often have a  $D_{\max}$  of  $> 2.0$  nm and an effective diameter ( $D_{\text{eff}}$ ) of  $> 1.1$  nm.

Disperse Red 179 and DAPI have a molecular weight of 394.45 and 404.32 g/mol, respectively (see Table 1), and their molecular structures are relatively uncomplicated; both these characteristics suggest some bioaccumulation capability of these substances. There are no clear relationships for establishing strict molecular size cut-offs for assessing bioaccumulation potential; however, a reduction in uptake rate can be associated with increasing cross-sectional diameter, as demonstrated by Dimitrov et al. (2002, 2005). The maximum diameter of Disperse Red 179 and DAPI (and their conformers) ranges from 1.3 to 2.108 nm and 1.414 to 2.075 nm, respectively (BBM 2008), suggesting that a potential for a significantly reduced uptake rate from water and in vivo bioavailability exists with these dyes.

Based on a lack of accumulation in bioconcentration tests with the analogue substance Disperse Orange 30 and other related azo disperse dyes and on the large molecular sizes of Disperse Red 179 and DAPI, these substances are expected to have a low potential for bioaccumulation. Therefore, Disperse Red 179 and DAPI do not meet the bioaccumulation criteria ( $\text{BCF}$  or  $\text{BAF} \geq 5000$ ), as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

## Potential to Cause Ecological Harm

### Ecological Effects Assessment

#### A - In the Aquatic Compartment

Few empirical ecotoxicity data were identified for Disperse Red 179 or DAPEP. Acute toxicity studies on two commercial products containing Disperse Red 179 and DAPEP using *Poecilia reticulata* (guppy) were submitted to Environment Canada in January 2009 (BMG 2000b, 2003c). Both studies were conducted according to OECD Guideline Procedure 203 (Fish acute toxicity testing) and EEC directive 92/69/EEC (Acute toxicity for fish). Results of both studies are presented in Table 7a.

In the first study, the toxicity of Disperse Red 179 was investigated using an aquatic toxicity screening test using the commercial product Foron Rubin RD-S (BMG 2003c). The test reported an acute 96-hour LC<sub>50</sub> of between 10 and 100 mg/L, and a no-observed-effect concentration (NOEC) of 10 mg/L, based on nominal concentrations. An assessment of the reliability of the study using a robust study summary found that the study was deemed to be of “low confidence” due to lack of details on the test substance (Appendix 1). Indeed, neither the proportion of Disperse Red 179 in Foron Rubin RD-S nor the solubility of Foron Rubin RD-S is reported.

The second aquatic toxicity test study (BMG 2000b) was conducted on Disperse Red 153, a substance that contains DAPEP and a structural isomer of DAPEP in unknown proportion (Nakagawa et al. 1996; CII 2002–). The test determined a NOEC 100 mg/L. This result may be interpreted as meaning that no acute effects were observed at saturation of the substance. Like the previous study, the study is considered to be of “low confidence” due to lack of details on the test substance (Appendix 1).

Although both studies are considered to be of low confidence, the results obtained in both studies are typical for disperse azo dyes.

**Table 7a. Empirical data for aquatic toxicity of Disperse Red 179 and DAPEP**

Test substance	Test organism	Type of test	Endpoint	Value (mg/L)	Reference
Foron Rubin RD-S (Disperse Red 179)	<i>Poecilia reticulata</i> (guppy)	Acute (96 hours)	LC <sub>50</sub> <sup>1</sup>	10–100 mg/L	BMG 2003c
			NOEC <sup>2</sup>	10 mg/L	
C.I. Disperse Red 153	<i>Poecilia reticulata</i> (guppy)	Acute (96 hours)	NOEC <sup>2</sup>	100 mg/L	BMG 2000b

<sup>1</sup> LC<sub>50</sub> – The concentration of a substance that is estimated to be lethal to 50% of the test organisms.

<sup>2</sup> NOEC – The no-observed-effect concentration is the highest concentration in a toxicity test not causing a statistically significant effect in comparison to the controls.

Empirical toxicity data are also available for another close analogue of both substances, ethanol, 2-((4-(2-(6-chloro-2-benzothiazolyl)diazenyl)phenyl)ethylamino)-, 1-acetate, CAS RN 70198-17-3 (see Table 7b). The molecular weight of this monoazo benzothiazole disperse dye (404.9 g/mol) and its chemical structure are similar to those of Disperse Red 179 and DAPEP. The 96-hour static toxicity test of the substance added to aquaria in a 0.05% acetone carrier was conducted with daphnids, flatworms, fathead minnows and snails (Health, Safety, and Human Factors Laboratory 1978). Results indicated low toxicity for fathead minnow and snails ( $LC_{50}$  values of  $> 100$  mg/L) and low toxicity for flatworms ( $LC_{50} = 32$  mg/L) but elevated toxicity for daphnids ( $LC_{50} = 0.12$  mg/L) (Health, Safety, and Human Factors Laboratory 1978). The low toxicity value of 0.12 mg/L for daphnids is of concern, but these data are considered of low confidence since the reliability of the toxicity testing could not be assessed due to a general lack of details reported in the study and the age of the study itself.

Environment Canada received ecotoxicological data for a structurally similar substance under the *New Substances Notification Regulations (Chemicals and Polymers)* (Environment Canada 1994) (see Table 7b). The molecular weight of this notified substance was 418.35 g/mol, which is similar to the molecular weight of Disperse Red 179 and DAPEP. The results for the 96-hour static toxicity test with rainbow trout on a substance containing 5% of the notified substance revealed that the  $LC_{50}$  for this species is 10 mg/L. However, while this toxicity value suggests moderate to low acute toxicity to fish, it was not considered indicative of the notified material due to the low concentration of notified substance in the tested product.

**Table 7b. Empirical data for aquatic toxicity for close analogues of Disperse Red 179 and DAPEP**

Common name or CAS RN	Test organism	Duration (hours)	End point	Reliability of the study	Value (mg/L)	Reference
CAS RN 70198-17-3	Fathead minnows	96	$LC_{50}^1$	Not available	$> 100$	Health, Safety, and Human Factors Laboratory 1978
	Snails	96	$LC_{50}^1$	Not available	$> 100$	
	Flatworms	96	$LC_{50}^1$	Not available	32	
	Daphnids	96	$LC_{50}^1$	Not available	0.12	
Confidential	Rainbow trout	96	$LC_{50}^1$	Low confidence	10	Environment Canada 1994

<sup>1</sup> $LC_{50}$  – The concentration of a substance that is estimated to be lethal to 50% of the test organisms.

Empirical toxicity data are available for the close analogue Disperse Orange 30 (see Table 7c). According to a study submitted to Environment Canada on behalf of ETAD (Brown 1992), a 96-hour  $LC_{50}$  of 710 mg/L for zebra fish, a 48-hour  $EC_{50}$  of 5.8 mg/L for *Daphnia magna*, and a 72-hour  $EC_{50}$  of 6.7 mg/L (growth) for *Scenedesmus subspicatus* have been obtained experimentally based on a toxicity study using Disperse Orange 30. However, the original studies have not been provided and their reliability therefore cannot be verified. Another result for Disperse Orange 30 established an  $LC_{50}$  for rainbow trout (*Oncorhynchus mykiss*) of  $> 700$  mg/L (Sandoz 1975). However, this study was, after review, considered to be unacceptable (see Appendix 1). Finally, another acute

toxicity study, using rainbow trout and submitted to Environment Canada in August 2008, indicated a 96-hour LC<sub>50</sub> of > 100 mg/L (Safepharm Laboratories Ltd. 1990). The assessment of the reliability of the study using a robust study summary deemed the study to be of “low confidence” due to lack of details (Appendix 1).

**Table 7c. Empirical data for aquatic toxicity for the analogue Disperse Orange 30**

Test organism	Type of test	Duration (hours)	Endpoint	Reliability of the study	Value (mg/L)	Reference
Rainbow trout	Acute	48	LC <sub>50</sub> <sup>1</sup>	Unacceptable	> 700	Sandoz 1975
Rainbow trout	Acute	96	LC <sub>50</sub>	Low confidence	> 100	Safepharm Laboratories Ltd. 1990
Zebra fish	Acute	96	LC <sub>50</sub>	Not available	710	Brown 1992
<i>Daphnia magna</i>	Acute	48	EC <sub>50</sub> <sup>2</sup>	Not available	5.8	
<i>Scenedesmus subspicatus</i>	Acute	72	EC <sub>50</sub>	Not available	6.7	
Bacteria	Acute	n/a	IC <sub>50</sub> <sup>3</sup>	Not available	> 100	

<sup>1</sup> LC<sub>50</sub> – The concentration of a substance that is estimated to be lethal to 50% of the test organisms.

<sup>2</sup> EC<sub>50</sub> – The concentration of a substance that is estimated to have some toxic sublethal effect on 50% of the test organisms.

<sup>3</sup> IC<sub>50</sub> – The concentration of a substance that is estimated to inhibit growth in 50% of the test organisms.

In another study, a summary of which was submitted to Environment Canada on behalf of ETAD (Brown 1992), 11 disperse dyes were tested on the following organisms: zebra fish, *Daphnia magna*, algae and bacteria. In this study there were some disperse dyes (non-azo compounds) that had toxicity levels reported as < 1 mg/L for algae. However, Brown (1992) reported that inhibition of growth in algae was due largely to light absorption by the dyes rather than biological activity. Three of the disperse dyes tested by Brown (1992) are analogues of Disperse Red 179 and DAPEP. These are Disperse Red 73, Disperse Orange 25, and Disperse Red 17 (Table 7c). These analogues showed moderate toxicity in *D. magna* (48-hour EC<sub>50</sub> = 23–110 mg/L) and moderate to low toxicity in zebra fish (96-hour LC<sub>50</sub> = 17–268 mg/L) (see Table 7d). Moderate toxicity was also observed for algae growth (EC<sub>50</sub> for growth = 7–54 mg/L), and no toxicity was detected for bacteria (IC<sub>50</sub> > 100 mg/L). The experimental details for the dyes tested were not provided, greatly limiting evaluation of these studies (Brown 1992). However, these data were considered usable and are included in this screening assessment as part of the weight of evidence, as they are in agreement with other data and concur with expected range of ecotoxicity values for these structures. These values would also therefore suggest that neither Disperse Red 179 nor DAPEP is highly hazardous to aquatic organisms.

**Table 7d. Empirical data for aquatic toxicity for analogues of Disperse Red 179 and DAPEP**

Common Name or CAS#	Test organism	Duration (hours)	Endpoint	Value (mg/L)	Reference
Disperse Red 73	Zebra fish	96	LC <sub>50</sub> <sup>1</sup>	17	Brown 1992
	<i>Daphnia magna</i>	48	EC <sub>50</sub> <sup>2</sup>	23	
	<i>Scenedesmus subspicatus</i>	72	EC <sub>50</sub> <sup>2</sup>	> 10	
	Bacteria	n/a	IC <sub>50</sub> <sup>3</sup>	> 100	
Disperse Red 17	Zebra fish	96	LC <sub>50</sub> <sup>1</sup>	103	Brown 1992
	<i>Daphnia magna</i>	48	EC <sub>50</sub> <sup>2</sup>	98	
	<i>Scenedesmus subspicatus</i>	72	EC <sub>50</sub> <sup>2</sup>	7	
	Bacteria	n/a	IC <sub>50</sub> <sup>3</sup>	> 100	
Disperse Orange 25	Zebra fish	96	LC <sub>50</sub> <sup>1</sup>	268	Brown 1992
	<i>Daphnia magna</i>	48	EC <sub>50</sub> <sup>2</sup>	110	
	<i>Scenedesmus subspicatus</i>	72	EC <sub>50</sub> <sup>2</sup>	54	
	Bacteria	n/a	IC <sub>50</sub> <sup>3</sup>	> 100	
Disperse Yellow 3	Fathead minnow	96	LC <sub>50</sub> <sup>1</sup>	> 180	Little and Lamb 1973

<sup>1</sup> LC<sub>50</sub> – The concentration of a substance that is estimated to be lethal to 50% of the test organisms.

<sup>2</sup> EC<sub>50</sub> – The concentration of a substance that is estimated to have some toxic sublethal effect on 50% of the test organisms.

<sup>3</sup> IC<sub>50</sub> – The concentration of a substance that is estimated to inhibit growth in 50% of the test organisms.

n/a : Not available

In general, due to their poor solubility (< 1 mg/L), disperse dyes are expected to have a low acute ecological impact (Hunger 2003). With the exception of the lone low 96-hour LC<sub>50</sub> value observed for daphnids (Health, Safety, and Human Factors Laboratory 1978), the results of empirical toxicity studies with both assessed substances and several analogues are consistent with this expectation, indicating LC<sub>50</sub> values in the 5 to 340 mg/L range, with *Daphnia* being the most sensitive organisms tested (EC<sub>50</sub>/LC<sub>50</sub> values from 4.5 to 100 mg/L). Although interpretation of results from these tests is complicated by the fact that these effect values are based on nominal concentrations sometimes more than 10 000 times greater than the estimated solubility of the substance (i.e., 0.012 mg/L for Disperse Red 179; 0.004 mg/L for DAPEP; 0.021–0.69 mg/L for analogue CAS RN 68133-69-7), they do represent possible worst-case environmental loadings.

The available empirical ecotoxicity information for analogues of Disperse Red 179 and DAPEP thus indicates that Disperse Red 179 and DAPEP are not likely to be highly hazardous to aquatic organisms.

## B - In Other Environmental Compartments

Since Disperse Red 179 and DAPEP may potentially be discharged to soil from application of sewage sludge to agricultural soils, it would be desirable to obtain toxicity

data for soil organisms. This is relevant because it has been shown that dyes are strongly adsorbed and stick to wastewater treatment plant sludge (Tincher 1988). However, no suitable ecological effects studies were found for these compounds or their analogues in media other than water. Although no suitable ecological effects studies were found for these compounds in soil, considering the toxicity data for aquatic organisms as well as the lack of bioaccumulation potential and their low bioavailability, potential for toxicity to soil-dwelling organisms is likely to be low. For the same reasons, the toxicity potential is also likely to be low for oxic sediment-dwelling species, although this cannot be substantiated due to lack of whole organism sediment toxicity data for these substances or suitable analogues. In addition, the toxicity potential of Disperse Red 179 and DAPEP in anoxic sediments will be low because of the low bioavailability of their anaerobic degradation products.

### **Ecological Exposure Assessment**

No data concerning concentrations of these substances in water in Canada have been identified; therefore, environmental concentrations are estimated from available information, including estimated quantities of the substances in commerce, release rates, and size of receiving water bodies.

### **Industrial releases**

Disperse Red 179 and DAPEP can be used in low volumes at some industrial facilities and can be released to water, where they will stay for an unknown period of time before settling to sediments. Since Disperse Red 179 and DAPEP are analogues, a single exposure scenario was modelled for both substances to determine a predicted environmental concentration (PEC) for the aquatic environment. A number of industrial sites were identified as sources of potential aquatic releases, and one site was selected for evaluation of a worst-case scenario due to the larger quantity of the substances used. Conservative assumptions were made regarding the amount of substance processed and released, the number of processing days, and the sewage treatment plant removal rate. The PEC for Disperse Red 179 and DAPEP was calculated based on a combined use quantity of 510 kg/year (350 kg/year and 160 kg/year for Disperse Red 179 and DAPEP, respectively), of which 22% is assumed to be released over a period of 250 days as a result of the dyeing process when the unfixed dye is washed off of the fibres and discharged with wastewater (Environment Canada 2009a, 2009b, 2009c, 2009d). The 22% released to wastewater (sewer) from industrial activities is a conservative estimate from the Mass Flow Tool (Environment Canada 2009b, 2009c, 2009d). The release amount was then assumed to be discharged directly to a local sewage treatment plant (STP), with a zero removal rate for the substances. Disperse Red 179 and DAPEP in the STP effluent was further assumed to be released to a receiving water body that has a dilution capacity of 10 times the effluent flow. Based on the highest possible release amount estimated and the above-mentioned assumptions, the highest concentration of Disperse Red 179 and DAPEP in the receiving water is  $1.7 \times 10^{-5}$  mg/L (Environment Canada 2009e).

