Screening Assessment for the Challenge

2,7-Naphthalenedisulfonic acid, 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)-, disodium salt
(Direct Black 38)

Chemical Abstracts Service Registry Number
1937-37-7

Environment Canada
Health Canada

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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 2,7-naphthalenedisulfonic acid, 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)-, disodium salt (Direct Black 38), Chemical Abstracts Service Registry Number 1937-37-7

This substance was identified as a high priority for screening assessment and included in the Challenge because it was initially found to meet the ecological categorization criteria for persistence, bioaccumulation potential and inherent toxicity to non-human organisms and was believed to be in commerce in Canada.

Direct Black 38 was not considered to be a high priority 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 primarily on information relevant to the evaluation of ecological risks.

Direct Black 38 is a synthetic dye that is used as a colorant primarily in the textile industry. Direct Black 38 is typical of other direct dyes in that it exhibits a high affinity for cellulose fibres. Other applications include paper, leather, plastics, inks and wood. This chemical is not naturally produced in the environment. As a result of industry surveys conducted pursuant to section 71 of CEPA 1999, no companies reported manufacturing, importing or using Direct Black 38 in Canada above the reporting thresholds in 2005 or 2006. Therefore, reporting thresholds are used throughout this screening assessment as a conservative estimate of the quantity of this substance in commerce in Canada.

Based on reported use patterns and certain assumptions related to dyes in general, potential releases of Direct Black 38 to the Canadian environment during the formulation and consumer use of products containing this substance are estimated to be 15% to sewers and 85% transferred to waste disposal sites. Direct Black 38 is an azo dye with two sulfonic acid groups, which dictate its adsorption characteristics and impart high water solubility. Dyes have an inherently high affinity to substrates, and a potentially large proportion can be removed during sewage treatment as a result of such substances being adsorbed to sludge.

Information on other disulfonated acid dyes, as well as results of QSAR modelling, indicate that Direct Black 38 will persist in aerobic environments (water, soil, sediment). Degradation of Direct Black 38 under anaerobic or reducing conditions may occur relatively rapidly, but would be limited to specific environments (e.g., deep layers of sediments), with potentially harmful metabolites being formed as a result of cleavage of its azo bonds. However, in these situations exposure to aquatic organisms would be limited. The high water solubility of this substance, as well as other physical and chemical properties (e.g., large molecular size), indicates that it has a low potential to accumulate in the lipid tissues of organisms. Therefore, Direct Black 38 meets the persistence criteria but does not meet the bioaccumulation criteria as set out in the *Persistence and Bioaccumulation Regulations*. In addition, experimental toxicity data for Direct Black 38 and other disulfonated acid dyes suggest that this substance is not expected to cause acute harm to aquatic organisms at low concentrations.

For this screening assessment, a conservative exposure scenario was selected in which an industrial operation discharges this substance into the aquatic environment through a single sewage treatment plant. While there were no reports of Direct Black 38 being in commerce, the reporting threshold of 100 kg was used to conservatively estimate release and exposure levels. The predicted environmental concentration in water of this substance was below the predicted no-effect concentration for sensitive aquatic organisms, resulting in a risk quotient of much lower than one.

It is concluded that Direct Black 38 is not 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.

Direct Black 38 has been subjected to controls in other jurisdictions, based on concern for its hazardous properties, including carcinogenicity. Although the likely high hazard of Direct Black 38 is recognized, on the basis of information which indicates that it is not manufactured in or imported into Canada in amounts above the reporting threshold, the likelihood of exposure in Canada is considered to be low; hence the risk to human health is likewise considered to be low and it is concluded that it is a substance that is not entering the environment in a quantity or concentration or under conditions that constitute a danger in Canada to human life or health.

Based on available information, it is concluded that Direct Black 38 does not meet any of the criteria set out in section 64 of CEPA 1999.

Because this substance is listed on the *Domestic Substances List*, its import and manufacture in Canada are not subject to notification under subsection 81(1). Given the hazardous properties of this substance, 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 the substance so that new manufacture, import or use of this substance is notified and undergoes ecological and human health risk assessments.

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 substance 2,7-naphthalenedisulfonic acid, 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)-, disodium salt (which will be referred to as Direct Black 38 for the purposes of this document) was identified as a high priority for assessment of ecological risk as it met the ecological categorization criteria for persistence, bioaccumulation potential and inherent toxicity to aquatic organisms, and was believed to be in commerce in Canada. The Challenge for this substance was published in the *Canada Gazette* on May 31, 2008 (Canada 2008a,b). A substance profile was released at the same time. The substance profile presented the technical information available prior to December 2005 that formed the basis for categorization of this substance. As a result of the Challenge, no information was received pertaining to Direct Black 38.

Although Direct Black 38 was determined to be a high priority for assessment with respect to the environment, and it met the criteria for high hazard to human health based on classifications of other national or international agencies for carcinogenicity, genotoxicity, developmental toxicity or reproductive toxicity, it did not meet the criteria for GPE or IPE. Therefore, this assessment focuses principally on information relevant to the evaluation of ecological risk.

Screening assessments focus on information critical to determining whether a substance meets the criteria for defining a chemical as toxic as 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 any information submitted under the Challenge. Data relevant to the screening assessment of this substance was identified in original literature, review and assessment documents, stakeholder research reports and from recent literature searches, up to March 2009. Key studies were critically evaluated; modelling results may have been used to reach conclusions. When available and relevant, information presented in a hazard assessment from other jurisdictions was considered. This 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.

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. 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 assessment remain the responsibility of Health Canada and Environment Canada. The critical information and considerations upon which this assessment is based are summarized below.

Substance Identity

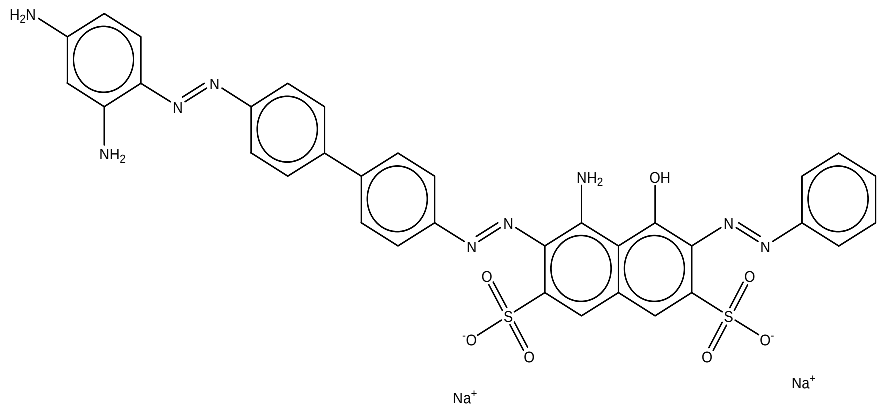
For the purposes of this document, 2,7-naphthalenedisulfonic acid, 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)-, disodium salt will be referred to as Direct Black 38, its Colour Index name (Colour Index Constitution Number: 30235; CII 2002). Information on this substance's identity is shown in Table 1 below.

