Screening Assessment

Phenol, 2-methyl-4,6-dinitro-
(DNOC)

Chemical Abstracts Service Registry Number
534-52-1

Environment Canada
Health Canada

November 2009

Synopsis

Under the *Canadian Environmental Protection Act*, *1999* (CEPA 1999), the Ministers of the Environment and of Health have conducted a screening assessment of Phenol, 2-methyl-4,6-dinitro-, also known as 4,6-dinitro-*o*-cresol (DNOC), Chemical Abstracts Service Registry Number 534-52-1, which was selected as one of 123 substances on the Domestic Substances List for a pilot project for screening assessments.

DNOC is used predominantly in the plastics industry as an inhibitor of polymerization in styrene and vinyl products. Results from a section 71 *Notice with Respect to Certain Substances on the Domestic Substances List (DSL)* conducted for the year 2000 indicated that, although DNOC was not manufactured in Canada, 100 to 1000 tonnes were imported at that time. DNOC is included in the National Pollutant Release Inventory (NPRI), and facilities manufacturing, importing or otherwise using more than 10 tonnes per year of the substance must report their releases. The one company that reported to the NPRI for the years 1994 to 2002 ceased use of DNOC in late 2002. There have been no reports to the NPRI for this substance since 2003.

DNOC was detected in surface water and sewage sludges but not in sediment in Canada in the early 1980s. No more recent monitoring data for these media were identified. DNOC was not detected in rural, urban or agriculture soil from various locations across Canada. No Canadian air or groundwater monitoring data were identified.

It is believed that industrial uses of DNOC could result in releases of the substance to surface waters. A conservative scenario developed to account for potential releases from industrial process losses indicated a low potential for risk to aquatic organisms.

Scientific studies have shown that DNOC may form in air by reaction with reactive species such as OH and NO radicals, although the extent and mechanisms of formation are not well understood at present. A conservative scenario based on concentrations of DNOC in precipitation that could be expected to enter Canadian receiving water indicated that the potential for risk to aquatic organisms from this source is low.

Toxicity data for DNOC are available for microorganisms, bacteria and mammals, and for aquatic and terrestrial plants, invertebrates and vertebrates. No data on toxicity were identified for amphibians or marine organisms.

DNOC is persistent in water and air but is not bioaccumulative. The substance therefore meets the persistence criterion but does not meet the bioaccumulation criterion set out in the *Persistence and Bioaccumulation Regulations.* Modelling indicates that it is not likely to be transported over very long distances, and a decreasing concentration with increasing latitude is expected.

Based on available information, it is concluded that DNOC is not entering the environment in a quantity or concentration or under conditions that have or may have an immediate or long-term

effect on the environment or its biological diversity or that constitute or may constitute a danger to the environment on which life depends. Therefore, it is concluded that DNOC does not meet criteria set out in paragraphs 64*(a)* and 64*(b)* of the *Canadian Environmental Protection Act, 1999*.

Sources of human exposure to DNOC in Canada are likely to be limited to fugitive releases from industrial sites and the combustion of fossil fuels. There is no indication that DNOC is present in consumer products.

Comparison of a conservatively selected lowest effect level (i.e., 2.5 mg/kg-bw per day) for slight changes in biochemical parameters in a 90-day study in rats to the highest of the upper bounding estimates of exposure for all age groups in the population (i.e., 0.06 μg/kg-bw per day) for the 0- to 6-month (formula-fed) age group resulted in a margin of exposure of approximately 41 700. In light of the moderate to high confidence in the databases on exposure and effects upon which this assessment is based, this margin is considered adequate to address elements of uncertainty associated with limitations of the database for health effects and population exposure and intraspecies and interspecies variations in sensitivity, as well as the biological adversity or severity of the effects deemed critical.

The outcome of this screening health assessment is that DNOC does not meet the criterion set out in paragraph 64*(c)* of CEPA 1999—i.e., it is 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. This determination is based on the adequacy of the sufficiently health-protective margin between a conservatively selected lowest effect level and upper-bounding estimates of exposure of individuals in the general population.