### Consumer releases (Megaflush)

As Disperse Red 179 and DAPEP are found in consumer products and are reported to be released to water (sewers), according to results from the Mass Flow Tool (Environment Canada 2009c, 2009d). Mega Flush (Environment Canada's spreadsheet model for estimating down-the-drain releases from consumer uses) was employed to estimate the potential concentration of the substances in multiple water bodies receiving sewage treatment plant (STP) effluents to which consumer products containing the substances may have been released (Environment Canada 2009f). The spreadsheet model is designed to provide these estimates based on conservative assumptions regarding the amount of substance used and released by consumers. By default, we assume primary and secondary STP removal rates to be 0%, losses from use to be 100%, consumer use of the substance to be over 365 days/year, and the receiving water flow rates at all sites to be the tenth percentile (low end). These estimates are made for approximately 1000 release sites across Canada, which account for most of the major STPs in the country.

The equation and inputs used to calculate the PEC of Disperse Red 179 in the receiving water bodies are described in Environment Canada (2009g). A scenario was run assuming a total consumer quantity of 104 kg/year predicted to be released to sewer (9.2% of total mass), as a result of the laundering of manufactured articles that contain this dye (articles either imported or manufactured in Canada) (Environment Canada 2009c). Using this scenario, the tool estimates that the PEC for Disperse Red 179 in the receiving water bodies ranges from  $1.3 \times 10^{-5}$  to  $1.6 \times 10^{-4}$  mg/L.

A similar scenario for releases from consumer uses was used to predict PECs of DAPEP (Environment Canada 2009h). The scenario was run for DAPEP assuming a total quantity of 47 kg/year (9.2 % of total mass) lost to sewers during the laundering of manufactured articles that contain this dye. Similar removal rates of 0%, were used. Using this scenario, the tool estimates that the PEC for DAPEP in the receiving water bodies ranges from  $5.9 \times 10^{-6}$  to  $7.2 \times 10^{-5}$  mg/L.

### Characterization of Ecological Risk

The approach taken in this ecological screening assessment was to examine a variety of supporting information and develop conclusions based on a weight-of-evidence approach and using precaution as required under CEPA 1999. Lines of evidence considered include results from conservative risk quotient calculations, as well as information on persistence, bioaccumulation, inherent toxicity, sources and fate of the substances.

Disperse Red 179 and DAPEP are expected to be persistent in water, soil and in sediment under aerobic conditions; they are also expected to have a low bioaccumulation potential. The low importation volumes of both substances into Canada indicate a low potential for release into the Canadian environment despite their industrial, commercial and consumer use. Once released into the aquatic environment, they will be found mainly in sediments.

They also are expected to generally demonstrate low to moderate potential for toxicity to aquatic organisms.

Risk quotient analysis integrating conservative estimates of exposure with toxicity information were performed for the aquatic medium to determine whether there is potential for ecological harm in Canada. A predicted no-effect concentration (PNEC) for both substances was conservatively estimated based on the extremely low 96-hour  $LC_{50}$  of 0.12 mg/L (Health, Safety, and Human Factors Laboratory 1978) for daphnids using the analogue substance CAS RN 70198-17-3. This is the lowest experimental analogue value from the acute toxicity data identified, and is an order of magnitude lower than the next lowest value. A factor of 100 was applied to account for extrapolating from acute to chronic (long-term) toxicity and from laboratory results for one species to other potentially sensitive species in the field. The resulting PNEC is 0.0012 mg/L.

When compared to the conservative PECs calculated above for the industrial release scenario, the resulting risk quotient for industrial discharges to the aquatic environment ( $PEC/PNEC$ ) is  $0.000\ 017 / 0.0012 = 0.0145$  for the combined releases of Disperse Red and DAPEP (Environment Canada 2009e). Therefore, it is estimated that concentrations of Disperse Red 179 or DAPEP in surface waters in Canada resulting from industrial discharges for a worst-case scenario site in Canada appear very unlikely to cause adverse effects on populations of aquatic organisms. Given that this industrial release scenario provides a conservative estimate of exposure and risk, the results indicate a low potential for ecological harm resulting from local exposure to point source industrial releases to the aquatic environment.

For exposure resulting from down-the-drain releases using moderately conservative consumer use scenarios, PECs estimated with Mega Flush do not exceed the PNEC at any sites (i.e., all risk quotients much  $< 1$ ) (Environment Canada 2009g, 2009h). This indicates that down-the-drain consumer releases of Disperse Red 179 and DAPEP are not expected to harm aquatic organisms.

Therefore, based on the evidence available, Disperse Red 179 and DAPEP are unlikely to be causing ecological harm in Canada.

### **Uncertainties in Evaluation of Ecological Risk**

Uncertainties in this risk assessment exist due to a lack of data on physical and chemical properties specific to Disperse Red 179 and DAPEP, notably their solubility in water, octanol-water partition coefficient and organic carbon-water partition coefficient. However, read-across approaches, close analogue data, and modelled data using the experimental value adjustment method of EPIsuite (2008) were used to fill critical data gaps within an acceptable margin of error.

The persistence assessment is limited by the uncertainty about the rate of degradation in anaerobic sediments and the extent to which degradation occurs in these sediments and whether the degradation products (e.g., amines) would be biologically available. Nevertheless, it is clear that anaerobic degradation of the bioavailable portion of azo dyes



in sediments to constitutive amines is much faster (half-lives in the order of days) than aerobic biodegradation. Although the amine degradation products are not expected to be biologically available because they form only in relatively deep anoxic sediment and can be tightly bound to sediment through nucleophilic addition and oxidative radical coupling (Weber et al. 2001; Colon et al. 2002), this issue is a source of uncertainty in the toxicity assessment of Disperse Red 179 and DAPEP.

Uncertainties are also associated with the fraction of the substances that is assumed to be released during use (i.e., during industrial activities and use of consumer products) and the lack of information on environmental concentrations in Canada of Disperse Red 179 and DAPEP. These uncertainties were addressed by making conservative assumptions in each of the modelling exercises.

The experimental concentrations associated with toxicity to aquatic organisms have an additional source of uncertainty in that these concentrations exceed the solubility of the chemical in water. Despite this, the low solubility of the substances and their limited bioavailability due to their molecular size suggest that Disperse Red 179 and DAPEP are not highly hazardous to aquatic organisms.

Also, regarding ecotoxicity, based on the predicted partitioning behaviour of these chemicals, the significance of soil and sediment as important media of exposure is not well addressed by the effects data available. Indeed, the only effects data identified apply primarily to pelagic aquatic exposures, although the water column may not be the medium of primary long-term concern. Nevertheless, based on the relatively low aquatic toxicity of these substances and the low masses in commerce, potential for harm to soil- and sediment-dwelling organisms is also expected to be low.

## Potential to Cause Harm to Human Health

### Exposure Assessment

No environmental measurements of Disperse Red 179 or DAPEP were identified in the literature. Based on the release information, concentrations in environmental media are expected to be negligible.

Disperse dyes such as Disperse Red 179 and DAPEP are used in the textile industry to colour synthetic fabrics such as polyesters and polyamides. Disperse dyes derive their name from the dyeing process employed (Danish EPA 1998). Because of their low water solubility, the dye compounds are typically milled to produce a fine powder and applied as a dispersion in water. The hydrophobic dye molecules adsorb to the hydrophobic textile, and heating induces uptake of the dye by the textile (Chudgar and Oakes 2003). Disperse Red 179 and DAPEP do not form chemical bonds with the textile; therefore, migration is possible. Disperse Red 179 and DAPEP may be used as dyeing agents for synthetic fibres for personal apparel and domestic textile uses.

The upper-bounding exposure estimates were derived for two scenarios. One scenario considered the dermal route, when an individual wears apparel made of a fabric dyed with these substances; the other was for the oral route, for mouthing of a fabric dyed with these substances by infants and young children. These are considered the most likely routes of exposure. The upper-bounding internal doses from dermal exposure to Disperse Red 179 and DAPEP were estimated to range from 0.1 to 4 µg/kg body weight (kg-bw) per day for all age groups wearing new, unwashed apparel possessing good to poor colourfastness properties (ETAD 2004). For infants and children, the estimated exposure via mouthing was less than 0.002 µg/kg-bw per day. A recent study found that the amount of a disperse dye that migrated onto the skin of human volunteers was 300–600 times lower than that leached by sweat simulants (Meinke et al. 2009). This supports the conservative nature of the upper-bound exposure estimates. In addition, the dyes in textiles are expected to be leached out of fabric primarily by laundering, so any potential exposures would decline over time. Details of the assumptions used in these calculations are given in Appendix 3 and Appendix 4.

### Health Effects Assessment

#### *Disperse Red 179 (CAS RN 16586-42-8)*

A single study testing Disperse Red 179 (cited as Disperse Violet 52) in an *in vitro* indicator genotoxicity assay (SOS/umu test) reported positive results with S9 activation and negative results without S9 (Kosaka and Nakamura 1990). The outputs of predictive QSAR models for Disperse Red 179 were mixed for carcinogenicity and genotoxicity endpoints (CASETOX 2008, DEREK 2008, Leadscope 2009 and Toxtree 2009) (Appendix 5). As only limited data were available with respect to the potential toxicity of

Disperse Red 179, relevant information on potential metabolites and analogues of this substance was also considered.

Since Disperse Red 179 is a member of the family of azo substances, relevant health effects information on its potential azo cleavage products was also considered. It has been demonstrated that certain azo substances can undergo reductive cleavage mediated by azoreductase enzymes found in mammalian tissues as well as bacteria of the intestine and skin (Platzek et al. 1999; Golka et al. 2004; Chen 2006; Xu et al. 2007; Stingley et al. 2010). While it is recognized that the degree of azo reduction is likely influenced by various factors (e.g., solubility of parent, presence and position of molecular substituents), in the absence of chemical-specific data, it is assumed that exposure to an azo substance may also lead to exposure to its corresponding azo cleavage products, typically aromatic amines. Accordingly, the predicted azo cleavage products for Disperse Red 179; namely, 6-nitro-2-aminobenzothiazole (CAS RN 6285-57-0) and propanenitrile, 3[(4-amino-3-methylphenyl)ethylamino]- (CAS RN 105294-34-6) (Appendix 6) are also considered in this screening assessment.

The predicted metabolite CAS RN 6285-57-0 was tested for *in vitro* mutagenicity in several studies. Three separate studies showed positive results in *Salmonella typhimurium* strains TA98, TA100 and TA1538 with and without S9 activation, while strains TA1535 and TA1537 were positive only without S9 (NTP 1982, 1983; Seifried et al. 2006). No mutagenicity was observed in mouse lymphoma cells with or without S9 activation for this substance (Seifried et al. 2006). CAS RN 6285-57-0 has also been linked to potential carcinogenic activity based on similarity to carcinogenic aromatic nitro compounds (Helmes et al. 1982). No empirical toxicity data were available for CAS RN 105294-34-6, the other predicted azo cleavage product of Disperse Red 179. Results of QSAR modelling for carcinogenicity and genotoxicity endpoints were mixed for both of these potential azo cleavage products (Appendix 5).

The only suitable analogue identified, Disperse Red 145 (CAS RN 25510-81-0) (Appendix 6), produced a positive result in the SOS/umu test with and without liver S9 activation (Kosaka and Nakamura 1990). It is also noted that the class of aminobenzothiazole azo dyes has been identified by the US Environmental Protection Agency (EPA) as a Toxic Substances Control Act (TSCA) New Chemicals Program (NCP) chemical category due to hazard concerns for related substances and potential metabolites (US EPA 2002).

The information obtained on Disperse Red 179, its potential azo cleavage products and a structural analogue (Disperse Red 145) suggests that there may be potential hazard associated with this Disperse Red 179. However, the confidence in the toxicity database is low due to the limited information available for this substance.

**DAPEP (25176-89-0)**

No empirical toxicity data were found for DAPEP. However, since DAPEP is one of two isomers that comprise Disperse Red 153 (CAS RN 78564-87-1),<sup>1</sup> data on this substance are considered to be representative of DAPEP. A single study showed Disperse Red 153 to be negative in the SOS/umu test with and without S9 activation (Kosaka and Nakamura 1990). In addition, the outputs of predictive QSAR models on both isomers of Disperse Red 153 were mixed for carcinogenicity and genotoxicity endpoints (Appendix 7). As only limited data were available with respect to the potential toxicity of DAPEP and Disperse Red 153, relevant information on potential metabolites and analogues was also considered.

Since DAPEP is a member of the family of azo substances, the potential azo cleavage products of DAPEP; namely, 5,6'-dichloro-2-aminobenzothiazole (CAS RN 24072-75-1) and propanenitrile, 3-[(4-aminophenyl)ethylamino]- (CAS RN 100894-10-8) (Appendix 8), are considered in this screening assessment (see previous section for rationale).

The predicted metabolite 5,6-dichloro-2-benzothiazoleamine (CAS RN 24072-75-1) was tested for mutagenicity both *in vivo* and *in vitro*. This substance was negative for mutagenicity both in *Salmonella typhimurium* (NTP 1986a, Seifried et al. 2006) and in mouse lymphoma cells (Seifried et al. 2006). The substance did not induce micronuclei in male or female mice exposed orally via the feed as part of an unpublished subchronic study (NTP 1993; Witt et al. 2000). While the full data set for this subchronic study are unpublished and not readily available (NTP 1986b), a brief report from the US National Toxicology Program indicated that some histopathological effects in the liver and kidney were observed in both rats and mice; however, incidence and doses were not provided (NTP 2000). It should also be noted that the US EPA identified the class of aminobenzothiazole azo dyes in part due to concerns for neurotoxicity of “chlorinated 2-aminobenzothiazole” as a reductive cleavage product (US EPA 2002). No empirical toxicity data were identified for the other potential metabolites CAS RN 25150-27-0 or CAS RN 100894-10-8 (the predicted azo cleavage product of the other dichloro isomer found in Disperse Red 153, see Appendix 8 for structure). The QSAR modelling results on carcinogenicity and genotoxicity were mixed for all of the predicted azo cleavage metabolites for DAPEP and the 6,7'-dichloro isomer of Disperse Red 153 (Appendix 7).

Besides Disperse Red 153 (CAS RN 78564-87-1 already discussed above (a mixture of isomers including DAPEP), the only other potential analogue with some hazard data identified was Disperse Red 152 (CAS RN 78564-86-0). Disperse Red 152 is also a mixture 5,6'- and 6,7'-dichloro isomers (see Appendix 8 for structure), similar to Disperse Red 153 and yielded a similar negative result in the SOS/umu test (Kosaka and Nakamura 1990). Although limited analogues were identified for DAPEP, it should also be noted that the class of aminobenzothiazole azo dyes to which DAPEP belongs has

<sup>1</sup> Disperse Red 153 (CAS RN 78564-87-1) is composed of two isomers with dichloro substitution on the benzothiazole either at the 5,6'- position (CAS RN 25176-89-0, DAPEP) or at the 6,7'- position (no CAS RN). See Substance Identity section for description and Appendix 8 for structures of both DAPEP and the 6,7'-dichloro isomer.

been identified by the US EPA as a TSCA NCP chemical category due to hazard concerns for related substances and potential metabolites (US EPA 2002).

The limited toxicological information obtained for DAPEP, a structural isomer, potential azo cleavage products, and an analogue (Disperse Red 152 suggests that there may be a potential hazard associated with DAPEP. However, the confidence in the toxicity database is low due to the limited information available for this substance.

### **Characterization of Risk to Human Health**

Although only limited empirical data were identified for Disperse Red 179 and DAPEP, their potential azo cleavage products, and analogues, the available information together with mixed QSAR results suggest a potential hazard. However, the limited health effects information available precludes selection of critical effect levels for use in risk characterization of these substances.

Disperse Red 179 and DAPEP belong to the group of azo substances that may potentially release aromatic amines by reductive cleavage of the azo linkage. Although hazard data is limited for the specific aromatic amines associated with Disperse Red 179 and DAPEP, the collective toxicity database for aromatic amines as a chemical class (Vineis and Pirastu 1997; Benigni and Passerini 2002; Talaska 2003) suggests that there may be a potential concern for these substances.