Table 1. Substance identity for Direct Black 38

Chemical Abstracts Service Registry Number (CAS RN)	**1937-37-7**
DSL name	**2,7-Naphthalenedisulfonic acid, 4-amino-3-[[4'-[(2,4-**

	diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy -6-(phenylazo)-, disodium salt
National Chemical Inventories (NCI) names[1]	*2,7-Naphthalenedisulfonic acid, 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)-, disodium salt* (AICS, ASIA-PAC, DSL, PICCS, TSCA) *disodium 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)naphthalene-2,7-disulphonate* (EINECS) *Direct Black 38* (ENCS) *Disodium 4-amino-3-[[4'-[(2,4-diaminophenyl)azo][1,1'-biphenyl]-4-yl]azo]-5-hydroxy-6-(phenylazo)-2,7-naphthalenedisulfonate* (ENCS) *C.I. Direct Black 38* (ECL) *1-AMINO-8-NAPHTHOL-36- DISULFONIC DIAZO COMPOUND* (PICCS) *C.I. DIRECT BLACK 38* (PICCS)
Other names	*Ahco Direct Black GX; Airedale Black ED; Aizen Direct Deep Black EH; Aizen Direct Deep Black GH; Aizen Direct Deep Black RH; Amanil Black GL; Amanil Black WD; Apomine Black GX; Atlantic Black BD; Atlantic Black C; Atlantic Black E; Atlantic Black EA; Atlantic Black GAC; Atlantic Black GG; Atlantic Black GXCW; Atlantic Black GXOO; Atlantic Black SD; Atul Direct Black E; Azine Deep Black EW; Azocard Black EW; Azomine Black EWO; Belamine Black GX; Bencidal Black E; Benzanil Black E; Benzo Deep Black E; Benzo Leather Black E; Benzoform Black BCN-CF; Black 2EMBL; Black 4EMBL; Brasilamina Black GN; Brilliant Chrome Leather Black H; C.I. 30235; C.I. Direct Black 38, disodium salt; Calcomine Black; Calcomine Black EXL; Carbide Black E; Chloramine Black C; Chloramine Black EC; Chloramine Black ERT; Chloramine Black EX; Chloramine Black EXR; Chloramine Black XO; Chloramine Carbon Black S; Chloramine Carbon Black SJ; Chloramine Carbon Black SN; Chlorazol Black E; Chlorazol Black EA; Chlorazol Black EN; Chlorazol Burl Black E; Chlorazol Leather Black ENP; Chlorazol Silk Black G; Chlorozol Black E; Chrome Leather Black E; Chrome Leather Black EC; Chrome Leather Black EM; Chrome Leather Black G; Chrome Leather Brilliant Black ER; Coir Deep Black; Columbia Black EP; Coranil Direct Black F; Diacotton Deep Black; Diacotton Deep Black RX; Diamine Deep Black EC; Diamine DirectBlack E; Diaphtamine Black V; Diazine Black E; Diazine Direct Black E; Diazine Direct Black G; Diazol Black 2V; Diphenyl Deep Black G; Direct Black A; Direct Black BRN; Direct Black CX; Direct Black CXR; Direct Black E; Direct Black EW; Direct Black EX; Direct Black FR; Direct Black GAC; Direct Black GW; Direct Black GX; Direct Black GXR; Direct Black JET; Direct Black Meta;*

	Direct Black methyl; Direct Black N; Direct Black RX; Direct Black SD; Direct Black WS; Direct Black Z; Direct Black ZSh; Direct Deep Black E; Direct Deep Black E Extra; Direct Deep Black E-EX; Direct Deep Black EA-CF; Direct Deep Black EAC; Direct Deep Black EW; Direct Deep Black EX; Direct Deep Black WX; Enianil Black CN; Erie Black B; Erie Black BF; Erie Black GAC; Erie Black GXOO; Erie Black JET; Erie Black NUG; Erie Black RXOO; Erie Brilliant Black S; Erie Fibre Black VP; Fenamin Black E; Fibre Black VF; Fixanol Black E; Formaline Black C; Formic Black C; Formic Black CW; Formic Black EA; Formic Black MTG; Formic Black TG; Hispamin Black EF; Interchem Direct Black Z; Kayaku Direct Deep Black EX; Kayaku Direct Deep Black GX; Kayaku Direct Deep Black S; Kayaku Direct Leather Black EX; Kayaku Direct Special Black AAX; Lurazol Black BA; Meta Black; Mitsui Direct Black EX; Mitsui Direct Black GX; Nippon Deep Black; Nippon Deep Black GX; Paper Black BA; Paper Black T; Paper Deep Black C; Paramine Black B; Paramine Black E; Peeramine Black E; Peeramine Black GXOO; Phenamine Black BCN-CF; Phenamine Black CL; Phenamine Black E; Phenamine Black E 200; Pheno Black EP; Pheno Black SGN; Pontamine Black E; Pontamine Black EBN; Sandopel Black EX; Seristan Black B; Telon Fast Black E; Tertrodirect Black E; Tertrodirect Black EFD; Tetrazo Deep Black G; Union Black EM; Vondacel Black N
Chemical group (DSL Stream)	Discrete organics
Major chemical class or use	Trisazo dyes
Major chemical sub-class	Aromatic amines, sulfonaphthalene, naphthols
Chemical formula	$C_{34}H_{25}N_9Na_2O_7S_2$

Chemical structure[2]	
SMILES[3]	Nc6ccc(N=Nc1ccc(cc1)c5ccc(N=Nc4c(N)c3c(O)c(N=Nc2ccccc2)c(cc3cc4S(O-Na)(=O)=O)S(O-Na)(=O)=O)cc5)c(N)c6
Molecular mass	781.7 g/mol dry powder at room temperature

1 National Chemical Inventories (NCI). 2007: 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); ENCS (Japanese Existing and New Chemical Substances); PICCS (Philippine Inventory of Chemicals and Chemical Substances); and TSCA (Toxic Substances Control Act Chemical Substance Inventory).

2 Source: chemBlink (2009)

3 Simplified Molecular Line Input Entry System

Physical and Chemical Properties

Direct Black 38 is an anionic azo dye. The azo bond (–N=N–) is the part of the molecule that produces colour (Danish EPA 1999). Direct Black 38 also contains two sulfonic acid groups, the presence of which contributes to its high water solubility. In addition to chemical structure, dyes may be classified according to their industrial applications and the methods by which they are applied to the substrate of interest (e.g. direct dyes) (ETAD 1995). This classification system tends to reflect groupings based on physical and chemical behaviour. A brief discussion of the uses of this dye can be found later in this document under the Uses section.