Based on the information available for environmental and human health considerations, it is concluded that DNOC does not meet any of the criteria set out in section 64 of the *Canadian Environmental Protection Act, 1999*.

Introduction

This screening assessment report was conducted pursuant to section 74 of the *Canadian Environmental Protection Act, 1999* (CEPA 1999). This section of the Act requires that the Ministers of the Environment and of Health conduct screening assessments of substances that satisfy the categorization criteria set out in section 73 of the Act in order to determine of they meet or may meet the criteria set out in section 64 of the Act.

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 (Canada 1999). Screening assessments examine scientific information and develop conclusions by incorporating a weight-of-evidence approach and precaution.

A screening assessment was undertaken on Phenol, 2-methyl-4,6-dinitro-, also known as 4,6-dinitro-*o*-cresol (DNOC; CAS RN 534-52-1), on the basis that this compound was included in the Domestic Substances List (DSL) pilot project for screening assessment as a substance likely to be prioritized because it met the criteria for persistence and/or bioaccumulation and inherent toxicity to non-human organisms and as a substance likely to be prioritized on the basis of greatest potential for human exposure.

Owing to the chemical nature of DNOC, it readily forms water-soluble sodium, potassium and ammonium salts, and virtually 100% of dissolved DNOC will be in the ionized form at environmentally relevant pHs (pH 6–8). Based on this information, Environment Canada reviewed the use of DNOC salts to determine if they should also be included in this screening assessment. The sodium, potassium and ammonium salts of DNOC are not on the Domestic Substances List (DSL), although DNOC sodium salt is on the Non-Domestic Substances List (NDSL). If a company were intending to manufacture or import these substances, they would be considered to be new to Canada and subject to notification under the *New Substances Notification Regulations (Chemicals and Polymers)*. Therefore, although the screening assessment focused on the uses of DNOC, a review of the fate and effects of its salts was also carried out.

This screening assessment includes consideration of information on chemical properties, hazards, uses and exposure. 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 August 2004 for ecological sections of the document and June 2003 for human health sections of the document. In addition, an industry survey was conducted in 2000 through a *Canada Gazette* notice issued under authority of section 71 of CEPA 1999.

The screening assessment report does not present an exhaustive or critical review of all available data. Rather, it presents the critical studies and lines of evidence pertinent the conclusion. One line of evidence includes consideration of risk quotients to identify potential for ecological effects. However, other concerns that affect current or potential risk, such as persistence, bioaccumulation, chemical transformation and trends in ambient concentrations, are also considered.

Evaluation of risk to human health involves consideration of data relevant to estimation of exposure (non-occupational) of the general population, as well as information on health hazards. Decisions for human health are based on the nature of the critical effect and/or margins between conservative effect levels and estimates of exposure, taking into account confidence in the completeness of the identified databases on both exposure and effects, within a screening context. The screening assessment does not represent an exhaustive or critical review of all available data. Rather, it presents a summary of the critical information upon which the conclusion is based.

This screening assessment was prepared by staff in the Existing Substances programs at Health Canada and Environment Canada. The substance matter in this report pertaining to ecological aspects has been subjected to external review. The report for a Screening Health Assessment was reviewed externally by V.C. Armstrong (Consultant) and staff of Toxicology Advice and Consulting Limited. 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. Additionally, the draft of this screening assessment was subject to a 60-day public comment period from June 23, 2007, to August 22, 2007. The State of the Science Report for a Screening Health Assessment has been posted on the Health Canada website since January 30, 2006, and the draft ecological screening assessment report on 4,6-dinitro-*o*-cresol (DNOC) has been posted on the Environment Canada website since July 2006.

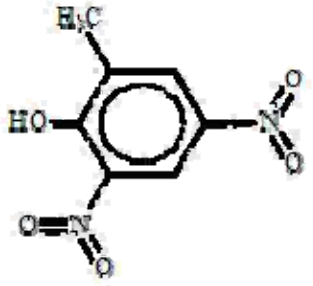
Information on ecological and human health screening assessments under CEPA 1999 may be linked from the CEPA Registry at www.ec.gc.ca/ceparegistry.