The potential for exposure of the general population to Disperse Red 179 and DAPEP from environmental media is expected to be negligible. Exposure of the general population of Canada to Disperse Red 179 and DAPEP from wearing personal apparel by all age groups and from incidental mouthing of fabrics by children has been quantified and is low.

As exposure of the general population in Canada based on the use of the substance as a synthetic textile dye is expected to be low, the expected risk to human health from the potential hazards is considered to be low.

**Uncertainties in Evaluation of Risk to Human Health**

There are uncertainties associated with the exposure assessment. Information was not available on migration factors for and solubilities of Disperse Red 179 and DAPEP. Sources of exposure have been broadly characterized as synthetic fabrics, as no specific consumer products were identified. However, confidence is high that the exposure estimates are conservative for the following reasons. The migration factor used in the assessment corresponds to daily exposure to new, unwashed fabrics with poor colourfastness, while leaching is expected to occur primarily during laundering. Additionally, a study identified that the amount of a disperse dye that migrated onto human volunteers was substantially lower than that leaching into solution.

Confidence in the toxicological database for these substances is considered to be low. There is limited hazard data available for Disperse Red 179, DAPEP, their analogues, and potential azo cleavage products. There is also a lack of information on potential for these substances to undergo azo cleavage, a major consideration when evaluating the toxicity of azo substances.

## **Conclusion**

Based on the information presented in this screening assessment, it is concluded that neither Disperse Red 179 nor DAPEP is entering the environment in a quantity or concentration or under conditions that have or may have an immediate or long-term harmful effect on the environment or its biological diversity or that constitute or may constitute a danger to the environment on which life depends.

Although limited empirical and modelling data for the substances, their potential metabolites and analogues provide an indication of a potential hazard, based upon consideration of the limited health effects information and low to negligible exposure of the general population to Disperse Red 179 and DAPEP, it is concluded that Disperse Red 179 and DAPEP are not entering the environment in a quantity or concentration or under conditions that constitute or may constitute a danger in Canada to human life or health.

It is therefore concluded that Disperse Red 179 and DAPEP do not meet the criteria set out in section 64 of CEPA 1999. Additionally, Disperse Red 179 and DAPEP meet the criteria for persistence; however, but do not meet the criteria for bioaccumulation as set out in the Persistence and Bioaccumulation Regulations (Canada 2000).

Because these substances are listed on the Domestic Substances List, their import and manufacture in Canada are not subject to notification under subsection 81(1). Given the potential hazardous properties of these substances, there is concern that new activities that have not been identified or assessed could lead to this substance meeting the criteria set out in section 64 of the Act. Therefore, it is recommended to amend the Domestic Substances List, under subsection 87(3) of the Act, to indicate that subsection 81(3) of the Act applies with respect to these substances so that new manufacture, import or use of this substance is notified and undergoes ecological and human health risk assessments.

In addition and where relevant, research and monitoring will support verification of assumptions used during the screening assessment.

## **Considerations for Follow-up**

Disperse Red 179 and DAPEP belong to a group of azo substances that may metabolize to aromatic amines, which as a chemical class are known to exhibit hazardous properties, including carcinogenicity. Therefore, additional activity (e.g., research, monitoring and surveillance, assessment) to characterize the risk to human health in Canada of this broader group of azo substances may be undertaken.

## References

- ACD/pK<sub>a</sub> DB [Prediction module]. 2005. Version 9.04. Toronto (ON): Advanced Chemistry Development. Available from: [http://www.acdlabs.com/products/phys\\_chem\\_lab/pka/](http://www.acdlabs.com/products/phys_chem_lab/pka/)
- Anliker R, Moser P. 1987. The limits of bioaccumulation of organic pigments in fish: their relation to the partition coefficient and the solubility in water and octanol. *Ecotoxicol Environ Safety* 13:43-52.
- Anliker R, Clarke EA, Moser P. 1981. Use of the partition coefficient as an indicator of bioaccumulation tendency of dyestuffs in fish. *Chemosphere* 10(3):263-274.
- Anliker R, Moser P, Poppinger D. 1988. Bioaccumulation of dyestuffs and organic pigments in fish. Relationships to hydrophobicity and steric factors. *Chemosphere* 17(8):1631-1644.
- Aronson D, Howard PH. 1999. Evaluating potential POP/PBT compounds for environmental persistence. North Syracuse (NY): Syracuse Research Corp., Environmental Science Centre. Report No. SRC-TR-99-020.
- Aronson D, Boethling B, Howard P, Stiteler W. 2006. Estimating biodegradation half-lives for use in chemical screening. *Chemosphere* 63:1953-1960.
- Baughman GL, Perenich TA. 1988. Fate of dyes in aquatic systems: I. Solubility and partitioning of some hydrophobic dyes and related compounds. *Environmental Toxicology and Chemistry* 7(3):183-199.
- Baughman GL, Weber EJ. 1994. Transformation of dyes and related compounds in anoxic sediment: kinetics and products. *Environmental Science and Technology* 28(2): 267-276.
- [BBM] Baseline Bioaccumulation Model. 2008. Gatineau (QC): Environment Canada, Existing Substances Division. [Model based on Dimitrov et al. 2005]. [cited 2008-11-21]. Available upon request.
- Benigni R, Passerini L. 2002. Carcinogenicity of the aromatic amines: from structure-activity relationships to mechanisms of action and risk assessment. *Mutation Research* 511(3):191-206.
- [BIOWIN] Biodegradation Probability Program for Windows [Estimation model]. 2000. Version 4.02. Washington (DC): U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics; Syracuse (NY): Syracuse Research Corporation. Available from: <http://www.epa.gov/oppt/exposure/pubs/episuite.htm>
- BMG. 2000a. C.I. Disperse Red 153, Test for inhibition of oxygen consumption by activated sludges: Respiration Inhibition Test. BMG report no. 800/b-00, December 2000.
- BMG. 2000b. Foron Rubin RD-S Presskuchen trocken, 96-hour acute toxicity to *Poecilia reticulata* (Guppy): Limit Test (100 mg/L). BMG report no. 800/a-00, December 2000.
- BMG. 2001. C.I. Disperse Red 153, Inherent biodegradability – Evaluation of the aerobic biodegradability in an aqueous medium: Zahn-Wellens / EMPA test. BMG report no. 800/c-00, January 2001.
- BMG. 2003a. Foron Rubin RD-S Presskuchen trocken, Inherent biodegradability – Evaluation of the aerobic biodegradability in an aqueous medium: Zahn-Wellens / EMPA test. BMG report no. 709/c-03, October 2003.
- BMG. 2003b. Foron Rubin RD-S Presskuchen trocken, Test for inhibition of oxygen consumption by activated sludges: Respiration Inhibition Test. BMG report no. 709/a-03, October 2003.



- BMG. 2003c. Foron Rubin RD-S Presskuchen trocken, 96-hour acute toxicity to *Poecilia reticulata* (Guppy): Screening Test. BMG report no. 709/b-03, October 2003.
- Boethling RS, Howard PH, Beauman JA, Larosche ME. 1995. Factors for intermedia extrapolations in biodegradability assessment. *Chemosphere* 30(4):741-752.
- Brown D. 1987. Effects of colorants in the aquatic environment. *Ecotoxicol Environ Safety* 13:1391-1347.
- Brown D (ICI Group Environmental Laboratory, Brixham, U.K.). 1992. Environmental assessment of dyestuffs. Prepared for Ecological and Toxicological Association of the Dyes and Organic Pigments Manufacturers, Basel, Switzerland. ETAD ecological sub-committee project E3020. Submitted to Environment Canada.
- [Canada]. 1999. *Canadian Environmental Protection Act, 1999*. S.C., 1999, c. 33. Canada Gazette. Part III, vol. 22, no. 3. Ottawa: Queen's Printer. Available from: <http://www.canlii.org/ca/sta/c-15.31/whole.html>
- [Canada]. 2000. *Canadian Environmental Protection Act, 1999: Persistence and Bioaccumulation Regulations*, P.C. 2000-348, 23 March 2000, SOR/2000-107. Canada Gazette. Part II, vol. 134, no. 7, p. 607-612. Ottawa: Queen's Printer. Available from: <http://www.gazette.gc.ca/archives/p2/2000/2000-03-29/pdf/g2-13407.pdf>
- Canada, Dept. of the Environment, Dept. of Health. 2006a. *Canadian Environmental Protection Act, 1999: Notice of intent to develop and implement measures to assess and manage the risks posed by certain substances to the health of Canadians and their environment*. Canada Gazette, Part I, vol. 140, no. 49, p. 4109-4117. Ottawa: Queen's Printer. Available from: <http://www.gazette.gc.ca/archives/p1/2006/2006-12-09/pdf/g1-14049.pdf>
- Canada, Dept. of the Environment, Dept. of Health. 2006b. *Canadian Environmental Protection Act, 1999: Notice with respect to selected substances identified as priority for action*. Canada Gazette, Part I, vol. 140, no. 9, p. 435-459. Ottawa: Queen's Printer. Available from: <http://www.gazette.gc.ca/archives/p1/2006/2006-03-04/pdf/g1-14009.pdf>
- Canada, Dept. of the Environment, Dept. of Health. 2008. *Canadian Environmental Protection Act, 1999: Notice of first release of technical information relevant to substances identified in the Challenge*. Canada Gazette, Part I, vol. 142, no. 35, p. 2497-2501. Ottawa: Queen's Printer. Available from: <http://www.gazette.gc.ca/rp-pr/p1/2008/2008-08-30/pdf/g1-14235.pdf>
- CASETOX [Prediction module]. 2008. Version 2.0. Beachwood (OH): MultiCASE. [cited 2009 Sep 30]. Available from: <http://www.multicase.com/products/prod03.htm> [restricted access].
- Chen H. 2006. Recent advances in azo dye degrading enzyme research. *Curr Protein Pept Sci* 7: 101-111.
- [CATABOL] Probabilistic assessment of biodegradability and metabolic pathways [Computer Model]. c2004-2008. Version 5.10.2. Bourgas (BG): Bourgas Prof. Assen Zlatarov University, Laboratory of Mathematical Chemistry. Available from: <http://oasis-lmc.org/?section=software&swid=1>
- [ChemID] ChemID Plus Advanced. [Database on the Internet]. 2009. [accessed 2009 April 20] U.S. National Library of Medicine (MD), National Institute of Health. Available from: <http://chem.sis.nlm.nih.gov/chemidplus/>
- Chen H. 2006. Recent advances in azo dye degrading enzyme research. *Current Protein & Peptide Science* 7: 101-111.
- Choi JH, Kim MH, Park JS, Jeon JM, Kim DO, Towns AD. 2007. Coloration of poly(lactic acid) with disperse dyes. II. Dyeing characteristics and color fastness. *Fibers and Polymers* 8(1):37-42.

Chudgar RJ, Oakes J. 2003. Dyes, azo. In: Kirk-Othmer encyclopedia of chemical technology, online version. Available from:  
<http://mrw.interscience.wiley.com/emrw/9780471238966/kirk/article/azochud.a01/current/abstract?hd=All,azoandhd=All,dye> [restricted access]

[CII] Color Index International [database on the Internet]. 2002 – . 4th ed. Research Triangle Park (NC): American Association of Textile Chemists and Colorists. [cited 2009 March 25] Available from:  
<http://www.colour-index.org/>

Clariant. 1996. IUCLID dataset for C.I. Disperse Blue 79 (CAS No 12239-34-8)

Colon D, Weber E, Baughman G. 2002. Sediment-associated reactions of aromatic amines. 2. QSAR Development. Environmental Science and Technology 36 (12):2443-2450.

[ConsExpo] Consumer Exposure Model [Internet]. 2006. Version 4.1. Bilthoven (NL): Rijksinstituut voor Volksgezondheid en Milieu (National Institute for Public Health and the Environment). [Available from:  
<http://www.rivm.nl/en/healthanddisease/productsafety/ConsExpo.jsp#tcm:13-42840>]

[CPOPs] Canadian POPs Model. 2008. Gatineau (QC): Environment Canada, Existing Substances Division; Bourgas (BG): Bourgas Prof. Assen Zlatarov University, Laboratory of Mathematical Chemistry. [Model developed based on Mekenyan et al. 2005]. Available upon request.

[Danish EPA] Danish Environmental Protection Agency. 1998. Azocolorants in Textiles and Toys, Environmental and Health Assessment. Environmental Project, no. 416 1998. [Available from:  
[http://www2.mst.dk/common/Udgivramme/Frame.asp?http://www2.mst.dk/udgiv/Publications/1998/87-7909-136-9/html/helepubl\\_eng.htm](http://www2.mst.dk/common/Udgivramme/Frame.asp?http://www2.mst.dk/udgiv/Publications/1998/87-7909-136-9/html/helepubl_eng.htm)]

de Bruijn J, Busser F, Seinen W, and Hermens J. 1989. Determination of octanol/water partition coefficients for hydrophobic organic chemicals with the “slow-stirring” method. Environ Toxicol Chem 8:499-512.

[DEREK] Deductive Estimation of Risk from Existing Knowledge [Prediction module on CD ROM]. 2008. Version 10.0.2. Cambridge (MA): Harvard University, LHASA Group. [cited 2009 Sep 30]. Available from: [http://www.lhasalimited.org/index.php?cat=2&sub\\_cat=2#](http://www.lhasalimited.org/index.php?cat=2&sub_cat=2#) [restricted access].

Dimitrov SD, Dimitrova NC, Walker JD, Veith GD, Mekenyan OG. 2002. Predicting bioconcentration factors of highly hydrophobic chemicals. Effects of molecular size. Pure and Applied Chemistry 74(10):1823-1830.

Dimitrov S, Dimitrova N, Parkerton T, Comber M, Bonnell M, Mekenyan O. 2005. Base-line model for identifying the bioaccumulation potential of chemicals. SAR and QSAR in Environmental Research 16(6):531-554.

[Environ] ENVIRON International Corporation. 2003a. Voluntary Children’s Chemical Evaluation Program Pilot (VCCEPP)–Tier 1 assessment of the potential health risks to children associated with exposure to the commercial pentabromodiphenyl ether product and appendices [Internet]. Emerville (CA): ENVIRON International Corporation. [Available from:  
<http://www.epa.gov/oppt/vccep/pubs/chem22a.html>]

[Environ] ENVIRON International Corporation. 2003b. Voluntary Children’s Chemical Evaluation Program Pilot (VCCEPP)–Tier 1 assessment of the potential health risks to children associated with exposure to the commercial octabromodiphenyl ether product and appendices [Internet]. Emerville (CA): ENVIRON International Corporation. [Available from: <http://www.epa.gov/oppt/vccep/pubs/chem23a.h>]

Environment Canada. 1994. .Acute Fish Toxicity Test Submission in Fulfillment of New Substances Notification Regulations to New Substances Branch, Environment Canada under New Substance Notification Program.

Environment Canada. Chemicals Evaluation Division. 2000. Environmental Categorization for Persistence, Bioaccumulation and Inherent Toxicity of Substances on the Domestic Substances List Using QSARs. Final Report. Environment Canada. July.

Environment Canada. 2006. Data for selected substances collected under the Canadian Environmental Protection Act, 1999, Section 71 Notice with respect to selected substances identified as priority for action. Data prepared by: Environment Canada, Health Canada, Existing Substances Program.

Environment Canada. 2009a. Data for Batch 7 substances collected under Canadian Environmental Protection Act, 1999, Section 71 Notice with respect to Batch 7 Challenge substances. Data prepared by: Environment Canada, Existing Substances Program.

Environment Canada. 2009b. Guidance for conducting ecological assessments under CEPA, 1999: science resource technical series, technical guidance module: Mass Flow Tool. Preliminary draft working document. Gatineau (QC): Environment Canada, Existing Substances Division.

Environment Canada. 2009c. Assumptions, limitations and uncertainties of the mass flow tool for Disperse Red 179, CAS RN 16586-42-8. Internal draft document. Gatineau (QC): Environment Canada, Existing Substances Division. Available on request.

Environment Canada. 2009d. Assumptions, limitations and uncertainties of the mass flow tool for DAPEP, CAS RN 25176-89-0. Internal draft document. Gatineau (QC): Environment Canada, Existing Substances Division. Available on request.

Environment Canada. 2009e. Site Specific Analysis report: CAS RN 16586-42-8 & 25176-89-0, Unpublished report. Gatineau (QC): Environment Canada, Ecological Assessment Division.