Commercially available dyes are formulated to contain additional chemicals to maintain the desired properties of the dye and to ensure their effectiveness in the dyeing process. Powdered dyes (like Direct Black 38) require de-dusting agents (e.g., hydrocarbon oils, polyalkylene glycol ethers), and commonly contain diluents to standardize dye strength, wetting agents and bacteriostats (ETAD 1995). The content of the active dye can therefore vary among formulations and is often confidential business information. Most sulfonated dyes are not produced or sold in pure form (ETAD 1995), which is likely one of the reasons why physical and chemical data on these substances are lacking.

Few experimental data on the physical and chemical properties of Direct Black 38 are available. At the Environment Canada-sponsored Quantitative Structure-Activity Relationship (QSAR) Workshop in 1999, invited modelling experts identified many structural classes of pigments and dyes as “difficult to model” using QSARs (Environment Canada 2000). The physical and chemical properties of many of the structural classes of pigments and 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 pigments and dyes are reviewed on a case-by-case basis.

Environment Canada has considered it generally inappropriate to use QSAR models to predict the physical and chemical properties of Direct Black 38. Consequently, a read-across approach has been used to estimate physical and chemical properties of Direct Black 38 based on similar acid dyes with available experimental data. Due to the paucity of information, acceptable analogous substances for the purposes of this assessment were chosen. These substances include Acid Red 111 and three anthraquinone dyes previously assessed under the Challenge. These substances all have two acid groups present (i.e., sulfonic acid) and they are relatively large molecules. Acid Red 111 is also an azo dye. They are therefore expected to have similar behaviour in the environment and similar toxicity to Direct Black 38. These substances also have available experimental data that could be used directly or with adjustment as an estimate of that parameter value for Direct Black 38.

Table 2 shows available information on the physical and chemical properties of Direct Black 38 and relevant analogous substances identified. These properties were subsequently used for further modelling and lines of evidence in this assessment.

Table 2. Physical and chemical properties for Direct Black 38 and its relevant analogues

Chemical	Type	Value	Temperature (°C)	Reference
Physical state				
Direct Black 38	Grey-black powder		25	NIEHS 2008
Acid Red 111[1]	Red powder		25	Study Submission 2007a
Decomposition (ºC)				
Acid Red 111[1]	Experimental	170–190		Study Submission 2007a
Acid Violet 48[1]	Experimental	> 200		MSDS 2003a
Acid Blue 80[1]	Experimental	> 190		MSDS 2006
Azo dyes	Read-across[2]	> 300		ETAD 1995
Density (kg/m^3)				
Acid Red 111[1]	Experimental, (bulk)	390		Study Submission 2007a
Henry's Law constant ($Pa{\cdot}m^3/mol$)				
Acid Violet 48	Calculated[3]	$\sim 1.2 \times 10^{-11}$	25	HENRYWIN 2000
Acid Blue 127	Calculated[3]	8.4×10^{-15}	25	HENRYWIN 2000
Log K_{ow} (Octanol-water partition coefficient) (dimensionless)				
Azo dyes	Read-across[2]	< 3	25	ETAD 1995
Water solubility (mg/L)				
Direct Black 38	Experimental	93 000	15–25	Isik and Sponza 2004
Acid Red 111[1]	Experimental	25 000	80	Study Submission 2007a
	Experimental	65 000		MSDS 2003b
	Experimental	20 000	100	Rosi Chemical 2000
Acid Violet 48[1]	Experimental	90 000	90	MSDS 2003a
	Experimental	~ 60 000	25	Clariant 2007
Acid Blue 80[1]	Experimental	10 000	25	MSDS 2006
Azo dyes	Read-across[2]	Readily soluble		ETAD 1995

[1] These physical and chemical properties are for the formulated product identified in the company's Material Safety Data Sheet (MSDS).

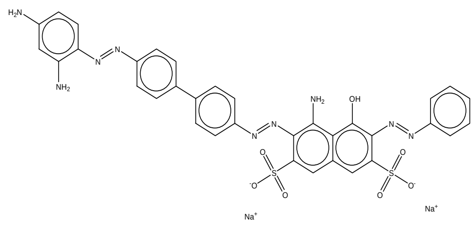
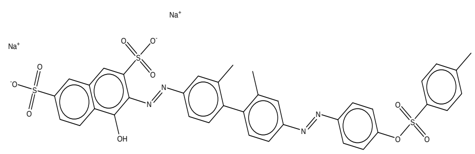
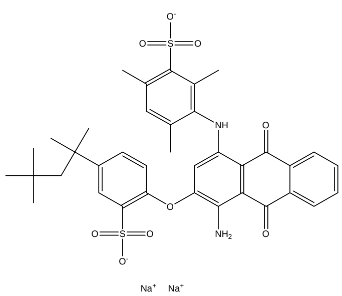
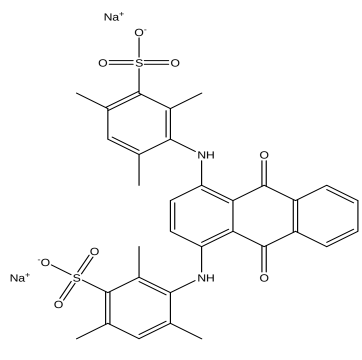
[2] The extrapolated values used for substances in the disulfonated acid dyes group are based on evidence on disulfonated acid dyes submitted to Environment Canada under the *New Substance Notification Regulations* and/or available evidence from other disulfonated acid dye analogues (e.g., ETAD 1995).

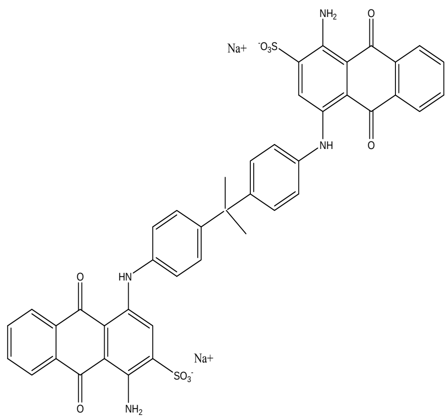
[3] Calculated using the following physical and chemical properties: water solubility (WS), vapour pressure (VP) and molecular weight (MW), using the following formula: (VP/WS)MW. For Acid Violet 48, water solubility at 60 000 mg/L and upper range limit of 10^{-8} Pa for vapour pressure were used in the calculation. For Acid Blue 127, water solubility at 10 000 mg/L and upper range limit of 10^{-8} Pa for vapour pressure were used in the calculation.

In general, sulfonated pure dyes whose properties have been studied have high melting points (usually above the 250–300°C range), whereas powder dyes have slightly lower melting points (> 200°C). In both cases, the substances decompose (char) at these levels (ETAD 1995). Therefore, a determination of boiling point is not meaningful for Direct Black 38.