The critical information and considerations upon which the assessment is based are summarized below.

Substance Identity

Substance name
For the purposes of this document, this substance will be referred to as DNOC, a common name for this substance.

Table 1. Substance identity for DNOC

Chemical Abstracts Service Registry Number (CAS RN)	**534-52-1**
DSL name	**Phenol, 2-methyl-4,6-dinitro-**
National Chemical Inventories (NCI) names[1]	*Phenol, 2-methyl-4,6-dinitro-* (TSCA, AICS, PICCS, ASIA-PAC, NZIoC) *2-methyl-4,6-dinitro-phenol* (EINECS) *4,6-dinitro-o-cresol* (ENCS) *2,2'-methylene-bis-(4-methyl-6-tert-butylphenol)* (PICCS)
Other names	*2,4-dinitro-6-methylphenol; 3,5-dinitro-2-hydroxytoluene; 4,6-dinitro-2-methylphenol; 6-methyl-2,4-dinitrophenol; Antinonin; Antinonnin; Arborol; Degrassan; Dekrysil; Detal; Dillex; Dinitro; Dinitro-o-cresol; Dinitrocresol; Dinitrodendtroxal*
Chemical group (DSL Stream)	Discrete organics
Major chemical class or use	Phenols
Major chemical sub-class	Aromatic phenols
Chemical formula[2]	$C_7H_6N_2O_5$
Chemical structure	
SMILES[3]	[O-][N+](=O)c1cc([N+]([O-])=O)cc(c1O)C
Molecular mass[2]	198.1348 g/mol

[1] National Chemical Inventories (NCI). 2006: AICS (Australian Inventory of Chemical Substances); ASIA-PAC (Asia-Pacific Substances Lists); EINECS (European Inventory of Existing Commercial Chemical Substances); ENCS (Japanese Existing and New Chemical Substances); NZIoC (New Zealand Inventory of Chemicals); PICCS (Philippine Inventory of Chemicals and Chemical Substances); and TSCA (Toxic Substances Control Act Chemical Substance Inventory).

[2] ChemFinder (2003).

[3] Simplified Molecular Line Input Entry System.

Physical and Chemical Properties

Experimental and modelled data are available for DNOC.

Table 2 summarizes data on experimental physical and chemical properties of DNOC that are relevant to its environmental fate.

Table 2. Summarized experimental physical and chemical properties for the neutral form of DNOC

Property	Value	Temperature (°C)	Reference
Physical characteristics	Yellow, crystalline solid, odourless		HSDB 2000; IPCS 2000
Melting point (ºC)	85.8–87.5		PhysProp 2003; Verschueren 2001; NLM 2000; IPCS 2000
Boiling point (ºC)	220–378		PhysProp 2003; Verschueren 2001; NLM 2000; IPCS 2000
Density (kg/m^3)	1.58	20	HSDB 2003; NLM 2000; IPCS 2000
Vapour pressure (Pa)	$1.6 \times 10^{-2} - 4.79 \times 10^{-2}$	20–35	HSDB 2003; ATSDR 1995; IPCS 2000
Henry's Law constant ($Pa \cdot m^3/mol$)	$2.490 \times 10^{-2} - 1.4 \times 10^{-1}$	25	HSDB 2003; NLM 2000; IPCS 2000
Log K_{ow} (Octanol-water partition coefficient) (dimensionless)	2.12 (neutral species) – 2.564 (neutral species); 1.78 at pH 4; 0.087 at pH 7; 1.32 at pH 10		HSDB 2003; NLM 2000; IPCS 2000; Schwarzenbach et al. 1988; UNEP/FAO 2002