Environment Canada. 2009f. Guidance for conducting ecological assessments under CEPA, 1999: science resource technical series, technical guidance module: Mega Flush consumer release scenario. Preliminary draft working document. Gatineau (QC): Environment Canada, Existing Substances Division.

Environment Canada. 2009g. Mega Flush report: CAS RN 16586-42-8, 2009-04-28. Unpublished report. Gatineau (QC): Environment Canada, Existing Substances Division.

Environment Canada. 2009h. Mega Flush report: CAS RN 25176-89-0, 2009-04-28. Unpublished report. Gatineau (QC): Environment Canada, Existing Substances Division.

[EPISuite] Estimation Programs Interface Suite for Microsoft Windows [Estimation model]. 2008. Version 4.0. Washington (DC): U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics; Syracuse (NY): Syracuse Research Corporation. Available from: <http://www.epa.gov/oppt/exposure/pubs/episuite.htm>

[ETAD] Ecological and Toxicological Association of Dyes and Organic Pigments Manufacturers. 1983. Extractability of dyestuffs from textiles. Basel (CH): Ecological and Toxicological Association of Dyes and Organic Pigments Manufacturers. ETAD Project A 4007.

[ETAD] Ecological and Toxicological Association of Dyes and Organic Pigments Manufacturers. 1992. Draft Guidelines for the Assessment of Environmental Exposure to Dyestuffs.

[ETAD] Ecological and Toxicological Association of Dyes and Organic Pigments Canadian Affiliates, Dayan J, Trebitz H, consultants. 1995. Health and environmental information on dyes used in Canada. Unpublished report submitted to Environment Canada, New Substances Division. On the cover: An

overview to assist in the implementation of the New Substances Notification Regulations under the Canadian Environmental Protection Act.

[ETAD 2004] The Ecological and Toxicological Association of Dyes and Organic Pigments Manufacturers. 2004. Setting a risk based detection limit of sensitizing disperse dyes on textiles. [Available from: [www.etad.com/documents/Downloads/publications/detectionlimit.pdf](http://www.etad.com/documents/Downloads/publications/detectionlimit.pdf)]

[ETAD] Ecological and Toxicological Association of Dyes and Organic Pigments Manufacturers. 2005. ETAD data for DSL categorization and screening submitted to Environment Canada on October 27, 2005. Study report by Intertek ASG. File Ref 2005/CC0157-001/REGIS.

Golka K, Kopps S., Myslak ZW. 2004. Carcinogenicity of azo colorants: influence of solubility and bioavailability. *Toxicology Letters*. 151:203–210

Health, Safety, and Human Factors Laboratory. 1978. Basic Toxicity of 2-[4-[(6-chloro-2-benzothiazolylazo)phenyl]-ethylamino]ethyl acetate. Toxicology Section, Health, Safety, and Human Factors Laboratory. Report submitted by Eastman Kodak Company in 1992 to the U.S. Environmental Protection Agency Registration and Agreement for the TSCA 8(a) Compliance Audit Program.

Health Canada. 1998. Exposure factors for assessing total daily intake of priority substances by the general population of Canada. Unpublished report. Ottawa (ON): Health Canada. Environmental Health Directorate.

Helmes CT, Fung VA, Lewin B, McCaleb KE, Malko S, Pawlovich AM. 1982. A study of aromatic nitro compounds for the selection of candidates for carcinogen bioassay. *Journal of Environmental Science and Health, Part A: Environmental Science and Engineering & Toxic and Hazardous Substance Control* 17: 75–128.

Hunger K, ed. 2003. *Industrial dyes; chemistry, properties, applications*. Weinheim (DE): WILEY-VCH Verlag GmbH & Co. KGaA.

Kosaka H & Nakamura S. 1990. Genotoxicity of synthetic dyes in umu test using *Salmonella typhimurium* TA1535/pSK1002 (1). *Jpn. J. Ind. Health*. 32:89-104. (also cited in Reifferscheid & Heil 1996)

[KOWWIN] Octanol-Water Partition Coefficient Program for Microsoft Windows [Estimation model]. 2000. Version 1.67. Washington (DC): U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics; Syracuse (NY): Syracuse Research Corporation. Available from: <http://www.epa.gov/oppt/exposure/pubs/episuite.htm>

Kraetke RM, Platzek T. 2005. Exposure to chemicals in clothing textiles: methods and models [Internet]. Abstract for Poster 74 at the conference “Occupational and environmental exposure of skin to chemicals—2005.” Washington (DC): National Institute for Occupational Safety and Health. Available from: <http://www.cdc.gov/niosh/topics/skin/OEESC2/AbPost074Kraetke.html>

[Leadscope] Leadscope Model Applier [Prediction module]. 2009. Version 1.2.0-3. Columbus (OH): Leadscope, Inc. [cited 2009 Sep 30]. Available from: [http://www.leadscope.com/all\\_products.php](http://www.leadscope.com/all_products.php) [restricted access].

Little LW, Lamb JC III. 1973. Acute toxicity of 46 selected dyes to the fathead minnow, *Pimephales promelas*. *Dyes and the Environment: Reports on Selected Dyes and Their Effects*. American Dye Manufacturers Institute, Inc. 1:130.

Maradiya HR. 2004. Disperse dyes based on 2-aminoheterocycles. *J Saudi Chem Soc*. 8(3):495-504.

Meinke M, Abdollahnia M, Gähr, Platzek T, Sterry W and Lademann J. 2009. Migration and penetration of a fluorescent textile dye into the skin – *in vivo* versus *in vitro* methods. *Experimental Dermatology* 18(9): 789-792.

Mekenyan G, Dimitrov SD, Pavlov TS, Veith GD. 2005. POPs: a QSAR system for creating PBT profiles of chemicals and their metabolites. *SAR QSAR Environ Res* 16(1-2):103-133.

[MITI] Ministry of International Trade & Industry (Jpn), Basic Industries Bureau, Chemical Products Safety Division. 1992. Biodegradation and bioaccumulation data of existing chemicals based on the CSCL Japan. Tokyo (Jpn): Japan Chemical Industry Ecology-Toxicology & Information Centre.

Mortelmans K, Haworth S, Lawlor T, Speck W, Tainer B, Zeiger E. 1986. *Salmonella* mutagenicity tests: II. Results from the testing of 270 chemicals. *Environmental Mutagenesis* 8: 1-119

Nakagawa M, Kawai K, Kawai K. 1996. Multiple azo disperse dye sensitization mainly due to group sensitizations to azo dyes. *Contact Dermatitis* 34:6-11.

[NCI] National Chemical Inventories [database on CD-ROM]. 2009. Columbus (OH): American Chemical Society. [cited 2009 March 11] Available from: <http://www.cas.org/products/cd/nci/index.html>

Noriris B and Smith S. 2002. Research into the mouthing behaviour of children up to 5 years old. Report commissioned by UK Department of Trade and Industry, London, UK.

[NTP] National Toxicology Program. 1982. Study ID 70250: Test result for an *in vitro* genetic toxicology test (mutagenicity in *Salmonella*) for 2-amino-6-nitrobenzothiazole (6285-57-0). National Institutes of Health's National Institute of Environmental Health Sciences (NIEHS). Research Triangle Park, NC. [Accessed from the NTP Database on Nov. 23, 2009]. Available from: [http://ntp-apps.niehs.nih.gov/ntp\\_tox/index.cfm?fuseaction=salmonella.salmonellaData&study\\_no=702501&cas\\_no=6285%2D57%2D0&endpointlist=SA](http://ntp-apps.niehs.nih.gov/ntp_tox/index.cfm?fuseaction=salmonella.salmonellaData&study_no=702501&cas_no=6285%2D57%2D0&endpointlist=SA) (also cited in Mortelmans et al. 1986)

[NTP] National Toxicology Program. 1983. Study ID 832791: Test result for an *in vitro* genetic toxicology test (mutagenicity in *Salmonella*) for 2-amino-6-nitrobenzothiazole (6285-57-0). National Institutes of Health's National Institute of Environmental Health Sciences (NIEHS). Research Triangle Park, NC. [Accessed from the NTP Database on Nov. 23, 2009]. Available from: [http://ntp-apps.niehs.nih.gov/ntp\\_tox/index.cfm?fuseaction=salmonella.salmonellaData&study\\_no=832791&cas\\_no=6285%2D57%2D0&endpointlist=SA](http://ntp-apps.niehs.nih.gov/ntp_tox/index.cfm?fuseaction=salmonella.salmonellaData&study_no=832791&cas_no=6285%2D57%2D0&endpointlist=SA) (also cited in Mortelmans et al. 1986)

[NTP] National Toxicology Program. 1986a. Study ID 461292: Test result for an *in vitro* genetic toxicology test (mutagenicity in *Salmonella*) for 5,6'-dichloro-2-benzothiazolamine (24072-75-1). National Institutes of Health's National Institute of Environmental Health Sciences (NIEHS). Research Triangle Park, NC. [Accessed from the NTP Database on Nov. 23, 2009]. Available from: [http://ntp-apps.niehs.nih.gov/ntp\\_tox/index.cfm?fuseaction=salmonella.overallresults&cas\\_no=24072-75-1&endpointlist=SA](http://ntp-apps.niehs.nih.gov/ntp_tox/index.cfm?fuseaction=salmonella.overallresults&cas_no=24072-75-1&endpointlist=SA) (also cited in Mortelmans et al. 1986)

[NTP] National Toxicology Program. 1986b. Study ID C61712: Short-term toxicity studies (2-week, 13-week, dosed-feed) in rats and mice for 5,6'-dichloro-2-benzothiazolamine (24072-75-1). (no Toxicity Report published). Available from: [http://ntp-apps.niehs.nih.gov/ntp\\_tox/index.cfm?fuseaction=shorttermbioassaydata.datasearch&chemical\\_name=5%2C6-Dichloro-2-benzothiazolamine&cas\\_no=24072-75-1&study\\_no=C61712&study\\_length=13%20Weeks](http://ntp-apps.niehs.nih.gov/ntp_tox/index.cfm?fuseaction=shorttermbioassaydata.datasearch&chemical_name=5%2C6-Dichloro-2-benzothiazolamine&cas_no=24072-75-1&study_no=C61712&study_length=13%20Weeks)

[NTP] National Toxicology Program. 1993. Study ID A42046: Test result for *in vivo* micronucleus induction in male and female mice for 5,6'-dichloro-2-benzothiazolamine (24072-75-1). National Institutes of Health's National Institute of Environmental Health Sciences (NIEHS). Research Triangle Park, NC. [Accessed from the NTP Database on Nov. 23, 2009]. Available from: [http://ntp-apps.niehs.nih.gov/ntp\\_tox/index.cfm?fuseaction=micronucleus.choosestudytype&cas\\_no=24072-75-1&endpointlist=MN](http://ntp-apps.niehs.nih.gov/ntp_tox/index.cfm?fuseaction=micronucleus.choosestudytype&cas_no=24072-75-1&endpointlist=MN) (also cited in Witt et al. 2000)

[NTP] National Toxicology Program. 2000. Short-term study toxicity. Prechronic toxicity for 5,6'-dichloro-2-benzothiazolamine (24072-75-1). Study ID C61712. National Institutes of Health's National Institute of Environmental Health Sciences (NIEHS). Research Triangle Park, NC. [Accessed from the NTP Database on Nov. 23, 2009]. Available from: <http://ntp-server.niehs.nih.gov/index.cfm?objectid=03621D4C-B63D-AED4-CB677FD7CC46D300>

[OASIS Forecast] Optimized Approach Based on Structural Indices Set [Internet]. 2005. Version 1.20. Bourgas (BG): Bourgas Prof. Assen Zlatarov University, Laboratory of Mathematical Chemistry. [. Available from: <http://oasis-lmc.org/?section==software>

[OECD] Organisation for Economic Co-operation and Development. 2004. Emission scenario document on adhesive formulation [Internet]. Final report. Paris (FR): OECD, Environment Directorate. (Series on Emission Scenario Documents). Available from: <http://ascouncil.org/news/adhesives/docs/EPAFormulation.pdf>

[OECD] Organisation for Economic Co-operation and Development. 2007. Draft emission scenario on textile manufacturing wool mills [Internet]. Paris (FR): OECD, Environment Directorate. Report No.: ENV/JM/EEA(2004)8/1/REV, JT00175156. Available from: <http://www.oecd.org/dataoecd/2/47/34003719.pdf>

Pagga U, Brown D. 1986. The degradation of dyestuffs: Part II Behaviour of dyestuffs in aerobic biodegradation tests. *Chemosphere* 15(4):479-491.

Peters AT, Gbadamosi NMA. 1992. 5,6-(6,7-) dichlorobenzothiazolylazo dyes for synthetic-polymer fibres. *Dyes and Pigments* 18:115-123.

Peters AT, Tsatsaroni E, Xisai M. 1992. Hetarylazo disperse dyes derived from 5,6-dichloro- and 6,7-dichloro-2 aminobenzothiazoles. *Dyes and Pigments* 20:41-51.

Platzek T, Lang C, Grohmann G, Gi US, Baltes W. 1999. Formation of a carcinogenic aromatic amine from an azo dye by human skin bacteria in vitro. *Human & Experimental Toxicology*. 18: 552-559.

Razo-Flores E, Luijten M, Donlon B, Lettinga G, Field J. 1997. Biodegradation of selected azo dyes under methanogenic conditions. *Water Science and Technology* 36(6-7):65-72.

Refferscheid G & Heil J. 1996. Validation of the SOS/umu test using results of 486 chemicals and comparison with the Ames test and carcinogenicity data. *Mutation Research*. 369:129-145.

[RIVM] Rijksinstituut voor Volksgezondheid en Milieu. 2005. ConsExpo 4.0. Consumer Exposure and Uptake Models. Program Manual [Internet]. Report No.: 320104004/2005. Bilthoven (NL): RIVM (National Institute for Public Health and the Environment). [cited 2009 November 16]. Available from: [<http://rivm.openrepository.com/rivm/bitstream/10029/7307/1/320104004.pdf>]

Safepharm Laboratories Ltd. 1990. Acute toxicity to rainbow trout. Project number 47/781. Challenge submission ID#11347.

Sakuratani Y, Noguchi Y, Kobayashi K, Yamada J, Nishihara T. 2008. Molecular size as a limiting characteristic for bioconcentration in fish. *Journal of Environmental Biology* 29(1):89-92.

Sandoz. 1975. Acute Fish Toxicity (Rainbow trout), 48 hr, Voluntary Data Submission for batch 5 substances collected under the Chemical Management Plan Challenge initiative. Data prepared by Environment Canada..

Sarex Overseas. 1995. Material Safety Data Sheet (MSDS) for SARAPERSE RED 4G [Internet]. Available from: <http://www.sarex.com/dyesmsds/PS616.HTM>

Savarino P, Viscardi G, Carpignano R, Barni E. 1989. Technical properties and photofading of disperse heterocyclic azo dyes. *Dyes and Pigments* 10:269-283.

Seifried HE, Seifried RM, Clarke JJ, Junghans TB, San RH. 2006. A compilation of two decades of mutagenicity test results with the Ames Salmonella typhimurium and L5178Y mouse lymphoma cell mutation assays. *Chemical Research in Toxicology*. 19:627-44.

Shen G, Hu S (Environmental Testing Laboratory, Shanghai Academy of Environmental Sciences, Shanghai, China). 2008. Bioconcentration Test of C.I. Disperse Orange 30 in Fish. Prepared for Dystar in the name of Ecological and Toxicological Association of the Dyes and Organic Pigments Manufacturers (ETAD), Basel, Switzerland. Report No. S-070-2007. Submitted to Environment Canada in April 2008. Challenge Submission ID#8351.

Sijm DTHM, Schuurmann G, deVries PJ, Opperhuizen A. 1999. Aqueous solubility, octanol solubility, and octanol/water partition coefficient of nine hydrophobic dyes. *Environmental Toxicology and Chemistry* 18(6):1109-1117.