Structural information for Direct Black 38 is presented in Table 3 along with that of other disulfonated acid dyes that are considered to be acceptable analogues for the purposes of this assessment. The molecular mass for these substances ranges from 678.69 to 872.83 g/mol. The cross-sectional diameters range from a minimum of 0.54–3.03 nm to a maximum of 2.01–3.53 nm. Available empirical data for these analogous substances (e.g., toxicity) are used in the weight of evidence to support the ecological assessment of Direct Black 38.

Table 3. Structural data for Direct Black 38 and selected **analogues**

Common name (CAS RN)	**Structure of analogue**	**Molecular mass (g/mol)**	**Cross-sectional diameter (nm)[1]**
Direct Black 38 (1937-37-7)		781.7	2.22–3.25
Acid Red 111 (6358-57-2)		830.82	3.03–3.53
Acid Violet 48 (72243-90-4)		764.82	0.54–2.04
Acid Blue 80 (4474-24-2)		678.69	1.67–2.01

Common name (CAS RN)	Structure of analogue	Molecular mass (g/mol)	Cross-sectional diameter (nm)[1]
Acid Blue 127 (6471-01-8)		872.83	0.54–2.8

[1] Minimum and maximum cross-sectional diameters. Model predictions using CPOPs (2008).

Direct Black 38 is a large, highly soluble disulfonated acid dye. Direct Black 38 contains three azo groups in its molecule, whereas the analogue Acid Red 111 contains only two azo groups. The anthraquinone dyes have a component triple ring structure with two oxygen atoms attached, and are not azo compounds. They are, however, disulfonated acids. The presence of sulfonic acid groups contributes to their solubility, which influences their environmental fate and toxicological properties. Molecular weights of all these substances are reasonably similar and water solubilities appear to be within an order of magnitude. The two azo dyes appear to be relatively larger than these anthraquinone dyes.

Sources

Direct Black 38 is 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, 2008b). These notices required submission of data on the Canadian manufacture and import of this substance. In the notice for 2006, data were also required on use quantities of Direct Black 38. In association with the section 71 notices for 2005 and 2006, companies that did not meet the mandatory reporting requirements but that had an interest in this substance were invited to identify themselves as stakeholders.

No companies reported importing or manufacturing Direct Black 38 above the 100-kg/year threshold in 2005 or 2006. No companies reported using Direct Black 38 above the 1000-kg/year threshold in 2006. In total, two companies reported a stakeholder interest in Direct Black 38 (Environment Canada 2006, 2008a).

During the development of the Domestic Substances List (DSL), 1 200 kg of Direct Black 38 were reported as being manufactured, imported or in commerce in Canada in 1986 (Environment Canada 1988). Total production of Direct Black 38 in the United States was reported as 1 705 507 kg (or 3 760 000 pounds) in 1976 and 373 307 kg (or 823 000 pounds) in 1978 (US DHHS 1980). The production in 1978 represented approximately 48% of the total production of benzidine-based dyes in the United States for that year (US DHHS 2005). Imports into the United States of Direct Black 38 were 32 093 kg (or 70 753 pounds) in 1976 and 77 311 kg (or 170 442 pounds) in 1978.

Manufacturers began phasing out the use of benzidine-based dyes in the mid- to late 1970s and replacing them with other dyes due to human health concerns. Direct Black 38 has been identified as a low production volume (LPV) chemical by the European Union, with production estimated between 10 000 and 1 000 000 kg per year (ESIS 2008). Direct Black 38 was also used in Denmark between 2002 and 2005 (SPIN 2008).

Products containing Direct Black 38 may enter Canada even if they are not identified in the section 71 survey as containing this substance, because they may be imported unknowingly in manufactured items, or may be in commerce in quantities below the 100-kg reporting threshold for the survey. Given the use of this dye in other countries, it is possible that it is entering the Canadian market as a component of manufactured items and consumer products.

Uses

Direct dyes, because of their larger and planar structure, exhibit high affinity for cellulose fibres and are applied directly to cellulose-containing materials in a neutral dye bath (ETAD 1995; Hunger 2003). They are used for dyeing cotton, rayon, and to a lesser extent wool, silk and nylon. Direct Black 38 may also be used in applications for paper, leather, plastics, ink and wood (CII 2002; ETAD 1995; NLM 2006). Direct Black 38 has also been used in hair dyes (NLM 2006).

Direct Black 38 is a benzidine-based dye. There have been no reports of its current use in Canada, and there are restrictions on the use of Direct Black 38 and other benzidine-based dyes in textiles in some jurisdictions (Danish EPA 1998; International Oeko-Tex® Association 2009). Additionally, an EU directive states that substances and preparations containing benzidine-based azo dyes should not be placed on the market for use by the general public (European Union 1999), and the handling of Direct Black 38 and 41 other benzidine-based dyes has been prohibited since 1993 in India, with an additional 70 azo dyes being prohibited since 1997 (India Textiles Committee 2009). Benzidine is listed on Schedule 1 (List of Toxic Substances) under CEPA 1999 and is regulated under the *Prohibition of Certain Toxic Substances Regulations, 2005* (Canada 1993, 2005; Environment Canada 2007a).

No information on current Canadian uses was collected for Direct Black 38 through the CEPA 1999 section 71 surveys (Canada 2006b; 2008b). Use codes identified during the DSL nomination (1984–1986) included colorant (pigment/stain/dye/ink) and specialty organic chemicals (Environment Canada 1988).

Releases to the Environment

Direct Black 38 is not reported to occur naturally in the environment nor are any releases reported as part of Environment Canada's National Pollutant Release Inventory.

Mass Flow

To estimate potential releases of this substance to the environment at different stages of its life cycle, a Mass Flow Tool was developed (Environment Canada 2008b). 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 of release to the different 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 and technical databases and documents. 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 Direct Black 38 to environmental media, chemical transformation during its life cycle and transfer to waste disposal sites, based on the Mass Flow Tool

Fate	Proportion of the mass (%)[1]	Major life cycle stage involved[2]
Released to receiving media:		
To soil	0.0	
To air	0.0	
To sewer[3]	14.6	Formulation, consumer use
Chemically transformed	0.0	
Transferred to waste disposal sites (e.g., landfill, incineration)	85.4	Waste disposal

[1] Information from the following OECD emission scenario documents was used to estimate releases to the environment for which the distribution of release of this substance is summarized above: textile manufacturing wool mills (OECD 2004); adhesive formulation (OECD 2007). Values presented for release to environmental media do not account for possible mitigation measures that may be in place at some locations. Specific assumptions used in the derivation of these estimates are summarized in Environment Canada (2008c).
[2] Potential applicable stage(s): production, formulation, industrial use, consumer use, service life of article/product, waste disposal.
[3] Wastewater before any form of treatment.