Log K_{oc} (Organic carbon-water partition coefficient) (dimensionless)	2.35–2.77; 1.3 (DNOC Na salt)		IPCS 2000
Water solubility (mg/L)	1000–198;	15–20	ChemFinder 2003; Schwarzenbach et al. 1988
	21.3–3300 (pH 4–10)		UNEP/FAO 2002
	100 000 (DNOC Na salt)		Vogue et al. 1994
Other solubilities (g/g)	4.3/100 (*ethanol*)		
	100/100 (*acetone*)		
	37/100 (*benzene*)		
pK_a (Acid dissociation constant) (dimensionless)	4.32		PALLAS (v. 4.0)
Conversion factor	1 ppm = 8.10 mg/m^3		NLM 2000; IPCS 2000

Sources

DNOC occurs in the environment primarily as a result of human activity, whether through direct release or through secondary transformation of atmospheric pollutants. It may form in the atmosphere following the reaction of 2-methylphenol with NO_x present in ambient air (ATSDR 1995). DNOC may also form in the atmosphere during the combustion of fossil fuels or as a result of photochemical reactions between precursor compounds (e.g., benzene, toluene) and hydroxyl radicals and nitrogen oxides (Tremp et al. 1993).

An industry survey was conducted by Environment Canada for the year 2000 (Environment Canada 2003a). Under section 71 of CEPA 1999, the *Notice with Respect to Certain Substances on the Domestic Substances List (DSL)* applied to any person who, during the 2000 calendar year, manufactured or imported DNOC, whether alone or in a mixture or in a product, in a total quantity greater than 10 000 kg. The survey results indicated that DNOC is not manufactured in Canada; however, between 100 and 1000 tonnes of DNOC were imported in 2000 by a single company (Environment Canada 2003a). Information received more recently indicates that the only company that had reported using DNOC in response to the section 71 notice ceased use of DNOC as of late 2002 (NOVA Chemicals Corporation 2007).

Two Canadian companies reported manufacture or import of DNOC in 1986, with amounts in the range of 100 to 1000 tonnes (Environment Canada 1990).

Uses

Historically, DNOC was used in Canada as an antioxidant, corrosion inhibitor, tarnish inhibitor and antiscaling agent, for a total of 99.9% of the Canadian market (Environment Canada 1990). Globally, the principal uses of DNOC are in the plastics industry as an inhibitor of polymerization in styrene and aromatic vinyl products; it is also used as an intermediate in the synthesis of fungicides, dyes and pharmaceuticals (IPCS 2000; UNEP/FAO 2002). Sources of exposure in the general environment are likely to be limited to fugitive releases from industrial sites and the combustion of fossil fuels. There is no indication that DNOC is present in consumer products.

DNOC was registered as an active ingredient in ten pesticides in Canada. Registration of the last two products that contained this active ingredient expired on December 31, 1990 (PMRA 2009). Three of the DNOC salts (sodium, ammonium and potassium) have been used as pesticides internationally but were never registered under the PCPA in Canada.

Releases to the Environment

Only one company reported releases of DNOC to the National Pollutant Release Inventory (NPRI) (Environment Canada 2003b). The facility, NOVA Chemicals, located in Sarnia, Ontario, reported only off-site transfers and no releases to water, air or soil. The company states

that all process water from its facility is collected, analyzed and sent to another facility for treatment in a biological oxidation unit, and that other quantities of waste DNOC would have been incinerated or sent for disposal. As such, NOVA Chemicals reports that there were no releases of DNOC to water from its facility (NOVA Chemicals Corporation 2007). Information about releases was requested in the survey conducted pursuant to section 71 of CEPA 1999, but no releases were reported (Canada 2001).

As indicated above, the use of DNOC by the NOVA Chemicals Corporation facility ceased as of late 2002 (NOVA Chemicals Corporation 2007). No facilities have reported to the NPRI for DNOC since 2003.

Environmental Fate

Environmental fate analysis combines information on the chemical behaviour of the substance with the properties of the receiving environment. The objective of fate analysis is to determine the multimedia distribution of the substance after its release into the environment. This includes consideration of the persistence and bioaccumulation of the substance in the environment.