S.M.S Technology Co., Ltd. [not dated]. Material Safety Data Sheet (MSDS) for Navacron Disperse Scarlet GS [Internet]. Available from: [http://www.intonline.org/product/nava-doc/navacron-msds/9-Navacron%20dyes/msds\(Navacron%20Scarlet%20GS\).pdf](http://www.intonline.org/product/nava-doc/navacron-msds/9-Navacron%20dyes/msds(Navacron%20Scarlet%20GS).pdf)

Stingley R, Zou W, Heinze T, Chen H, Cerniglia C. 2009. Metabolism of azo dyes by human skin microbiota. *Journal of Medical Microbiology*. 2009 Sep 3. [Epub ahead of print].

Talaska G. 2003. Aromatic amines and human urinary bladder cancer: exposure sources and epidemiology. *Journal of Environmental Science and Health, Part C: Environmental Carcinogenesis & Ecotoxicology Reviews* 21(1): 29–43.

Tincher WC. 1988. Dyes in the environment: dyeing wastes in landfill. Study sponsored by the U.S. Operating Committee of ETAD.

[TOPKAT] Toxicity Prediction Program [Internet]. 2004. Version 6.2. San Diego (CA): Accelrys Software Inc. Available from: <http://www.accelrys.com/products/topkat/index.html>

Toxtree version 1.60. 2009. Developed by Ideaconsult Ltd Bulgaria.

[US EPA] United States Environmental Protection Agency. 2002a. PBT Profiler Methodology [Internet]. Washington (DC): U.S. EPA, Office of Pollution Prevention and Toxics. [cited 2008 August] Available from: <http://www.pbtp profiler.net/methodology.asp>

[US EPA] United States Environmental Protection Agency. 2002b. TSCA New Chemicals Program (NCP) Chemical Categories: Aminobenzothiazole Azo Dyes. Available from: <http://www.epa.gov/oppt/newchemicals/pubs/cat02.htm#Aminobenzothiazole%20Azo%20Dyes>

[US EPA] United States Environmental Protection Agency. 2009. Inventory update reporting, past IUR data: Non-confidential production volume information submitted by companies under the 1986, 1990, 1994, 1998, and 2002 Inventory Update Reporting Regulation: CAS RN 16586-42-8 & CAS RN 25176-89-0 [Internet]. Washington (DC): U.S. Environmental Protection Agency [cited 2009 March 25] Available from: <http://www.epa.gov/oppt/iur/tools/data/2002-vol.htm>

Vineis P, Pirastu R. 1997. Aromatic amines and cancer. *Cancer Causes Control* 8(3): 346–355.

[WATERNT] Water Solubility Program [Estimation model]. 2002. Version 1.00. Washington (DC): U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics; Syracuse (NY): Syracuse Research Corporation. Available from: <http://www.epa.gov/oppt/exposure/pubs/episuite.htm>

- Weber EJ, Adams RL. 1995. Chemical and sediment –mediated reduction of the azo dye Disperse Blue 79. *Environmental Science and Technology* 29(5):1163-1170.
- Weber E, Colon D, Baughman GL. 2001. Sediment-associated reactions of aromatic amines. 1. Elucidation of sorption mechanisms. *Environmental Science and Technology* 35(12):2470-2475.
- Witt KL, Knapton A, Wehr CM, Hook GJ, Mirsalis J, Shelby MD, MacGregor JT. 2000. Micronucleated erythrocyte frequency in peripheral blood of B6C3F(1) mice from short-term, prechronic, and chronic studies of the NTP carcinogenesis bioassay program. *Environ Mol Mutagen*. 2000;36(3):163-94.
- [WSKOWWIN] Water Solubility for Organic Compounds Program for Microsoft Windows [Estimation Model]. 2000. Version 1.41. Washington (DC): U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics; Syracuse (NY): Syracuse Research Corporation.. Available from: [www.epa.gov/oppt/exposure/pubs/episuite.htm](http://www.epa.gov/oppt/exposure/pubs/episuite.htm)
- Xu H, Heinze TM, Chen S, Cerniglia CE, Chen H. 2007. Anaerobic Metabolism of 1-Amino-2-Naphthol-Based Azo Dyes (Sudan Dyes) by Human Intestinal Microflora. *Applied and Environmental Microbiology*. 73:7759-62.
- Yen CC, Perenich TA, Baughman GL. 1989. Fate of dyes in aquatic systems II. Solubility and octanol/water partition coefficients of disperse dyes. *Environmental Toxicology and Chemistry* 8(11):981-986.
- Yen CC, Perenich TA, Baughman GL. 1991. Fate of commercial disperse dyes in sediments. *Environmental Toxicology and Chemistry* 10:1009-1017.



## Appendix I - Robust Study Summaries for key studies

### Evaluation of experimental data using Kollig's approach\*: Water solubility

Item	Weight	Response	Mark
<b>Reference:</b> Sijm DTHM, Schuurmann G, De Vries PJ, and Opperhuizen A. 1999. Aqueous solubility, octanol solubility, and octanol/water partition coefficient of nine hydrophobic dyes. Environ Toxicol Chem 18(6):1109-1117.			
<b>Test substance:</b> CAS RN: 68133-69-7			
<b>Parameter:</b> Water solubility			
Could you repeat the experiment with available information?	5	Yes	4
Is a clear objective stated?	1	Yes	1
Is water quality characterized or identified (distilled or deionized)?	2	Yes, distilled	2
Are the results presented in detail, clearly and understandably?	3	Yes	2
Are the data from a primary source and not from a referenced article?	3	Primary source	3
Was the chemical tested at concentrations below its water solubility?	5	N/A	N/A
Were particulates absent?	2	Assumed	2
Was a reference chemical of known constant tested?	3	No	0
Were other fate processes considered?	5	N/A	N/A
Was a control (blank) run?	3	Not indicated	0
Was temperature kept constant?	5	Not indicated but assumed. The water solubility was estimated using a generator column as was done by Opperhuizen 1986	5
Was the experiment done near room temperature (15–30°C)?	3	Not indicated but assumed	3
Is the purity of the test chemical reported (> 98%)?	3	No, but the chemicals were obtained from Bayer AG and then recrystallized in dichloromethane to remove any additives prior to use	3
Was the chemical's identity proven?	3	Yes, guaranteed by Bayer AG	3
Is the source of the chemical reported?	1	Yes, Bayer AG	1
<b>Score:</b>	29/37 = 78%		
<b>Degree of reliability**</b>	2 Satisfactory confidence		

\* Kollig, H.P. 1988. Criteria for evaluating the reliability of literature data on environmental process constants. Environ Toxicol Chem 17:287-311.

\*\* The reliability code for ecotoxicological studies of DSL categorization is used.

**Evaluation of experimental data using Kollig's approach\*: Octanol/water partition coefficient ( $K_{ow}$ )**

Item	Weight	Response	Mark
<b>Reference:</b> Sijm DTHM, Schuurmann G, De Vries PJ, and Opperhuizen A. 1999. Aqueous solubility, octanol solubility, and octanol/water partition coefficient of nine hydrophobic dyes. Environ Toxicol Chem 18(6):1109-1117.			
<b>Test substance:</b> CAS RN: 68133-69-7			
<b>Parameter:</b> Octanol/water partition coefficient ( $K_{ow}$ )			
Could you repeat the experiment with available information?	5	Yes	5
Is a clear objective stated?	1	Yes	1
Is water quality characterized or identified (distilled or deionized)?	2	Distilled	2
Are the results presented in detail, clearly and understandably?	3	Yes	3
Are the data from a primary source and not from a referenced article?	3	Yes	3
Was the chemical tested at concentrations below its water solubility?	5	N/A	N/A
Were particulates absent?	2	Yes	2
Was a reference chemical of known constant tested?	3	No	0
Were other fate processes considered?	5	N/A	N/A
Was a control (blank) run?	3	Not indicated	0
Was temperature kept constant?	5	Yes, used the slow stirring method protocol from De Bruijn et al. 1989	5
Was the experiment done near room temperature (15–30°C)?	3	Yes, 25°C (slow stirring method protocol from De Bruijn et al. 1989)	3
Is the purity of the test chemical reported (> 98%)?	3	No, but the chemicals were obtained from Bayer AG and then recrystallized in dichloromethane to remove any additives prior to use	3
Was the chemical's identity proven?	3	Yes, guaranteed by Bayer AG	3
Is the source of the chemical reported?	1	Yes, Bayer AG	1
<b>Score:</b>	31/37 = 83%		
<b>Degree of reliability**</b>	1 High confidence		

\* Kollig, H.P. 1988. Criteria for evaluating the reliability of literature data on environmental process constants. Environ Toxicol Chem 17:287-311.

\*\* The reliability code for ecotoxicological studies of DSL categorization is used.

Robust Study Summary Form: Persistence in Water, Sediments, and Soil				
No	Item	Weight	Yes/No	Specify
1	Reference: 13365Submission027 Foron Rubin RD-S Presskuchen trocken (Disperse Red 179). Inherent Biodegradability - Evaluation of the Aerobic Biodegradability in an Aqueous Medium: Zahn-Wellens / EMPA Test. BMG report no. 709/c-03, October 2003. Submitted to Environment Canada through the section 71 survey (Environment Canada 2009a)			
2	Substance identity: CAS RN	n/a	n	Not specified, but it is Disperse Red 179 (16586-42-8)
3	Substance identity: chemical name(s)	n/a	n	Foron Rubin RD-S
4	Chemical composition of the substance	2	n	Only the TOC content of the substance is reported. No mention of secondary products.
5	Chemical purity	1	n	The commercial product itself, Foron Rubine RD-S, is tested. It contains 34.3% w/w of 16586-42-8.
<b>Method</b>				
6	Reference	1	Y	The test is Zahn-Wellens / EMPA Test
7	OECD, EU, national, or other standard method?	3	Y	
8	Justification of the method/protocol if a non-standard method was used	2		Not applicable
9	GLP (good laboratory practice)	3	n	Not clearly indicated
<b>Test design/ conditions</b>				
10	Test type (hydrolysis, biodegradation, etc.)	n/a	y	Biodegradation
11	Test conditions type (aerobic or anaerobic)	n/a	Y	Aerobic
12	Test medium (water, sediment, or soil)	n/a	Y	Activated sludge
13	Test duration	n/a	Y	28 days
14	Negative or positive controls?	1	Y	Positive control with diethyleneglycol
15	Number of replicates (including controls)	1	Y	2 replicates for the test, 2 replicates for the blank and 1 for the test control
16	Measured concentrations reported?	3	Y	The degradation of the test material was monitored by the determination of the inorganic carbon (IC) at regular time intervals. Concentrations of the chemical of interest were not measured

				during the test.
17	Analytical method / instrument	1	Y	Inorganic carbon (IC) was determined in the same way as DOC without sparging the samples before analysis.
<b>Details on Biodegradation</b>				
18	Type of biodegradation (ready or inherent) reported?	2	y	Inherent biodegradation investigated according to Zahn-Wellens test
19	When type of biodegradation (ready or inherent) is not reported, is there is indirect information allowing for identification of biodegradation type?	1		n/a
20	Inoculum source	1	Y	It is mentioned that the inoculum is from a waste treatment plant. The name of the plant is not mentioned, however.
21	Inoculum concentration or number of micro-organisms	1	Y	0.2 g/L of dry matter
22	Were inoculum pre-conditioning and pre-adaptation reported?	1	N	
23	Were inoculum pre-conditioning and pre-adaptation appropriate for the method used?	n/a		n/a
24	Temperature	1	Y	22 ± 0.5°C, in dark room
25	Has percentage degradation of the reference compound reached the pass levels by day 14?	n/a	Y	Diethyleneglycol was 99% degraded by the 14th day.
26	<b>Soil:</b> soil moisture reported?	1		n/a
27	<b>Soil and sediments:</b> background SOM (Soil Organic Matter) content reported?	1		n/a
28	<b>Soil and sediments:</b> clay content reported?	1		n/a
29	<b>Soil and sediments:</b> CEC (Cation Exchange Capacity) reported?	1		n/a
<b>Details on Hydrolysis</b>				
30	pH values reported?	1		n/a
31	Temperature	1		n/a
32	Were appropriate concentrations of the substance used?			n/a

33	If solvent was used, was it done appropriately?			n/a
<b>Details on Photodegradation</b>				
34	Temperature	1		n/a
35	Light source	1		n/a
36	Light spectrum (nm)	1		n/a
37	Relative intensity based on sunlight intensity	1		n/a
38	Spectrum of a substance	1		n/a
39	<b>Indirect photolysis:</b> sensitizer (type)	1		n/a
40	<b>Indirect photolysis:</b> concentration of sensitizer	1		n/a
<b>Results</b>				
41	Endpoint and value	n/a	n/a	0% degradation. The 99% compound elimination is due to adsorption or sedimentation, not biodegradation.
42	Breakdown products	n/a		
43	<b>Score: ... %</b>	<b>68.2</b>		
44	<b>Environment Canada reliability code:</b>	<b>2</b>		
45	<b>Reliability category (high, satisfactory, low):</b>	<b>Satisfactory Confidence</b>		

Robust Study Summary Form: Persistence in Water, Sediments, and Soil				
No	Item	Weight	Yes/No	Specify
1	Reference: 13365Submission028, CI Disperse Red 153. Inherent Biodegradability - Evaluation of the Aerobic Biodegradability in an Aqueous Medium. BMG report no. 800/c-00 (CAS RN 25176-89-0) January 2000. Submitted to Environment Canada through the section 71 survey (Environment Canada 2009a)			
2	Substance identity: CAS RN	n/a	n	CAS RN 25176-89-0
3	Substance identity: chemical name(s)	n/a	n	C.I. Disperse Red 153
4	Chemical composition of the substance	2	n	Only the TOC content of the substance is reported. No mention of secondary products (i.e., is this 100% 25176-89-0).
5	Chemical purity	1	n	The product tested is C.I. Disperse Red 153
<b>Method</b>				
6	Reference	1	Y	The test is Zahn-Wellens / EMPA Test
7	OECD, EU, national, or other standard method?	3	Y	
8	Justification of the method/protocol if a non-standard method was used	2		n/a
9	GLP (good laboratory practice)	3	n	Not clearly indicated
<b>Test design/conditions</b>				
10	Test type (hydrolysis, biodegradation, etc.)	n/a	y	Biodegradation
11	Test conditions type (aerobic or anaerobic)	n/a	Y	Aerobic
12	Test medium (water, sediment, or soil)	n/a	Y	Activated sludge
13	Test duration	n/a	Y	28 days
14	Negative or positive controls?	1	Y	Positive control with diethyleneglycol
15	Number of replicates (including controls)	1	Y	2 replicates for the test, 2 replicates for the blank and 1 for the test control
16	Measured concentrations reported?	3	Y	The degradation of the test material was monitored by the determination of the dissolved organic carbon (DOC) at regular time intervals. Concentrations of the chemical of interest were not measured during the test.