Results summarized in Table 4 indicate that potential releases of Direct Black 38 related to formulation and consumer use (i.e., the service life of products containing the substance) are estimated to be 15% to sewers, while 85% is estimated to be transferred to waste disposal sites (Environment Canada 2008c). The Mass Flow Tool does not quantify releases from a landfill and does not consider the effectiveness of sewage treatment. The industrial use scenario assumes that this substance is used as a dyeing agent for textiles. Various assumptions have been made in estimating releases to sewers and include residues from container handling (0.3%), process emissions (none to air), the dye fixation level (87%), the cleaning of transfer lines and vessels, and potential releases associated with consumer use during laundering of articles. Releases could occur to sewer water during the dyeing of textiles at the manufacturing site, which would be considered a non-dispersive use. Releases to sewer water from laundering of articles during the life of the textile would be considered a widely dispersive use.

Environmental Fate

Dyes have an inherently high affinity to substrates, with fixation levels ranging from 64 to 98% for direct dyes and acid dyes (with more than one sulfonic acid group) (ETAD 1995). With respect to wastewater treatment, most of the adsorption/desorption research on dyes has been done using activated sewage sludge or carbon (ETAD 1995), with dyestuffs generally being adsorbed to the extent of 40–80% (Clarke and Anliker 1980). Although dyes are not easily biodegraded aerobically, combined removal through biodegradation and adsorption on waste treatment sludge has been shown to exceed 95% in some cases (ETAD 1995). Some portion of dyes may still end up in the aquatic environment depending on their chemical structure and the properties of the dye product (Danish EPA 1999).

Direct Black 38 may be released to sewer water, as well as transferred to waste disposal sites (as described previously using the Mass Flow Tool). If released to the aquatic environment, this substance's adsorption characteristics would be dictated by its two sulfonic acid groups, which also impart high water solubility. Other factors such as increasing molecular size, hardness of the water and salinity, as well as decreasing pH, are thought to favour some sorption of azo dyes to suspended solids (Danish EPA 1999; NLM 2006). Acid dyes have a high fixation rate with positively charged substrates and can adsorb to positively charged particulates (e.g., nitrogen-containing particles) and have the potential to settle out to bed sediments or wastewater treatment plant sludge (ETAD 1995). Razo-Flores et al. (1997) suggest that due to the recalcitrant nature of azo dyes in aerobic environments, they eventually end up in anaerobic sediments, shallow aquifers and groundwater.

Sludge from wastewater treatment plants would contain dyes as a result of the removal processes (ETAD 1995), and dyes may end up being indirectly applied to soil through sludge enrichment or deposited in landfills. In soil, the ionic nature of these dyes may result in ion-exchange processes with clay that would retard leaching (NLM 2006). Volatilization from dry or moist soil surfaces is expected to be an unimportant fate process based upon the low estimated vapour pressures for azo dyes in general (Danish EPA 1999; NLM 2006).

Given the intended use in aqueous-based treatments, Direct Black 38 is not expected to be released to air and is not expected to partition to this compartment, based on very low modelled Henry's Law constants for other acid dyes (e.g., Acid Violet 48: ~1.2×10^{-11} $Pa \cdot m^3/mol$). Moreover, air is not considered to be a carrying medium for dyes (including acid dyes), as these substances exhibit low or negligible volatility (Brown and Hamburger 1987; Danish EPA 1999; ETAD 1995).

The environmental persistence and potential for bioaccumulation of Direct Black 38 are briefly discussed in the following sections.

Persistence and Bioaccumulation Potential

Environmental Persistence

Dyes must have a high degree of chemical and photolytic stability in order to be useful, so most are generally considered non-degradable under environmentally relevant aerobic conditions (Danish EPA 1999; ETAD 1995). Studies applying commonly accepted screening tests (e.g., OECD guidelines) for ready and inherent biodegradability have confirmed this point (ETAD 1992; Pagga and Brown 1986). Abiotic degradation, including photolysis and hydrolysis, is not thought to play a significant role in the environmental fate of azo dyes (Danish EPA 1999), although one study showed strongly accelerated photo decomposition of azo dyes in the presence of natural humic materials (Brown and Anliker 1988).

Biotic degradation of azo dyes may take place relatively rapidly under anaerobic or reducing conditions (Baughman and Weber 1994; Danish EPA 1999; ETAD 1995; Isik and Sponza 2004; Yen et al. 1991). Permeability through the bacterial cell wall has been found to be the rate-limiting step in the reduction process (Danish EPA 1999). Azo dyes have a high tendency to cleave at the azo bond with the formation of aromatic amines (Danish EPA 1999; Hunger 2005). The carcinogenic potential of aromatic amines varies considerably with molecular structure, with carcinogenic breakdown products being associated with the moieties of benzidine, aniline, toluene or naphthalene. There is concern that biodegradation products of Direct Black 38 include benzidine and potentially other mutagenic and carcinogenic substances (Isik and Sponza 2004; NLM 2006). However, the formation of such metabolites in deep anoxic sediments would typically not result in exposure to aquatic organisms. Total mineralization or further degradation of these metabolites could take place if they are transferred (e.g., by sediment resuspension) to aerobic environments (Danish EPA 1999; Isik and Sponza 2004). Aromatic amines may also be present as impurities in commercially available azo dyes, although the metabolic cleavage of azo dyes is the main source of these compounds (Danish EPA 1999).

There are no empirical data on the persistence of Direct Black 38 under environmentally relevant conditions. There are, however, studies that evaluate the effectiveness of various sequential systems designed to treat wastewaters, which focus on the decolourization and reduction in toxicity of wastewater effluent (such as for Direct Black 38: Bafana et al. 2009; Danish EPA 1999; Isik and Sponza 2004).

Persistence was thus examined using predictive QSAR models for biodegradation. These models are considered acceptable for use in this situation as they are based on chemical structure, and the trisazo structure is represented in the training sets of all the BIOWIN models used, thereby increasing the reliability of the predictions. Given the ecological importance of the water compartment, the fact that most of the available models apply to water and the fact that Direct Black 38 is expected to be released to this compartment, persistence in water was primarily examined. Direct Black 38 does not contain functional groups expected to undergo hydrolysis.

Table 4 summarizes the results of available QSAR models for biodegradation of Direct Black 38 in water.

Table 4. Modelled data for biodegradation of Direct Black 38

Fate process	Model and model basis	Model output	Expected half-life (days)
WATER			
Primary biodegradation			
Biodegradation (aerobic)	BIOWIN 2000[1] Sub-model 4: Expert Survey (qualitative results)	2.39 (biodegrades somewhat slowly)	< 182
Ultimate biodegradation			
Biodegradation (aerobic)	BIOWIN 2000[1] Sub-model 3: Expert Survey (qualitative results)	0.63 (biodegrades very slowly)	> 182
Biodegradation (aerobic)	BIOWIN 2000[1] Sub-model 5: MITI linear probability	-1.84 (biodegrades very slowly)	> 182
Biodegradation (aerobic)	BIOWIN 2000[1] Sub-model 6: MITI non-linear probability	0.0 (biodegrades very slowly)	> 182
Biodegradation (aerobic)	TOPKAT 2004 Probability	0.0 (biodegrades very slowly)	> 182
Biodegradation (aerobic)	CATABOL c2004–2008 % BOD[2]	0 (biodegrades very slowly)	> 182

1 EPIsuite (2007); model run using SMILES from Table 1.
2 Biological oxygen demand

The results in Table 4 reveal that the majority of aerobic biodegradation models (i.e., the ultimate biodegradation models BIOWIN 3, 5, 6, TOPKAT and CATABOL) indicate this dye biodegrades very slowly, if at all. The half-life results from the primary survey model (BIOWIN 4) equates to “weeks to months,” indicating that this substance undergoes a somewhat slow rate of primary biodegradation but that the half-life is expected to be less than 182 days.