The results of Level III fugacity modelling (EQC 2003) indicate that, if the chemical is released into water, the majority of DNOC would remain in water with a minor amount partitioning to sediments and less than one percent to air and soil (Table 3). With emissions solely to air, the majority of DNOC would partition to air, soil and water, with a negligible amount adsorbing to sediment. If DNOC were to be released equally to all three major environmental compartments (air, water, and soil), it would mainly partition to water and soil, with some DNOC partitioning to air.

Table 3. Results of the Level III fugacity modelling (EQC 2003)

	Percentage of substance partitioning into each compartment			
Substance released to:	**Air**	**Water**	**Soil**	**Sediment**
Air (100%)	47.7	20.1	32.0	0.212
Water (100%)	0.04	98.9	1.04	0.02
Soil (100%)	0.04	1.93	98.0	0.02
Air, water, soil (33% each)	6.65	63.3	29.3	0.66

Persistence and Bioaccumulation Potential

The information below was considered in evaluating whether DNOC meets the criteria for persistence and bioaccumulation as defined under the *Persistence and Bioaccumulation Regulations* under CEPA 1999 (Canada 2000). Persistence criteria are half-lives of greater than or equal to 2, 182, 365 and 182 days for air, water, sediment and soil, respectively. Bioaccumulation criteria are bioaccumulation factors (BAFs) or bioconcentration factors (BCFs) of greater than or equal to 5000 or a log K_{ow} of greater than or equal to 5.0.

Environmental Persistence

When DNOC is released to the environment, measured data demonstrate that DNOC will persist in air with an atmospheric oxidation half-life of 129 days (Table 4a). Predicted values indicate that photoxidation half-lives range from 8 hours to 53 days (Table 4b).

Empirical and predicted half-lives of 7, 37.5 and 58 days indicate that DNOC is expected to biodegrade in surface water, but at a relatively slow rate. The substance was reported to be difficult to degrade in activated sludge (< 20% degradation under aerobic conditions) (Tables 4a and 4b).

DNOC is expected to disappear from soil within 14 hours to 2 months (Callahan et al. 1979). DNOC is not expected to volatilize from dry or moist soils, based on its vapour pressure of 1.6 – 4.79 x 10^{-2} Pa (Table 2). After an 80-day lag period, DNOC rapidly degraded at a rate of 2.1 µg/L/day in groundwater and sediment (Table 4a).

Table 4a. Empirical data for degradation of DNOC

Medium	Fate process	Degradation value	Degradation endpoint / units	Reference
Air	Photoxidation	129	Half-life (days)	Howard et al. 1991; Atkinson 1987
Water	Biodegradation	7–58	Half-life (days)	Capel and Larson 1995; Mabey et al. 1981; IPCS 2000
Soil	Biodegradation	14 hours to < 2 months		Callahan et al. 1979
Waste water	Biodegradation	< 20%	28 days	Zahn and Wellens 1980
Sediment	Biodegradation	2.1	µg/L/day	Tuxen et al. 2000

Table 4b. Modelled data for degradation of DNOC

Fate process	Model and Model Basis	Model Result and Prediction	Extrapolated Half-life (days or hours)
Atmospheric oxidation	AOPWIN 2000	t½ = 35-53 days	>2 days
Ozone reaction	AOPWIN v. 2000	Half-life (days)	Not reactive
Biodegradation	BIOWIN 2000, Sub-model 3: Expert Survey (ultimate biodegradation)	2.4 "biodegrades fast" Half-life (days)	<182
Biodegradation	BIOWIN 2000 Sub-model 6: MITI non-linear probability	0.0009	>182

The empirical and modelled data (tables 4a and 4b) demonstrate that DNOC meets the persistence criteria for air (half-life in air > 2 days) and water (indicated by a degradation rate of less than 20% within the 28-day test period) (Zahn and Wellens 1980).