17	Analytical method / instrument	1	Y	The DOC was determined in duplicate with a Shimadzu 5050 TOC-Analyzer using the NPOC-mode. Inorganic carbon (IC) was determined in the same way as DOC without sparging the samples before analysis.
<b>Details on Biodegradation</b>				
18	Type of biodegradation (ready or inherent) reported?	2	Y	Inherent biodegradation investigated according to Zahn-Wellens test
19	When type of biodegradation (ready or inherent) is not reported, is there is indirect information allowing for identification of biodegradation type?	1		n/a
20	Inoculum source	1	Y	The inoculum is from a waste treatment plant. The name of the plant is not mentioned, however.
21	Inoculum concentration or number of micro-organisms	1	Y	0.2 g/L of dry matter
22	Were inoculum pre-conditioning and pre-adaptation reported?	1	N	
23	Were inoculum pre-conditioning and pre-adaptation appropriate for the method used?	n/a		n/a
24	Temperature	1	Y	22 ± 0.5°C, in dark room
25	Has percentage degradation of the reference compound reached the pass levels by day 14?	n/a	Y	The reference compound reached 87% degradation after 14 days.
26	<b>Soil:</b> soil moisture reported?	1		n/a
27	<b>Soil and sediments:</b> background SOM (Soil Organic Matter) content reported?	1		n/a
28	<b>Soil and sediments:</b> clay content reported?	1		n/a
29	<b>Soil and sediments:</b> CEC (Cation Exchange Capacity) reported?	1		n/a
<b>Details on Hydrolysis</b>				
30	pH values reported?	1		n/a
31	Temperature	1		n/a
32	Were appropriate concentrations of the substance used?			n/a
33	If solvent was used, was it done appropriately?			n/a
<b>Details on Photodegradation</b>				
34	Temperature	1		n/a
35	Light source	1		n/a
36	Light spectrum (nm)	1		n/a

37	Relative intensity based on sunlight intensity	1		n/a
38	Spectrum of a substance	1		n/a
39	<b>Indirect photolysis:</b> sensitiser (type)	1		n/a
40	<b>Indirect photolysis:</b> concentration of sensitiser	1		n/a
<b>Results</b>				
41	Endpoint and value	n/a	n/a	Not degraded
42	Breakdown products	n/a		
43	<b>Score: ... %</b>	<b>68.2</b>		
44	<b>Environment Canada reliability code:</b>	<b>2</b>		
45	<b>Reliability category (high, satisfactory, low):</b>	<b>Satisfactory Confidence</b>		



Robust Study Summary Form: Aquatic iT				
No	Item	Weight	Yes/No	Specify
1	Reference: 13365 Submission 025 Foron Rubin RD-S Presskuchen trocken (Disperse Red 179) 96-hr Acute Toxicity to <i>Poecilia reticulata</i> (Guppy). BMG report no. 709/b-03, October 2003. Submitted to Environment Canada through the section 71 survey (Environment Canada 2009a)			
2	Substance identity: CAS RN	n/a	Y	The chemical tested is Foron Rubin RD-S Presskuchen trocken
3	Substance identity: chemical name(s)	n/a	Y	Foron Rubin RD-S Presskuchen trocken (Disperse Red 179)
4	Chemical composition of the substance	2	N	Composition of product not presented
5	Chemical purity	1	N	Indicated by section 71, not the toxicity study. The test indicates 100% active ingredient, which is impossible according to section 71.
6	Persistence/stability of test substance in aquatic solution reported?	1	N	No information
<b>Method</b>				
7	Reference	1	Y	The test was completed according to OECD Guideline Procedure 203 and EEC directive 92/69/EEC
8	OECD, EU, national, or other standard method?	3	Y	OECD and European Economic Community
9	Justification of the method/protocol if a non-standard method was used	2		Not applicable
10	GLP (good laboratory practice)	3	N	Not specified
<b>Test organism</b>				
11	Organism identity: name	n/a	Y	<i>Poecilia reticulata</i> (guppy)
12	Latin or both Latin and common names reported?	1	Y	<i>Poecilia reticulata</i> (guppy)
13	Life cycle age / stage of test organism	1	N	The life cycle stage of the test organisms is not specified, but it is believed that there are discrepancies due to the variation in length and especially weight.
14	Length and/or weight	1	Y	This is an issue since large variation can be observed between fish.
15	Sex	1	N	
16	Number of organisms per replicate	1	Y	Minimum allowed by test protocol: 7 fish
17	Organism loading rate	1	Y	Organism loading rates are < 1 g fish/L. Those are 0.533 for 100 mg/L, 0.563 for 10 mg/L and 0.538 for 1 mg/L.
18	Food type and feeding periods during the acclimation period	1	Y	
<b>Test design/ conditions</b>				
19	Test type (acute or chronic)	n/a	Y	Acute

20	Experiment type (laboratory or field)	n/a	Y	Laboratory
21	Exposure pathways (food, water, both)	n/a	Y	Water
22	Exposure duration	n/a	Y	96 hours
23	Negative or positive controls (specify)	1	Y	Positive
24	Number of replicates (including controls)	1	Y	A total of 4 replicates (1 for each concentration and 1 for the control)
25	Nominal concentrations reported?	1	Y	4 including control
26	Measured concentrations reported?	3	N	In fact, the toxicity reported exceeds the compound's solubility.
27	Food type and feeding periods during the long-term tests	1		Not applicable
28	Were concentrations measured periodically (especially in the chronic test)?	1	N	
29	Were the exposure media conditions relevant to the particular chemical reported? (e.g., for metal toxicity – pH, DOC/TOC, water hardness, temperature)	3	Y	
30	Photoperiod and light intensity	1	Y	Photoperiod of 16, no idea of actual intensity
31	Stock and test solution preparation	1	Y	Due to the limited water solubility, the individual test concentrations were prepared by adding the respective amounts of an acetonic stock solution to the empty glass vessels. After complete evaporation of the solvent, the tap water was added. Details of the stock solutions are also available.
32	Was solubilizer/emulsifier used if the chemical was poorly soluble or unstable?	1	Y	Due to the limited water solubility, the individual test concentrations were prepared by adding the respective amounts of an acetonic stock solution to the empty glass vessels. After complete evaporation of the solvent, the tap water was added.
33	If solubilizer/emulsifier was used, was its concentration reported?	1	N	No, but the acetone evaporated.
34	If solubilizer/emulsifier was used, was its ecotoxicity reported?	1	N	The acetone evaporated and therefore was absent.
35	Analytical monitoring intervals	1	N	
36	Statistical methods used	1	N	
<b>Information relevant to the data quality</b>				
37	Was the endpoint directly caused by the chemical's toxicity, not by the organism's health (e.g., when mortality in the control >10%) or physical effects (e.g. "shading effect")?	n/a		
38	Was the test organism relevant to the Canadian environment?	3	N	

39	Were the test conditions (pH, temperature, DO, etc.) typical for the test organism?	1	Y	pH was a little high at 8.1–8.5; oxygen concentrations were normal at 6.9–7.9 mg/L.
40	Do system type and design (static, semi-static, flow-through; sealed or open; etc.) correspond to the substance's properties and the organism's nature/habits?	2	Y	
41	Was pH of the test water within the range typical for the Canadian environment (6 to 9)?	1	Y	
42	Was temperature of the test water within the range typical for the Canadian environment (5 to 27°C)?	1	Y	
43	Was toxicity value below the chemical's water solubility?	3	N	
<b>Results</b>				
44	Toxicity values (specify endpoint and value)	n/a	n/a	
45	Other endpoints reported - e.g., BCF/BAF, LOEC/NOEC (specify)?	n/a	Y	NOEC > 10 mg/L based on nominal concentration
46	Other adverse effects (e.g., carcinogenicity, mutagenicity) reported?	n/a	Y	Loss of coordination, hypoactivity and swimming on the back were also reported.
47	<b>Score: ... %</b>	<b>50.0</b>		
48	<b>Environment Canada reliability code:</b>	<b>3</b>		
49	<b>Reliability category (high, satisfactory, low):</b>	<b>Low Confidence</b>		

Robust Study Summary Form: Aquatic iT				
No	Item	Weight	Yes/No	Specify
1	Reference: 13365 Submission 026 C.I. Disperse Red 153. 96-hour Acute Toxicity to <i>Poecilia reticulata</i> (Guppy) Limit Test (100 mg/L). BMG report no. 800/a-00 submitted to Environment Canada through the section 71 survey (Environment Canada 2009a)			
2	Substance identity: CAS RN	n/a		25176-89-0
3	Substance identity: chemical name(s)	n/a		C.I. Disperse Red 153
4	Chemical composition of the substance	2	N	The substance is identified as C.I. Disperse Red 153, Batch # 99L094 Muster 100811B, with 100% active ingredient. Little information.
5	Chemical purity	1	N	The test report mentions 100% purity, but it is unclear whether this refers to 100% purity of commercial product or CAS RN.
6	Persistence/stability of test substance in aquatic solution reported?	1	N	No information
<b>Method</b>				
7	Reference	1	Y	The test was completed according to OECD Guideline Procedure 203 and EEC directive 92/69/EEC
8	OECD, EU, national, or other standard method?	3	Y	OECD and European Economic Community
9	Justification of the method/protocol if a non-standard method was used	2		Not applicable
10	GLP (good laboratory practice)	3	N	Not specified
<b>Test organism</b>				
11	Organism identity: name	n/a	Y	<i>Poecilia reticulata</i> (guppy)
12	Latin or both Latin and common names reported?	1	Y	<i>Poecilia reticulata</i> (guppy)
13	Life cycle age / stage of test organism	1	N	The life cycle stage of the test organisms is not specified, but it is believed that there are discrepancies due to the variation in length and especially weight.
14	Length and/or weight	1	Y	This is an issue since large variation can be observed between fish.
15	Sex	1	N	
16	Number of organisms per replicate	1	Y	Minimum allowed by test protocol: 7 fish

17	Organism loading rate	1	Y	Organism's loading rates are < 1 gram of fish/L. It is 0.584 g for 100 mg/L.
18	Food type and feeding periods during the acclimation period	1	Y	
<b>Test design/conditions</b>				
19	Test type (acute or chronic)	n/a	Y	Acute
20	Experiment type (laboratory or field)	n/a	Y	Laboratory
21	Exposure pathways (food, water, both)	n/a	Y	Water
22	Exposure duration	n/a	Y	96 hours
23	Negative or positive controls (specify)	1	Y	Positive
24	Number of replicates (including controls)	1	Y	Two replicates (100 mg/L and control)
25	Nominal concentrations reported?	1	Y	Only one nominal concentration
26	Measured concentrations reported?	3	N	
27	Food type and feeding periods during the long-term tests	1		Not applicable
28	Were concentrations measured periodically (especially in the chronic test)?	1	N	
29	Were the exposure media conditions relevant to the particular chemical reported? (e.g., for metal toxicity - pH, DOC/TOC, water hardness, temperature)	3	Y	
30	Photoperiod and light intensity	1	Y	Photoperiod of 16, no idea of actual intensity.
31	Stock and test solution preparation	1	Y	Due to the limited water solubility, the individual test concentrations were prepared by adding the respective amounts of an acetonic stock solution to the empty glass vessels. After complete evaporation of the solvent, the tap water was added. Details of the stock solutions are also available.
32	Was solubilizer/emulsifier used if the chemical was poorly soluble or unstable?	1	Y	Due to the limited water solubility, the individual test concentrations were prepared by adding the respective amounts of an acetonic stock solution to the empty glass vessels. After complete evaporation of the solvent, the tap water was added.
33	If solubilizer/emulsifier was used, was its concentration reported?	1	N	No, but the acetone evaporated.
34	If solubilizer/emulsifier was used, was its ecotoxicity reported?	1	N	The acetone evaporated and therefore was absent.
35	Analytical monitoring intervals	1	N	
36	Statistical methods used	1	N	

<b>Information relevant to the data quality</b>				
37	Was the endpoint directly caused by the chemical's toxicity, not by the organism's health (e.g., when mortality in the control >10%) or physical effects (e.g., "shading effect")?	n/a		
38	Was the test organism relevant to the Canadian environment?	3	N	
39	Were the test conditions (pH, temperature, DO, etc.) typical for the test organism?	1	Y	pH was a little high at 8.6–8.9; oxygen concentration were normal at 7.5–7.9 mg/L.
40	Do system type and design (static, semi-static, flow-through; sealed or open; etc.) correspond to the substance's properties and the organism's nature/habits?	2	Y	
41	Was pH of the test water within the range typical for the Canadian environment (6 to 9)?	1	Y	
42	Was temperature of the test water within the range typical for the Canadian environment (5 to 27°C)?	1	Y	
43	Was toxicity value below the chemical's water solubility?	3	N	
<b>Results</b>				
44	Toxicity values (specify endpoint and value)	n/a	n/a	
45	Other endpoints reported - e.g., BCF/BAF, LOEC/NOEC (specify)?	n/a	Y	NOEC > 100 mg/L based on nominal concentrations
46	Other adverse effects (e.g., carcinogenicity, mutagenicity) reported?	n/a	Y	Loss of coordination, hypoactivity and swimming on the back were looked for.
47	<b>Score: ... %</b>	<b>50.0</b>		
48	<b>Environment Canada reliability code:</b>	<b>3</b>		
49	<b>Reliability category (high, satisfactory, low):</b>	<b>Low Confidence</b>		

Robust Study Summary Form: Aquatic B				
No	Item	Weight	Yes/No	Specify
1	<b>Reference:</b> Shen G, and Hu S. 2008. Bioconcentration Test of C.I. Disperse Orange 30 in Fish. Prepared by Environmental Testing Laboratory, Shanghai Academy of Environmental Sciences, Shanghai, China, for Dystar in the name of Ecological and Toxicological Association of the Dyes and Organic Pigments Manufacturers (ETAD), Basel, Switzerland. Report No. S-070-2007. Submitted to Environment Canada in April 2008. Challenge Submission ID#8351.			
2	Substance identity: CAS RN	n/a	Y	5261-31-4
3	Substance identity: chemical name(s)	n/a	Y	Propanenitrile, 3-[[2-(acetyloxy)ethyl][4-[(2,6-dichloro-4-nitrophenyl)azo]phenyl]amino]-
4	Chemical composition of the substance	2	N	
5	Chemical purity	1	N	
6	Persistence/stability of test substance in aquatic solution reported?	1	N	
7	If test material is radiolabelled, were precise position(s) of the labelled atom(s) and the percentage of radioactivity associated with impurities reported?	2	n/a	
	<b>Method</b>			
8	Reference	1	Y	OECD guidelines for the testing of chemicals No. 305B-1996
9	OECD, EU, national, or other standard method?	3	Y	OECD
10	Justification of the method/protocol if a non-standard method was used	2		
11	GLP (good laboratory practice)	3	N	
<b>Test organism</b>	<b>Test organism</b>			
12	Organism identity: name	n/a	Y	Zebra fish, <i>Brachydanio rerio</i>
13	Latin or both Latin and common names reported?	1	Y	Both
14	Life cycle age / stage of test organism	1	N	
15	Length and/or weight	1	Y	Mean body length $3.91 \pm 0.18$ cm and mean body weight $0.32 \pm 0.06$ g
16	Sex	1	N	
17	Number of organisms per replicate	1	Y	7
18	Organism loading rate	1	Y	20 mg/L
19	Food type and feeding periods during the acclimation period	1	Y	Fed a commercial fish diet until one day before start of test
	<b>Test design/conditions</b>			
20	Experiment type (laboratory or field)	n/a	Y	Laboratory

21	Exposure pathways (food, water, both)	n/a	Y	Water
22	Exposure duration	n/a	Y	28 days
23	Number of replicates (including controls)	1	Y	
24	Concentrations	1	Y	20 mg/L
25	Food type/composition and feeding periods during the test	1	Y	Fish were fed two hours before water renewal.
26	If BCF/BAF derived as a ratio of chemical concentration in the organism and in water, was experiment duration equal to or longer than the time required for the chemical concentrations to reach steady state?	3	Y	28 days
27	If BCF/BAF derived as a ratio of chemical concentration in the organism and in water, were measured concentrations in both water and organism reported?	3	Y	
28	Were concentrations in the test water measured periodically?	1	Y	On three separate days
29	Were the exposure media conditions relevant to the particular chemical reported? (e.g., for metal toxicity - pH, DOC/TOC, water hardness, temperature)	3	Y	Yes, every second day
30	Photoperiod and light intensity	1	Y	12:12
31	Stock and test solution preparation	1	Y	
32	Analytical monitoring intervals	1	Y	Every second day for dissolved oxygen, pH and temperature
33	Statistical methods used	1	Y	
34	Was solubilizer/emulsifier used if the chemical was unstable or poorly soluble?	n/a	N	
	<b>Information relevant to the data quality</b>			
35	Was the test organism relevant to the Canadian environment?	3	Y	
36	Were the test conditions (pH, temperature, DO, etc.) typical for the test organism?	1	Y	
37	Do system type and design (static, semi-static, flow-through; sealed or open; etc.) correspond to the substance's properties and the organism's nature/habits?	2	Y	Semi-static
38	Was pH of the test water within the range typical for the Canadian environment (6 to 9)?	1	Y	7.22–7.84
39	Was temperature of the test water within the range typical for the Canadian environment (5 to 27°C)?	1	Y	22–23