When the overall results of the modelled data are considered, the weight of evidence (for models predicting ultimate biodegradation) indicates that the aerobic biodegradation half-life of this substance in water is ≥ 182 days. This finding is consistent with what would be expected for this chemical structure (i.e., few degradable functional groups, solid particle) and its intended use as a dye.

Using an extrapolation ratio of 1:1:4 for a water:soil:sediment biodegradation half-life (Boethling et al. 1995), the ultimate degradation half-life in aerobic soil is expected to be ≥ 182 days and the half-life in aerobic sediment is expected to be ≥ 365 days. As mentioned previously, experimental evidence indicates that this dye may not persist in the deeper, anoxic layers of sediments where azo bonds are readily reduced under anaerobic conditions.

Based on the weight of evidence provided by the available literature on azo dyes and modelled data described above (see Table 4), Direct Black 38 meets the persistence criteria in water, soil and sediment (half-lives in soil and water ≥ 182 days and half-life in sediment ≥ 365 days) as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

Potential for Bioaccumulation

In this assessment, a variety of lines of evidence have been used to determine the bioaccumulation potential of Direct Black 38, which is different from the K_{ow}-based QSAR approach used during categorization. As indicated in Table 2, this substance has a high water solubility (93 000 mg/L) and read-across data for azo dyes suggests relatively low log K_{ow} values (< 3.0), which would indicate low bioaccumulation potential. Ionic dyes (including acid and direct dyes) are generally considered to have a very low bioaccumulation potential based on results of studies with various dyes (ETAD 1995).

Estimated and experimental log K_{ow} values were compared with experimental bioconcentration factors (BCFs) for fish for a number of dyes (Anliker et al. 1981; Danish EPA 1999; ETAD 1995). With respect to the data for 6 acid dyes and one direct dye (only dye classes were reported), reported BCFs were less than 10, indicating that these very hydrophilic (ionic) dyes are not likely to bioconcentrate or bioaccumulate in aquatic organisms.

There are no empirical bioaccumulation data available for Direct Black 38. Therefore, available data on water solubility, molecular weight and cross-sectional diameter have also been considered (including those for the analogues chosen) in order to determine the bioaccumulation potential of Direct Black 38. Given this substance’s high water solubility, ionic nature and high degree of dissociation under typical environmental conditions, its lipid partitioning tendency is expected to be limited.

Direct Black 38 is a relatively large molecule with a high molecular weight (781.7 g/mol), similar to the other analogues chosen. The minimum and maximum cross-sectional diameter for Direct Black 38 ranges from 2.22 to 3.25 nm. These characteristics suggest a low bioaccumulation potential for this substance. A report prepared for Environment Canada (Environment Canada 2007b) points out that there are no clear relationships for establishing strict molecular size cut-offs for assessing bioaccumulation potential. However, the report does not dispute the notion that a reduction in uptake rate can be associated with increasing cross-sectional diameter as demonstrated by Dimitrov et al. (2002, 2005). ETAD (1995) has stated that substances with a molecular weight of > 450 g/mol and a cross-sectional diameter of > 1.05 nm have a low potential for bioaccumulation. Recent investigations (BBM 2008; Dimitrov et al. 2002, 2005) suggest 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 lowers appreciably when cross-sectional diameter is > ~1.5 nm and more significantly for molecules having a cross-sectional diameter of > 1.7 nm. Sakuratani et al. (2008) have also investigated the effect of cross-sectional diameter on passive diffusion from a test set of about 1200 new and existing chemicals. They observed that substances having a low bioconcentration potential often have a D_{max} > 2.0 nm and an effective diameter (D_{eff}) > 1.1 nm.

Direct Black 38 is expected to have a low bioaccumulation potential due to its physical and chemical properties (i.e., high molecular weight, large cross-sectional diameter, high water solubility), the physical and chemical properties of the relevant analogues (disulfonated dyes) used in this assessment (i.e., high molecular weights, high decomposition points, relatively large cross-sectional diameters, high water solubilities), and limited information regarding experimental log K_{ow} values and low experimental BCFs for some other acid and direct dyes. Therefore, considering the available evidence, Direct Black 38 does not meet the bioaccumulation criteria (BCF, BAF ≥ 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

A variety of lines of evidence have been used to determine the toxicological potential of Direct Black 38, which is different from the primarily K_{ow}-based QSAR approach used during categorization. The available data to support this assessment are discussed below.

One empirical toxicity study was available for Direct Black 38. Static bioassays were performed to evaluate the acute toxicity of various dyes (47 in total) to fathead minnow (*Pimephales promelas*) (Little and Lamb 1972). Bioassays were carried out according to published standard methods and data sheets for each test were prepared, including pertinent information on the test organisms, dilution water and test conditions. The experiment was designed to estimate the TL_{50}, the concentration at which 50% of the experimental animals survived after 96 hours. However, no mortality was observed in fathead minnows exposed to Direct Black 38 at the highest concentration tested (180 mg/L).

Although empirical ecotoxicological data for Direct Black 38 is very limited, predictions for ecotoxicological data using QSAR models are considered unreliable, as anionic dye classes are difficult to model because the properties of these dyes fall outside the domains of applicability of the available models. Consequently, data for the acceptable analogues of Acid Red 111 and two anthraquinone dyes were also considered. These data are summarized in Table 6 below.

Table 6. Empirical data for aquatic toxicity of relevant analogues

Test organism	Type of test	Endpoint	Value (mg/L)	Reference
Acid Red 111 (6358-57-2)				
Rainbow trout (*Oncorhynchus mykiss*)	Acute (48 hours)	LC_{50}	~11	Study Submission 2007a
Acid Blue 80 (4474-24-2)				
Rainbow trout (*Oncorhynchus mykiss*)	Acute (48 hours)	LC_{50}	75	Study Submission 2007b
Acid Violet 48 (72243-90-4)				
Rainbow trout (*Oncorhynchus mykiss*)	Acute (48 hours)	LC_{50}	~33	MSDS 2003a
Rainbow trout (*Oncorhynchus mykiss*)	Acute (96 hours)	LC_{50}	> 10 and < 100	Study Submission 2008a
Bacteria (species not specified)	Respiratory Inhibition Test (OECD No. 209)	IC_{50}	> 100	Study Submission 2008b

LC_{50} – The concentration of a substance that is estimated to be lethal to 50% of the test organisms.
IC_{50} – The concentration of a substance that is estimated to cause inhibition of 50% of the test organisms.