Potential for Bioaccumulation

A bioaccumulation factor of 25 was estimated by Gobas and Arnot (2003) for DNOC. This is significantly lower than the bioaccumulation criteria of BAF or BCF greater than 5000 as laid out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

Based on the criteria in the *Persistence and Bioaccumulation Regulations* (Canada 2000), DNOC is persistent in air and water and is not bioaccumulative.

Potential to Cause Ecological Harm

A scenario was developed using conservative assumptions to estimate potential environmental concentrations of DNOC that could result from its release from a hypothetical industrial facility. This estimation was done in spite of the absence of specific data indicating releases of DNOC to the environment, in recognition of two factors. First, it is recognized that some industrial facility operators are not fully aware of all potential sources of release of substances to the environment, including rinsate from the cleaning of reactors, transport and storage vessels, either directly at the facility or through third parties such as transporters or container recyclers. Second, there is the possibility that facilities that have not yet been identified are also using the substance. Of note, only facilities that are using greater than 10 000 kg per year of DNOC are required to report to NPRI. The requirement to respond to the section 71 notice for the year 2000 was also based on a threshold of 10 000 kg.

It was assumed in the conservative scenario that one customer was receiving the total annual import quantity (100–1000 tonnes). The scenario assumed releases of 0.2% of the annual import quantity of DNOC into the St. Clair River; based on professional judgement, this recognizes routine process losses and waste from equipment cleaning for a substance handled in bulk. This accounts for releases to solid waste and wastewater; using this percentage results in an estimated annual release of 200–2000 kg. If it is further assumed that DNOC is in use throughout the year and that there is continuous release (24 hours per day) over the year (350 operating days); daily releases would correspond to approximately 0.57–5.7 kg/day. Sewage treatment plant (STP) removal rates were also considered. The STP model (STP 2001) estimated that 27% of DNOC would be removed and that 73% would enter the environment in the form of final wastewater effluent from an STP.

Two main sources of atmospheric nitrophenols (a category that includes DNOC) have been reported in the literature. These include secondary formation by reactions in the troposphere and emissions from automobiles. Researchers have examined the atmospheric occurrence and formation of DNOC (Nojima et al. 1976; Alber et al. 1989; Richartz et al. 1990). DNOC has been shown to form as a secondary pollutant via the reaction of toluene and 2-methylphenol with nitrogen monoxide and hydroxyl radicals. It is difficult to estimate the quantity that may result from the anthropogenic release of precursor species. Direct emission of DNOC from car exhaust is likely only of minor importance. Under experimental conditions, exhaust from an automobile motor was found to contain DNOC at a rate of < 0.01 ng/m^3 (Tremp et al. 1993).

The NPRI (Environment Canada 2003b) reported that amounts of up to 2 tonnes of DNOC and its salts were, prior to 2002, annually "transferred for disposal" by NOVA Chemicals. For all years before 2002, the methods of treatment were biological, such as biooxidation, and incineration or thermal. For the year 2002, disposal was to a landfill.

Ecological Effects Assessment

Biotic Effects

Key studies of the toxicity of DNOC to organisms in different environmental media are presented in tables 6 to 9. Studies primarily on the acute toxicity of DNOC to microorganisms,

aquatic invertebrates, insects, terrestrial invertebrates and vertebrates were located in the literature. No acute or chronic marine toxicity data were identified.