40	Was lipid content (or lipid-normalized BAF/BCF) reported?	2	Y	
41	Were measured concentrations of a chemical in the test water below the chemical's water solubility?	3	N	
42	If radiolabelled test substance was used, was BCF determination based on the parent compound (i.e., not on total radiolabelled residues)?	3	n/a	
	<b>Results</b>			
43	Endpoints (BAF, BCF) and values	n/a	n/a	BCF
44	BAF or BCF determined as: 1) the ratio of chemical concentration in the organism and in water, or 2) the ratio of the chemical uptake and elimination rate constants	n/a	n/a	1
45	Was BAF/BCF derived from a 1) tissue sample or 2) whole organism?	n/a	n/a	2
46	Was 1) average or 2) maximum BAF/BCF used?	n/a	n/a	1
47	<b>Score: ... %</b>	<b>75.0</b>		
48	<b>Environment Canada reliability code:</b>	<b>2</b>		
49	<b>Reliability category (high, satisfactory, low):</b>	<b>Satisfactory Confidence</b>		
50	<b>Comments</b>	<b>The present procedure is based on semi-static conditions (renewal of test solutions every 2 days). Therefore, test chemicals with very low water solubility can also be characterized as to their bioconcentration potential without adding solvents or other auxiliary substances which may affect the results.</b>		

Robust Study Summary Form: Aquatic iT				
No	Item	Weight	Yes/No	Specify
1	Reference: Sandoz. 1975. Acute fish toxicity (rainbow trout) 48hr			
2	Substance identity: CAS RN	n/a	Y	5261-31-4
3	Substance identity: chemical name(s)	n/a	Y	
4	Chemical composition of the substance	2	N	
5	Chemical purity	1	N	
6	Persistence/stability of test substance in aquatic solution reported?	1	N	
<b>Method</b>				
7	Reference	1	Y	
8	OECD, EU, national, or other standard method?	3	Y	
9	Justification of the method/protocol if a non-standard method was used	2		
10	GLP (good laboratory practice)	3	Y	
<b>Test organism</b>				
11	Organism identity: name	n/a	Y	Rainbow trout
12	Latin or both Latin and common names reported?	1	Y	
13	Life cycle age / stage of test organism	1	N	
14	Length and/or weight	1	Y	
15	Sex	1	N	
16	Number of organisms per replicate	1	N	
17	Organism loading rate	1	N	
18	Food type and feeding periods during the acclimation period	1	N	
<b>Test design/conditions</b>				
19	Test type (acute or chronic)	n/a	Y	Acute
20	Experiment type (laboratory or field)	n/a	Y	Laboratory
21	Exposure pathways (food, water, both)	n/a		
22	Exposure duration	n/a	Y	48
23	Negative or positive controls (specify)	1	N	
24	Number of replicates (including controls)	1	N	
25	Nominal concentrations	1	N	

	reported?			
26	Measured concentrations reported?	3	N	
27	Food type and feeding periods during the long-term tests	1	N	
28	Were concentrations measured periodically (especially in the chronic test)?	1	N	
29	Were the exposure media conditions relevant to the particular chemical reported? (e.g., for metal toxicity - pH, DOC/TOC, water hardness, temperature)	3	N	
30	Photoperiod and light intensity	1	N	
31	Stock and test solution preparation	1	N	
32	Was solubilizer/emulsifier used if the chemical was poorly soluble or unstable?	1	N	
33	If solubilizer/emulsifier was used, was its concentration reported?	1		
34	If solubilizer/emulsifier was used, was its ecotoxicity reported?	1		
35	Analytical monitoring intervals	1	N	
36	Statistical methods used	1	N	
<b>Information relevant to the data quality</b>				
37	Was the endpoint directly caused by the chemical's toxicity, not by the organism's health (e.g., when mortality in the control > 10%) or physical effects (e.g., "shading effect")?	n/a		
38	Was the test organism relevant to the Canadian environment?	3	Y	
39	Were the test conditions (pH, temperature, DO, etc.) typical for the test organism?	1	N	
40	Do system type and design (static, semi-static, flow-through; sealed or open; etc.) correspond to the substance's properties and the organism's nature/habits?	2	N	
41	Was pH of the test water within the range typical for the Canadian environment (6 to 9)?	1	N	
42	Was temperature of the test water within the range typical for the Canadian environment (5 to 27°C)?	1	Y	

43	Was toxicity value below the chemical's water solubility?	3	N	
<b>Results</b>				
44	Toxicity values (specify endpoint and value)	n/a	n/a	48-hour LC <sub>50</sub> > 700 mg/L
45	Other endpoints reported - e.g., BCF/BAF, LOEC/NOEC (specify)?	n/a		
46	Other adverse effects (e.g., carcinogenicity, mutagenicity) reported?	n/a		
47	<b>Score: ... %</b>	<b>28.9</b>		
48	<b>Environment Canada reliability code:</b>	<b>4</b>		
49	<b>Reliability category (high, satisfactory, low):</b>	<b>Not Satisfactory</b>		
50	<b>Comments</b>			

Robust Study Summary Form: Aquatic iT				
No	Item	Weight	Yes/No	Specify
1	Reference: Safepharm Laboratories Ltd. 1990. Acute toxicity to rainbow trout. Project number 47/781			
2	Substance identity: CAS RN	n/a	Y	5261-31-4
3	Substance identity: chemical name(s)	n/a	Y	
4	Chemical composition of the substance	2	N	
5	Chemical purity	1	N	
6	Persistence/stability of test substance in aquatic solution reported?	1	N	
<b>Method</b>				
7	Reference	1	N	
8	OECD, EU, national, or other standard method?	3	N	
9	Justification of the method/protocol if a non-standard method was used	2	N	
10	GLP (good laboratory practice)	3		n/a
<b>Test organism</b>				
11	Organism identity: name	n/a		Rainbow trout
12	Latin or both Latin and common names reported?	1	Y	
13	Life cycle age / stage of test organism	1	Y	
14	Length and/or weight	1	Y	
15	Sex	1		n/a
16	Number of organisms per replicate	1	Y	Three to ten
17	Organism loading rate	1	Y	0.70 g body weight/L
18	Food type and feeding periods during the acclimation period	1		n/a since acute test
<b>Test design / conditions</b>				
19	Test type (acute or chronic)	n/a		Acute
20	Experiment type (laboratory or field)	n/a		Lab
21	Exposure pathways (food, water, both)	n/a		Water
22	Exposure duration	n/a		96 hours
23	Negative or positive controls (specify)	1	Y	Positive
24	Number of replicates (including controls)	1	Y	Two at definitive study
25	Nominal concentrations reported?	1	Y	3
26	Measured concentrations reported?	3	N	
27	Food type and feeding periods during the long-term tests	1		n/a
28	Were concentrations measured periodically (especially in the chronic test)?	1	N	

29	Were the exposure media conditions relevant to the particular chemical reported? (e.g., for metal toxicity - pH, DOC/TOC, water hardness, temperature)	3	Y	
30	Photoperiod and light intensity	1	N	
31	Stock and test solution preparation	1	N	
32	Was solubilizer/emulsifier used if the chemical was poorly soluble or unstable?	1	N	
33	If solubilizer/emulsifier was used, was its concentration reported?	1		n/a
34	If solubilizer/emulsifier was used, was its ecotoxicity reported?	1		n/a
35	Analytical monitoring intervals	1	Y	
36	Statistical methods used	1	N	
<b>Information relevant to the data quality</b>				
37	Was the endpoint directly caused by the chemical's toxicity, not by the organism's health (e.g., when mortality in the control > 10%) or physical effects (e.g., "shading effect")?	n/a	Y	
38	Was the test organism relevant to the Canadian environment?	3	Y	
39	Were the test conditions (pH, temperature, DO, etc.) typical for the test organism?	1	Y	
40	Do system type and design (static, semi-static, flow-through; sealed or open; etc.) correspond to the substance's properties and the organism's nature/habits?	2		n/a
41	Was pH of the test water within the range typical for the Canadian environment (6 to 9)?	1	N	No pH given
42	Was temperature of the test water within the range typical for the Canadian environment (5 to 27°C)?	1	Y	
43	Was toxicity value below the chemical's water solubility?	3	N	Water solubility for this substance was 0.07.
<b>Results</b>				
44	Toxicity values (specify endpoint and value)	n/a		96-hour LC <sub>50</sub> > 100 mg/L
45	Other endpoints reported - e.g., BCF/BAF, LOEC/NOEC (specify)?	n/a	N	
46	Other adverse effects (e.g., carcinogenicity, mutagenicity) reported?	n/a	N	
47	<b>Score: ... %</b>	<b>43.6</b>		
48	<b>Environment Canada reliability code:</b>	<b>3</b>		
49	<b>Reliability category (high, satisfactory, low):</b>	<b>Low Confidence</b>		
50	<b>Comments</b>			

## Appendix 2 – PBT Model Inputs Summary Table

Model Inputs for Disperse Red 179 (CAS RN 16586-42-8)			
	Physical and Chemical Fate	PBT Profiling	Persistence
Model input parameters	EPI Suite (all models, including AOPWIN, KOCWIN, BCFWIN, BIOWIN and ECOSAR)	Canadian-POPs (including CATABOL)	TOPKAT
SMILES code	<chem>N(=O)(=O)c(ccc(nc(N=Nc(c(cc(N(CCC(#N))CC)c1)C)c1)s2)c23)c3</chem>	<chem>N(=O)(=O)c(ccc(nc(N=Nc(c(cc(N(CCC(#N))CC)c1)C)c1)s2)c23)c3</chem>	<chem>N(=O)(=O)c(ccc(nc(N=Nc(c(cc(N(CCC(#N))CC)c1)C)c1)s2)c23)c3</chem>
Molecular weight (g/mol)	394.45		
Melting point (°C)			
Boiling point (°C)			
Data temperature (°C)			
Density (kg/m <sup>3</sup> )			
Vapour pressure (Pa)			
Henry's Law constant (Pa·m <sup>3</sup> /mol)			
Log K <sub>aw</sub> (air-water partition coefficient; dimensionless)			
Log K <sub>ow</sub> (octanol-water partition coefficient; dimensionless)	5.09 (value modelled using the "Experimental value adjustment method" of KOWWIN (2000), which estimated the log K <sub>ow</sub> of the substances based on the experimental log K <sub>ow</sub> value of 4.08 for the analogue CAS RN 68133-69-7 (Sijm et al. 1999))	Same as EPIWEB	Same as EPIWEB
K <sub>ow</sub> (octanol-water partition coefficient; dimensionless)			

<b>Log K<sub>oc</sub></b> (organic carbon-water partition coefficient – L/kg)			
<b>Water solubility (mg/L)</b>	0.012 (value modelled using the "Experimental value adjustment method" of WATERNT [2002], which estimated the water solubility of the substances based on the water solubility values of the analogue CAS 68133-69-7. The water solubility of the analogue [0.048mg/L] is a geometric average of CAS 68133-69-7 experimental solubility values [Sijm et al. 1999])		
<b>Log K<sub>oa</sub></b> (Octanol-air partition coefficient; dimensionless)			
<b>Soil-water partition coefficient (L/kg)<sup>1</sup></b>			
<b>Sediment-water partition coefficient (L/kg)<sup>1</sup></b>			
<b>Suspended particles-water partition coefficient (L/kg)<sup>1</sup></b>			
<b>Fish-water partition coefficient (L/kg)<sup>2</sup></b>			
<b>Aerosol-water partition coefficient; dimensionless<sup>3</sup></b>			
<b>Vegetation-water partition coefficient; dimensionless<sup>1</sup></b>			
<b>Enthalpy (K<sub>ow</sub>)</b>			
<b>Enthalpy (K<sub>aw</sub>)</b>			
<b>Half-life in air (days)</b>			
<b>Half-life in water (days)</b>			
<b>Half-life in sediment (days)</b>			
<b>Half-life in soil (days)</b>			



<b>Half-life in vegetation (days)<sup>4</sup></b>			
<b>Metabolic rate constant (1/days)</b>			
<b>Biodegradation rate constant (1/days) or (1/hr) -specify</b>			
<b>Biodegradation half-life in primary clarifier (t<sub>1/2-p</sub>) (hr)</b>			
<b>Biodegradation half-life in aeration vessel (t<sub>1/2-s</sub>) (hr)</b>			
<b>Biodegradation half-life in settling tank (t<sub>1/2-s</sub>) (hr)</b>			

<sup>1</sup> Derived from log K<sub>oc</sub><sup>2</sup> Derived from BCF data<sup>3</sup> Default value<sup>4</sup> Derived from half-life in water

Model Inputs for DAPEP (CAS RN 25176-89-0)			
	Physical and Chemical Fate	PBT Profiling	Persistence
Model input parameters	EPI Suite (all models, including AOPWIN, KOCWIN, BCFWIN, BIOWIN and ECOSAR)	Canadian-POPs (CATABOL)	TOPKAT
SMILES code	<chem>c12N=C(N=Nc3ccc(N(CC)CCC(#N))cc3)Sc1cc(Cl)c(Cl)c2</chem>	<chem>c12N=C(N=Nc3ccc(N(C(C)CCC(#N))cc3)Sc1cc(Cl)c(Cl)c2</chem>	<chem>c12N=C(N=Nc3ccc(N(CC)CCC(#N))cc3)Sc1cc(Cl)c(Cl)c2</chem>
Molecular weight (g/mol)	404.32		
Melting point (°C)			
Boiling point (°C)			
Data temperature (°C)			
Density (kg/m <sup>3</sup> )			
Vapour pressure (Pa)			
Henry's Law constant (Pa·m <sup>3</sup> /mol)			
Log K <sub>aw</sub> (air-water partition coefficient; dimensionless)			
Log K <sub>ow</sub> (octanol-water partition coefficient; dimensionless)	6.01 (value modelled using the "Experimental value adjustment method" of KOWWIN 2000, which estimated the log K <sub>ow</sub> of the substances based on the experimental log K <sub>ow</sub> value of 4.08 for the analogue CAS RN 68133-69-7 (Sijm et al. 1999))	Same as EPIWEB	Same as EPIWEB
K <sub>ow</sub> (octanol-water partition coefficient; dimensionless)			
Log K <sub>oc</sub> (organic carbon-water partition coefficient – L/kg)			

<b>Water solubility (mg/L)</b>	0.004 (value modelled using the "Experimental value adjustment method" of WATERNT [2002], which estimated the water solubility of the substances based on the water solubility values of the analogue CAS 68133-69-7. The water solubility of the analogue (0.048mg/L) is a geometric average of CAS 68133-69-7 experimental solubility values [Sijm et al. 1999])		
<b>Log K<sub>oa</sub> (Octanol-air partition coefficient; dimensionless)</b>			
<b>Soil-water partition coefficient (L/kg)<sup>1</sup></b>			
<b>Sediment-water partition coefficient (L/kg)<sup>1</sup></b>			
<b>Suspended particles-water partition coefficient (L/kg)<sup>1</sup></b>			
<b>Fish-water partition coefficient (L/kg)<sup>2</sup></b>			
<b>Aerosol-water partition coefficient; dimensionless<sup>3</sup></b>			
<b>Vegetation-water partition coefficient; dimensionless<sup>1</sup></b>			
<b>Enthalpy (K<sub>ow</sub>)</b>			
<b>Enthalpy (K<sub>aw</sub>)</b>			
<b>Half-life in air (days)</b>			
<b>Half-life in water (days)</b>			
<b>Half-life in sediment (days)</b>			
<b>Half-life in soil (days)</b>			
<b>Half-life in vegetation (days)<sup>4</sup></b>			
<b>Metabolic rate constant (1/days)</b>			

<b>Biodegradation rate constant (1/days) or (1/hr) -specify</b>			
<b>Biodegradation half-life in primary clarifier (<math>t_{1/2-p}</math>) (hr)</b>			
<b>Biodegradation half-life in aeration vessel (<math>t_{1/2-s}</math>) (hr)</b>			
<b>Biodegradation half-life in settling tank (<math>t_{1/2-s}</math>) (hr)</b>			

### Appendix 3. Upper-bounding exposure estimates of Disperse Red 179 and DAPEP from Textiles.

Consumer product	Upper-bounding exposure estimates (mg/kg-bw per day) of Disperse Red 179 and DAPEP by various age groups. <sup>1</sup>				
	0–6 months <sup>2</sup>	0.5–4 years <sup>3</sup>	5–11 years <sup>4</sup>	12–19 years <sup>5</sup>	20+ years <sup>6</sup>
Dermal: wearing of textiles	$(0.2 - 4) \times 10^{-3}$	$(0.2 - 3) \times 10^{-3}$	$(0.2 - 3) \times 10^{-3}$	$(0.1 - 2) \times 10^{-3}$	$(0.1 - 2) \times 10^{-3}$
Oral: mouthing	$0.002 \times 10^{-3}$	$0.001 \times 10^{-3}$	NA	NA	NA

<sup>1</sup> Upper-bounding leachable fraction was estimated to range from 0.03% for colourfast textiles (ETAD 2004), to 0.5% for textiles with poor colourfastness (Kraetke and Platzek 2005).