Toxicological data were reported for a formulated product, Lanasyn Scarlet F-3GL 130, which contains Acid Red 111 (Study Submission 2007a). The information listed in the material safety data sheet included an acute LC_{50} (48 hours) of approximately 11 mg/L for rainbow trout (*Oncorhynchus mykiss*). Study details were not reported.

According to the data available for these analogous substances (Table 6), the lowest LC_{50} of > 10 mg/L was observed for rainbow trout in an acute toxicity study for Acid Violet 48 (Table 6). Experimental and methodological details were not available for either study on rainbow trout for Acid Violet 48, thus the data are considered to have limited reliability.

However, the results shown in Table 6 agree within an order of magnitude, and indicate that Direct Black 38 would have a moderate to low potential for toxicity.

In general, the above results are similar to what has been reported for acid and direct dyes in the literature (Danish EPA 1999). The Danish EPA summarized short-term test results for Zebra fish, *Daphnia magna*, algae and bacteria from a study by ETAD covering 47 dyes of different chemical dye classes (although specific dyes included in the study were not reported). The results for 96-hour LC_{50} tests on Zebra fish indicated that toxicity was observed for 2 acid dyes between 1 and 10 mg/L, for 3 acid dyes between 10 and 100 mg/L and 6 other acid dyes above 100 mg/L. For direct dyes, all 7 dyes tested reported toxicity for Zebra fish at levels > 100 mg/L. Effects were observed between 10 and 100 mg/L and above 100 mg/L in 48-hour EC_{50} tests (endpoint not specified) on *D. magna* for 9 acid dyes. Similar tests with direct dyes resulted in adverse effects to *D. magna* at levels > 100 mg/L. Algal toxicity (measured in 72-hour EC_{50} tests) was observed in 2 cases for acid dyes below 1 mg/L, as well as above this level (for the remaining 7 acid dyes), whereas toxicity results for the 7 direct dyes tested were all above 1 mg/L. Algae appeared to be the most susceptible organisms for all dye classes tested; the effect was thought to be related to light inhibition at high dye concentrations (coloration of water can occur above 1mg/L). Generally, bacteria were the least susceptible to most of the different classes of dyes tested, compared with the other organisms (IC_{50}s > 100 mg/L). In this study, it appeared that direct dyes are less toxic than the acid dyes.

As mentioned previously, cleavage of the azo bond under anaerobic or reducing conditions (e.g., deep layers of sediments) is known to result in aromatic amines, some of which are known to be potentially harmful. In the case of Direct Black 38, these include benzidine and 4-aminobiphenyl (4-ABP), which have been found to be mutagenic and carcinogenic substances (Bafana et al. 2009; Isik and Sponza 2004), and acutely toxic (LC_{50} < 1 mg/L) to some crustaceans and juvenile fish (Danish EPA 1999). However, because they are only formed in deep anoxic sediment, there is little likelihood that aquatic organisms are exposed to these more harmful metabolites.

Experience with over 200 acid dyes has led to the observation that the potential toxicity of such substances may generally be predicted by the number of acid groups present (US EPA 2002). Some monoacid and diacid dyes have shown high to moderate toxicity (i.e., acute values < 1 mg/L and < 100 mg/L, respectively) to fish and aquatic organisms. Dyes with three or more acid groups showed low toxicity (i.e., acute values > 100 mg/L) towards fish and invertebrates. All acid dyes showed moderate toxicity to green algae, with further analysis suggesting that such effects may have been related to shading. For these generalizations to be applicable, the acid dyes must have some water solubility and molecular weights generally need to be near or below 1000.

Furthermore, Environment Canada has evaluated numerous acid dyes under the *New Substances Notification Regulations* and has generally found anionic dyes to be of low toxicity regardless of the number of acid groups, but some exceptions have been found (e.g., when a reactive functional group is not hindered). Given these findings, Direct

Black 38, as an anionic dye with two sulfonic acid groups and reactive functional groups (i.e., aniline group), are expected to have moderate toxicity to aquatic organisms.

Given the information summarized above, Direct Black 38 likely has the potential for moderate toxicity (LC_{50}s > 10 mg/L).

B - In Other Environmental Compartments

No ecological effects studies were found for Direct Black 38 in media other than water. As such, effect levels for this substance have not been estimated for soil and sediment. However, this substance could end up in these media as a result of release to the aquatic environment, landfill disposal of sludge from wastewater treatment plants, disposal of products containing these substances, or sludge application to soils. Therefore, toxicity data for soil and sediment organisms would be desirable.

This being said, the toxicity potential is likely to be low in sediment- and soil-dwelling species, considering the low bioaccumulation potential and the physical and chemical properties of this substance. However, this cannot be substantiated due to the lack of suitable whole organism toxicity data.

Ecological Exposure Assessment

No information has been identified concerning the concentrations of Direct Black 38 in the Canadian environment (air, water, soil, sediment). A concentration in water of 0.002 mg/L of an acid red dye (unspecified) was reported in the Coosa River, USA (Danish EPA 1999).

Releases and losses associated with the formulation, consumer use and waste disposal of this dye has been estimated using the Mass Flow Tool (15% estimated release to sewer, 85% estimated transfer to waste disposal sites; see Table 4). It is expected that exposure to Direct Black 38 in the Canadian environment is very low, particularly given it is not in commerce above reporting thresholds. As this substance may be used in industrial processes and can be released to water, Environment Canada's Industrial Generic Exposure Tool (IGETA) for the aquatic environment was used to estimate a conservative concentration of this substance in a generic watercourse receiving industrial effluents (Environment Canada 2009a).

The generic scenario is designed to provide exposure estimates based on conservative assumptions regarding the amount of substance processed and released, the number of processing days, the sewage treatment plant (STP) removal rate, and the size of the receiving watercourse. This information is then used to calculate a predicted environmental concentration (PEC).

Direct Black 38 is not currently reported to be in commerce in Canada; however, the reporting threshold in Canada of 100 kg was conservatively assumed to be the quantity used industrially. Assumptions made in calculating the conservative PEC for this substance include a 15% release or loss during industrial processes (textile dying) over a period of 150 days (to represent seasonal activities), with no removal during sewage treatment and release into a relatively small receiving watercourse with a flow of 0.4 m^3/s. The equation and inputs are described in Environment Canada (2009a,b). Based on these assumptions, the PEC resulting from industrial releases of Direct Black 38 to water is 0.003 mg/L.