Table 5. Empirical data for toxicity of DNOC to aquatic organisms

Test organism	Endpoint[1]	Value (mg/L)	Reference
Microorganisms			
Bacterium *Pseudomonas putida*	Toxic threshold, 16-h (cell multiplication inhibition)	16	Bringmann and Kühn 1980
Cyanobacterium *Microcystis aeruginosa*	Toxic threshold, 72-h (cell multiplication inhibition)	0.15	Bringmann and Kühn 1978
Protozoan *Entosiphon sulcatum*	Toxic threshold,16-h (cell multiplication inhibition)	5.4	Bringmann and Kühn 1980
Protozoan *Chilomonas paramecium*	Toxic threshold, 72-h (growth inhibition)	5.4	Bringmann and Kühn 1981
Protozoan *Uronaemia parduczi*	Toxic threshold, 72-hour (growth inhibition)	0.012	Bringmann and Kühn 1981
Aquatic plants			
Green alga *Scenedesmus quadricauda*	Toxic threshold, 16-h (cell multiplication inhibition)	13	Bringmann and Kühn 1980
Green alga *Scenedesmus subspicatus*	96-hour EC_{50} (biomass) 48-hour EC_{50} (growth rate)	6 12	Sewell et al. 1995a Sewell et al. 1995a
Lemna minor	Specific growth rate, 7-day exposure	0.32	Sloof and Canton 1983
Aquatic invertebrates			
Water flea *Daphnia magna*	24-hour LC_{50} 14-day LC_{50} 14-day NOEC (reproduction) 24-hour LC_{50} 24-hour NOEC (mortality) 21-day NOEC (reproduction)	5.7 1.6 0.6 2.3 1.5 1.3	van der Hoeven 1984 van der Hoeven 1984 van der Hoeven 1984 Kühn et al. 1989 Kühn et al. 1989 Kühn et al. 1989
Water flea *Daphnia pulex*	48-hour EC_{50} 3-hour LC_{50} (DNOC sodium salt)	0.145 3.5	Mayer and Ellersieck 1986 PAN 2004
Amphipod *Gammarus fasciatus*	96-hour LC_{50}	0.11	Mayer and Ellersieck 1986
Stonefly *Pteronarcys californica*	96-hour LC_{50}	0.32	Mayer and Ellersieck 1986
Vertebrates (fish)			
Bluegill *Lepomis macrochirus*	96-hour LC_{50} 96-hour LC_{50}	0.95 0.36	Sewell et al. 1995b Mayer and Ellersieck 1986
Rainbow trout *Oncorhynchus mykiss*	96-hour LC_{50} 96-hour NOEC 96-hour LC_{50}	0.45 0.32 0.066	Sewell et al. 1995c Sewell et al. 1995c Mayer and Ellersieck 1986
Atlantic salmon *Salmo salar*	96-hour LC_{50}	0.20	Zitko et al. 1976
Bluegill *Lepomis macrochirus*	96-hour LC_{50}	0.23	Buccafusco et al. 1981
Goldfish *Carassius auratus*	48-hour LC_{50} (DNOC sodium salt)	0.45	PAN 2004
Common carp	13-day NOEC (pH 6.9–9.0)	$\leq$ 0.25	Ghillebaert et al. 1995

Test organism	Endpoint[1]	Value (mg/L)	Reference
Cyprinus carpio	13-day NOEC (pH 7.8) 13-day NOEC (pH 9.0)	0.5–1.0 no effect	
Common carp *Cyprinus carpio*	48-hour LC_{50} (DNOC sodium salt)	0.17	PAN 2004
Medaka *Oryzias latipes*	48-hour LC_{50} (DNOC sodium salt)	0.20	PAN 2004

EC50 = the concentration of a substance that is estimated to cause some effect to 50% of the test organisms
LC_{50} = the concentration of a substance that is estimated to be lethal to 50% of the test organisms
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

Table 6. Acute toxicity of DNOC to terrestrial plants

Organism	Endpoint	Concentration (mg/L)	Reference
Tobacco *Nicotiana sylvestris*	3-hour ED_{50} (growth inhibition of the pollen tube culture)	0.466	Strube et al. 1991

ED_{50} =The dose estimated to produce an effect to50% of the population

Table 7. Acute toxicity of DNOC to terrestrial invertebrates

Organism	Endpoint	Concentration	Reference
Earthworm *Eisenia fetida*	7-day LC_{50} 14-day LC_{50} 14-day NOEC	17 mg DNOC/kg of soil 15 mg DNOC/kg of soil 10 mg DNOC/kg of soil	van der Hoeven 1992
Honey bee *Apis mellifera*	LD_{50} (oral) LD_{50} (contact)	2.04 ± 0.25 µg DNOC/bee 406 ± 27µg DNOC/bee	Beran and Neururer 1955

LC_{50} = the concentration estimated to be lethal to 50% of the organisms
NOEC = no-observed-effect concentration; LD_{50} = the dose estimated to be lethal to 50% of the organisms.