<sup>2</sup> Assumed to weigh 7.5 kg, have body surface area (excluding head and hands) of 0.28 m<sup>2</sup> (Health Canada 1998) and spend 23 min/d mouthing (Norris and Smith 2002).

<sup>3</sup> Assumed to weigh 15.5 kg, have body surface area (excluding head and hands) of 0.46 m<sup>2</sup> (Health Canada 1998) and spend 29 min/d mouthing (Norris and Smith 2002).

<sup>4</sup> Assumed to weigh 31.0 kg and have body surface area (excluding head and hands) of 0.80 m<sup>2</sup> (Health Canada 1998).

<sup>5</sup> Assumed to weigh 59.4 kg and have body surface area (excluding head and hands) of 1.4 m<sup>2</sup> (Health Canada 1998).

<sup>6</sup> Assumed to weigh 70.9 kg and have body surface area (excluding head and hands) of 1.6 m<sup>2</sup> (Health Canada 1998).

#### Appendix 4: Exposure estimates from dyed textiles

Consumer product scenario	Assumptions	Upper-bounding Estimated exposure
Wearing of dyed clothing made from synthetic fabrics	<p>Exposure scenario: ConsExpo 4.0, direct dermal contact with product: migration (RIVM 2005). Example for infants aged 0–6 months.</p> <p>Concentration: 1 wt % (Kraetke and Platzek 2005) Fabric Density: 100g/m<sup>2</sup> (Kraetke and Platzek 2005)</p> <p><b>General assumptions</b></p> <ul style="list-style-type: none"> <li>- Exposure frequency: 365 times/year</li> <li>- Body weight: 7.5 kg (Health Canada 1998)</li> <li>- Body Surface Area, excluding head and hands<sup>1</sup>: 0.28 m<sup>2</sup> (Health Canada 1998)</li> </ul> <p><b>Dermal route</b></p> <ul style="list-style-type: none"> <li>- Exposed area<sup>1</sup>: 0.28m<sup>2</sup> (Health Canada 1998)</li> <li>- Leachable Fraction: (Kraetke and Platzek 2005) 0.5%</li> <li>- Product amount<sup>2</sup>: 0.28g</li> <li>- Skin Contact Factor: 1 (fraction)</li> <li>- Uptake Fraction: 2 % (Kraetke and Platzek 2005)</li> </ul>	<p><b>Dermal chronic</b> Internal dose = 0.004 mg/kg-bw per day</p>
Mouthing of dyed fabrics	<p>Exposure is estimated below for infants of age 0–6 months (body weight 7.5 kg).</p> <p>The estimated daily intake for ingestion from mouthing:</p> $= \frac{WS \times V_s \times CF \times FR \times AF_o \times EF}{BW}$ <p>where;  <math>WS</math> = water solubility of Disperse Red 179 and DAPEP (Table 3) = 0.69 mg/L (Baughman and Perenich 1988)  <math>V_s</math> = salivary flow rate = 0.22 mL/min (Environ 2003a, b)  <math>CF</math> = Convert L to mL = 0.001 L/mL  <math>FR</math> = Fractional extraction by saliva = 0.5% [ETAD 1983]<sup>3</sup>  <math>AF_o</math> = Absorption factor by oral =1  <math>EF</math> = Exposure frequency of mouthing behaviour = 23 min/d (Norris and Smith 2002)  <math>BW</math> = body weight = 7.5 kg (infants, age 0–6 months) (Health Canada 1998)</p> <p>= (0.69 mg/L × 0.22 mL/min × 0.001 L/mL × 0.005 × 1 × 23 min)/7.5 kg  = 6x10<sup>-6</sup> mg/kg-bw per day</p>	<p><b>Oral chronic</b> internal dose = 6x10<sup>-6</sup> mg/kg-bw per day</p>

<sup>1</sup> This is assumed to equal the amount of fabric in contact the skin.

<sup>2</sup> Product amount = Fabric Density×Amount of Fabric×Concentration = (100g/m<sup>2</sup>)×(0.28 m<sup>2</sup>)×(1wt%) = 0.28g

<sup>3</sup> Maximum amount of dye extracted by simulated saliva from child oriented synthetic textiles after 4 hours was 0.13%, 0.5% is used to represent an upper bound.

## Appendix 5. (Q)SAR predictions for Disperse Red 179 (16586-42-8) and its potential azo cleavage products

### Carcinogenicity predictions

ID	CAS RN	Derek <sup>1</sup>	Toxtree <sup>2</sup>		Model Applier <sup>3</sup>				Casetox <sup>4</sup>			
		Cancer	SA gtx	Cancer	m-rat	f-rat	m-mice	f-mice	m-rat	f-rat	m-mice	f-mice
Parent	16586-42-8	P	P	N	P	IC	ND	ND	ND	ND	ND	ND
Metabolite 1	6285-57-0	P	P	-	P	P	ND	IC	P	N	N	P
Metabolite 2	105294-34-6	P	P	N	P	N	P	N	ND	ND	ND	ND

### Genotoxicity predictions

ID	CAS RN	Ames				ChrAb		Micronuclei induction		
		Derek	TT <sup>5</sup>	MA	CT	MA	CT <sup>6</sup>	TT	MA	CT
Parent	16586-42-8	P	P	P	ND	ND	IC	P	ND	ND
Metabolite 1	6285-57-0	P	-	P	ND	ND	ND	P	N <sup>7</sup>	P
Metabolite 2	105294-34-6	P	P	P	P	N	P	P	N	IC

CAS RN, Chemical Abstracts Service Registry Number; ChrAb, chromosomal aberration; CT, Casetox; f, female; IC, inconclusive; ID, identification; m, male; MA, Model Applier; N, negative; ND, not in domain of model; SA gtx, structural alert for genotoxic carcinogen; P, positive; TT, Toxtree; -, no result

<sup>1</sup> [DEREK] Deductive Estimation of Risk from Existing Knowledge [Prediction module on CD ROM]. 2008. Version 10.0.2. Cambridge (MA): Harvard University, LHASA Group. [cited 2009 Sep 30]. Available from: [http://www.lhasalimited.org/index.php?cat=2&sub\\_cat=2#](http://www.lhasalimited.org/index.php?cat=2&sub_cat=2#) [restricted access].

<sup>2</sup> Toxtree version 1.60. 2009. Developed by Ideaconsult Ltd Bulgaria.

<sup>3</sup> [Leadscope] Leadscope Model Applier [Prediction module]. 2009. Version 1.2.0-3. Columbus (OH): Leadscope, Inc. [cited 2009 Sep 30]. Available from: [http://www.leadscope.com/all\\_products.php](http://www.leadscope.com/all_products.php) [restricted access].

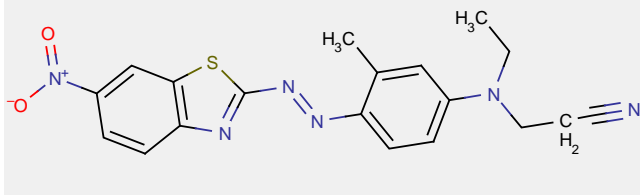
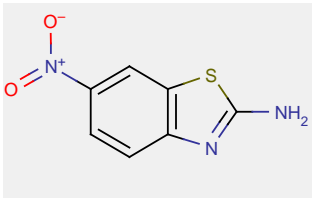
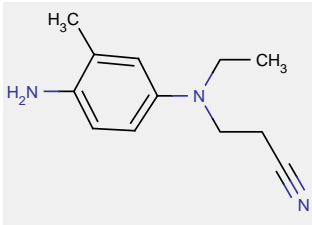
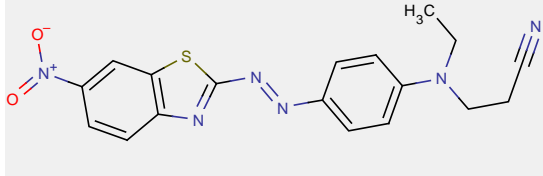
<sup>4</sup> CASETOX [Prediction module]. 2008. Version 2.0. Beachwood (OH): MultiCASE. [cited 2009 Sep 30]. Available from: <http://www.multicase.com/products/prod03.htm> [restricted access].

<sup>5</sup> TA100 strain of *Salmonella typhimurium*.

<sup>6</sup> *In vitro* test (in cultured Chinese hamster ovary cells).

<sup>7</sup> Weak negative.

## Appendix 6. Potential azo cleavage products and structural analogue of Disperse Red 179

Basis for consideration	CAS RN / name	Structure
Parent	16586-42-8 Disperse Red 179	
Metabolite 1 Potential azo cleavage product	6285-57-0 2-amino-6-nitrobenzothiazole	
Metabolite 2 Potential azo cleavage product	105294-34-6 propanenitrile, 3[(4-amino-3-methylphenyl)ethylamino]-	
Structural analogue <sup>1</sup>	25510-81-0 Disperse Red 145	

<sup>1</sup> Tanimoto score = 92 (SciFinder similarity search).



**Appendix 7 Q) SAR predictions for DAPEP (5,6-dichloro isomer of Disperse Red 153), 6,7-dichloro isomer of Disperse Red 153 and their potential azo cleavage products**

**Carcinogenicity predictions**

ID	CAS RN	Derek <sup>1</sup>	Toxtree <sup>2</sup>		Model Applier <sup>3</sup>				Casetox <sup>4</sup>			
		Cancer	SA gtx	Cancer	m-rat	f-rat	m-mice	f-mice	m-rat	f-rat	m-mice	f-mice
Parent 1 (DAPEP, 5,6'-dichloro isomer)	25176-89-0	P	P	N	P	N	ND	ND	ND	ND	ND	ND
Parent 2 (6,7'-dichloro isomer)	no CASRN	P	P	N	P	N	ND	ND	ND	ND	ND	ND
Metabolite 1 (for DAPEP, 5,6'-dichloro isomer)	24072-75-1	P	P	-	IC	IC	ND	ND	ND	N	N	P
Metabolite 2 (for 6,7'-dichloro isomer)	25150-27-0	P	P	-	IC	IC	ND	ND	ND	ND	N	N
Metabolite 3 (for both isomers)	100894-10-8	-	P	N	N	N	N	N	ND	ND	ND	ND

**Genotoxicity predictions**

ID	CAS RN	Ames				ChrAb		Micronuclei induction		
		Derek	TT <sup>5</sup>	MA	CT	MA	CT <sup>6</sup>	TT	MA	CT
Parent 1 (DAPEP, 5,6'-dichloro isomer)	25176-89-0	P	P	N	ND	ND	P	P	ND	ND
Parent 2 (6,7'-dichloro isomer)	no CASRN	P	P	N	ND	ND	ND	P	N	N
Metabolite 1 (for DAPEP, 5,6'-dichloro isomer)	25150-27-0	P	-	N	N	N	ND	P	IC	N
Metabolite 2 (for 6,7'-dichloro isomer)	105294-34-6	P	P	P	P	N	P	P	N	IC
Metabolite 3 (for both isomers)	100894-10-8	-	P	IC	ND	N	P	P	N	N

CAS RN, Chemical Abstracts Service Registry Number; ChrAb, chromosomal aberration; CT, Casetox; f, female; IC, inconclusive; ID, identification; m, male; MA, Model Applier; N, negative; ND, not in domain of model; SA gtx, structural alert for genotoxic carcinogen; P, positive; TT, Toxtree; -, no result

<sup>1</sup> [DEREK] Deductive Estimation of Risk from Existing Knowledge [Prediction module on CD ROM]. 2008. Version 10.0.2. Cambridge (MA): Harvard University, LHASA Group. [cited 2009 Sep 30]. Available from: [http://www.lhasalimited.org/index.php?cat=2&sub\\_cat=2#](http://www.lhasalimited.org/index.php?cat=2&sub_cat=2#) [restricted access].

<sup>2</sup> Toxtree version 1.60. 2009. Developed by Ideacon Ltd Bulgaria.

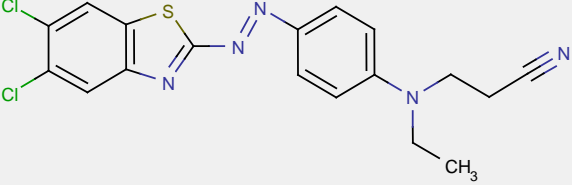
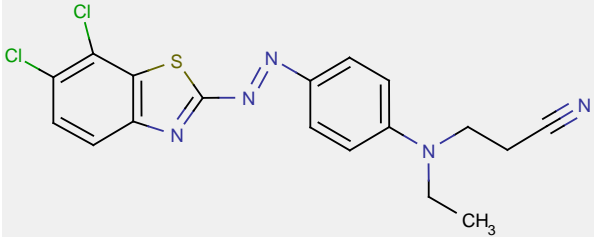
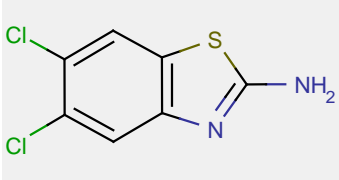
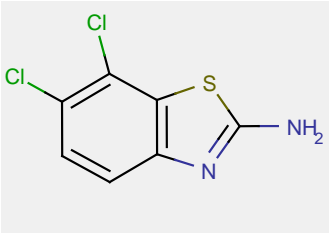
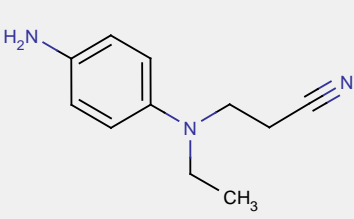
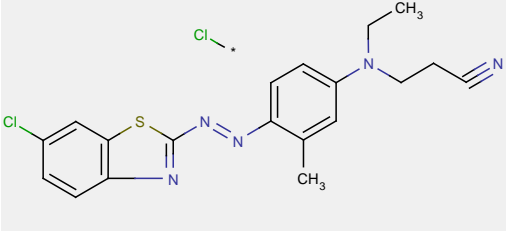
<sup>3</sup> [Leadscope] Leadscope Model Applier [Prediction module]. 2009. Version 1.2.0-3. Columbus (OH): Leadscope, Inc. [cited 2009 Sep 30]. Available from: [http://www.leadscope.com/all\\_products.php](http://www.leadscope.com/all_products.php) [restricted access].

<sup>4</sup> CASETOX [Prediction module]. 2008. Version 2.0. Beachwood (OH): MultiCASE. [cited 2009 Sep 30]. Available from: <http://www.multicase.com/products/prod03.htm> [restricted access].

<sup>5</sup> TA100 strain of *Salmonella typhimurium*.

<sup>6</sup> *In vitro* test (in cultured Chinese hamster ovary cells).

**Appendix 8. Structures for DAPEP (5,6'-dichloro isomer), the 6,7'-dichloro isomer, potential azo cleavage products, and an analogue Disperse Red 152 (mix of isomers)**

Basis for consideration	CAS RN / name	Structure
Parent 1 DAPEP (5,6'-dichloro isomer of Disperse Red 153)	25176-89-0	
Parent 2 6,7'-dichloro isomer of Disperse Red 153	no CASRN	
Metabolite 1 Potential azo cleavage product for DAPEP, the 5,6'-dichloro isomer of Disperse Red 153	24072-75-1 5,6-dichloro-2-benzothiazolamine	
Metablite 2 Potential azo cleavage product for the 6,7'-dichloro isomer of Disperse Red 153	25150-27-0 6,7-dichloro-2-benzothiazolamine	
Metabolite 3 Potential azo cleavage product for both the 5,6'- and 6,7'- isomers of Disperse Red 153	100894-10-8 Propanenitrile, 3-[(4-aminophenyl)ethylamino] -	
Structural analogue <sup>1</sup>	78564-86-0 Disperse Red 152 (mix of 5,6'- and 6,7'-dichloro isomers)	

<sup>1</sup> Tanimoto score = 90 (SciFinder similarity search).