Environment Canada's Mega Flush model was also used to estimate down-the-drain releases from consumer uses and potential concentrations in multiple water bodies receiving sewage treatment plant effluents containing this substance released from consumer products (Environment Canada 2009c). The equation and inputs used in this consumer use release scenario are described in Environment Canada (2009d). A Mass Flow analysis was done to predict releases to water (sewers) from consumer use of products containing this substance (see section on Releases to the Environment). The use quantity as mentioned above for the industrial scenario was applied to this release scenario (i.e., 100 kg of Direct Black 38). Only the quantity remaining after the industrial stage in Canada, however, was considered to be present in consumer products (i.e., after considering 15.3% loss to sewers at the industrial stage). In addition, potential quantities in imported textiles in Canada were estimated assuming a 30:70 ratio for manufactured to imported textiles (Environment Canada 2008c). The total quantity considered for a consumer use scenario of textile dyes was therefore 281 kg. There was a 10% annual loss of textile dyes predicted to be released to water, as a result of loss to sewers during the laundering of manufactured articles that contain these dyes (Environment Canada (2009d). Other assumptions include consumer use of the substance over 365 days/year, and the 10th percentile value flow rate for receiving water bodies at all sites. Exposure concentrations were estimated for approximately 1000 release sites across Canada, which account for most of the major STPs in Canada. The resulting maximum PEC from Mega Flush was 4.3×10^{-5} mg/L.

Characterization of Ecological Risk

The approach taken in this ecological screening assessment was to examine available scientific information and develop conclusions based on a weight-of-evidence approach and precaution as required under section 76.1 of CEPA 1999.

Direct Black 38 is not present in Canadian commerce above the reporting thresholds, and therefore, potential releases of the substance to the Canadian environment are likely to be low.

Based on modelled biodegradation and information on similar substances, Direct Black 38 is predicted to persist in aerobic water, soil and sediment. Under anaerobic conditions (e.g., deep layers of sediment), this dye could undergo relatively rapid azo reduction to potentially hazardous metabolites, although exposure of aquatic organisms to these breakdown products would be limited and would likely present a low risk. Data on water solubility, molecular size and cross-sectional diameter, as well as empirical bioaccumulation data for some similar substances, indicate that Direct Black 38 has a low potential to bioaccumulate in aquatic organisms. Direct Black 38 is considered to have potentially moderate toxicity to aquatic organisms (LC_{50}s > 10 mg/L).

A risk quotient analysis, integrating a conservative predicted environmental concentration (PEC) with a conservative estimate of a predicted no-effect concentration (PNEC) was conducted for the aquatic environment and the resulting risk quotient (PEC/PNEC) was used as a key part of the weight of evidence regarding the potential to cause environmental harm.

A conservative PNEC was derived using the lowest available experimental toxicity value for relevant analogues, as experimental data available for Direct Black 38 were limited. The LC_{50} value of > 10 mg/L for an acute (96-hour) test on rainbow trout was reported for Acid Violet 48. This value was divided by an application factor of 100 to account for uncertainty regarding the necessary reliance on limited data from relevant analogues, potential differences in species sensitivities and to extrapolate from a laboratory-based measure of acute effects to a predicted chronic no-effect concentration in the field. The resulting PNEC calculated for Direct Black 38 was > 0.1 mg/L.

When compared to the PEC calculated above for industrial releases to water (0.003 mg/L), the resulting risk quotient (PEC/PNEC) is <0.03. Therefore, it is estimated that concentrations of Direct Black 38 in surface waters in Canada resulting from industrial point source releases do not harm aquatic organisms.

For exposure resulting from down-the-drain releases through consumer product use, Mega Flush results similarly indicate that the PEC for this substance will not exceed the PNEC at any site (i.e., all risk quotients < 1). This indicates that down-the-drain consumer releases of Direct Black 38 are not likely to harm aquatic organisms.

Overall, results of this assessment indicate that Direct Black 38 is unlikely to pose a risk to the Canadian environment.

Uncertainties in Evaluation of Ecological Risk

In general, Direct Black 38 is a data-poor substance. Information was available on the water solubility of this substance. Very little other physical and chemical data exist on this substance. As a result, a read-across approach using data from selected analogues was used to estimate physical and chemical properties, given that they too are disulfonated dyes.

This paucity of information necessitated the generation of model predictions for Direct Black 38 for biodegradation, and inference of bioaccumulation potential using available data on physical and chemical properties. Only one empirical toxicity study was available for Direct Black 38. Therefore, available acute toxicity data from relevant analogues were also used to assess the potential toxicity of Direct Black 38. Long-term (chronic) toxicity data would be beneficial in evaluating substances such as this one that are determined to be persistent in the environment. The use of an assessment factor in determining a predicted no-effect concentration is intended to address these uncertainties. The significance of soil and sediment as possible media of exposure was not well addressed by the effects data available. However, given the low quantity that may be in use in Canada, exposure of organisms to soils or sediments containing these substances is not likely to be significant.

The lack of information on environmental concentrations of this substance (e.g., monitoring data) in Canada resulted in the need to evaluate risk based on predicted concentrations in water near industrial point sources. Conservative assumptions were made when using models to estimate concentrations in such receiving water bodies. Conservative assumptions were also made in modelling down-the-drain releases resulting from consumer use of products containing this substance.

There is the possibility that Direct Black 38 may be in commerce in quantities below the 100-kg reporting threshold for the section 71 surveys. Because of this possibility, when modelling industrial exposure concentrations, it was conservatively assumed that 100 kg of Direct Black 38 are being used in industrial applications in Canada.

Potential to Cause Harm to Human Health

Direct Black 38 has been subjected to controls in other jurisdictions, based on concern for its hazardous properties, including carcinogenicity and genotoxicity. The International Agency for Research on Cancer (IARC) classified Direct Black 38 as Group 2A (probably carcinogenic to humans) (IARC 1987); more recently, IARC has upgraded the class of benzidine based dyes to Group 1 (carcinogenic to humans) on the basis that these substances are known to be metabolized by azoreductase mediated cleavage of the azolinkage of the dye to benzidine, a known human carcinogen (Baan et al. 2008).

However, based on the results of a survey under section 71 of CEPA 1999, Direct Black 38 is not believed to be manufactured in or imported into Canada in quantities above the reporting limits. Therefore, the likelihood of exposure in Canada is considered to be low; hence the risk to human health is likewise considered to be low.

Conclusion

Based on the information presented in this screening assessment, it is concluded that Direct Black 38 is not 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 the high hazard of Direct Black 38 is recognized, on the basis of information which indicates that it is not manufactured in or imported into Canada in amounts above the reporting threshold, it is concluded that it is a substance that is not entering the environment in a quantity or concentration or under conditions that constitute a danger in Canada to human life or health.

It is therefore concluded that Direct Black 38 does not meet the definition of toxic as set out in section 64 of CEPA 1999. Additionally, Direct Black 38 meets the criteria for persistence, but does not meet the criteria for bioaccumulation potential as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

Considerations for Follow-up

In light of the potential high hazard of substances which metabolize to aromatic amines that may be of concern for carcinogenicity, including these substances, additional activity (e.g., research, monitoring and surveillance, assessment) to characterize the risk to human health in Canada of this broader group will be undertaken.

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