Table 8. Toxicity of DNOC to terrestrial vertebrates

Organism	Endpoint	Concentration (mg/kg-bw)	Reference
Japanese quail *Coturnix japonica*	24-hour LD_{50}	14.8 (95% CI 13–17)	Dickhaus and Heisler 1980
Japanese quail *Coturnix japonica*	8-day LC_{50}	106	Til and Kengen 1980
Pheasant	LD_{50}	8.4	Janda 1970
Partridge	LD_{50}	8.3	Janda 1970
Rat	90-day LOEL	2.5 (per day)	Den Tonkelaar et al. 1983

LD_{50} = the dose estimated to be lethal to 50% of the organisms.
LC_{50} = the concentration estimated to be lethal to 50% of the organisms.
LOEL = lowest-observed-effect level.
CI = confidence interval.

The most sensitive aquatic vertebrates reported in the literature are rainbow trout (Mayer and Ellersieck 1986; Sewell et al. 1995c). The authors reported LC_{50} values (the concentration estimated to be lethal to 50% of the organisms) of 0.066 and 0.45 mg/L, respectively. The 96-hour LC_{50} study reported by Sewell et al. (1995c) is an unpublished study; however, it was cited in a peer-reviewed report (IPCS 2000). Atlantic salmon and bluegill are also sensitive, with 96-

hour LC_{50} values of 0.20 mg/L and 0.23 mg/L, respectively (Zitko et al. 1976; Buccafusco et al. 1981).

The effect of DNOC on terrestrial vertebrates (mink and otter) (Critical Toxicity Value [CTV] for wildlife) was calculated using the repeated mammalian (rat) oral dose toxicity data provided for the substance (2.5 mg/kg-bw per day for a 90-day rat dietary exposure study, lowest-observed-effect level [LOEL]) (Den Tonkelaar et al. 1983). The $CTV_{wildlife}$ is calculated by taking the chronic value (geometric mean of the no-observed-effect level [NOEL] and LOEL) from the rat study and correcting it for body weight of a predictive sentinel species (Sample et al. 1996). In this case, the predictive sentinel species are the piscivorous mammals mink and river otter.

The $CTV_{wildlife}$ is thus calculated as:

$$CTV_{wildlife} = ChV_{ts} \cdot (BW_{ts}/BW_{pss})$$

where:

ChV_{ts} = chronic value for test species (geometric mean of LOEL [2.5 mg/kg-bw per day] and NOEL [0.25 mg/kg-bw per day] = 0.8 mg/kg-bw per day)
BW_{ts} = mean body weight of test species (0.35 kg)
BW_{pss} = body weight of predictive sentinel species (0.807 kg for mink; 6.01 kg for otter) (2004 personal communicaton from P. Martin, Canadian Wildlife Service, Environment Canada, Ontario Region; unreferenced).

Therefore, $CTV_{wildlife} = 0.8 \times (0.35/0.807) = 0.35$ for mink and $0.8 \times (0.35/6.01) = 0.047$ for otter.

The $PNEC_{wildlife}$ is calculated from the $CTV_{wildlife}$ as follows:

$$PNEC_{wildlife} = CTV_{wildlife}/AF$$

where:

$PNEC_{wildlife}$ = wildlife predicted no-effect concentration (mg/kg-bw per day)
AF = application factor (interspecies variation, laboratory to field extrapolation) (10).

Therefore, the $PNEC_{mink}$ is 0.035 mg/kg-bw per day, and the $PNEC_{otter}$ is 0.0047 mg/kg-bw per day.

Ecological Exposure Assessment

Concentrations in the Atmosphere and Precipitation

No monitoring data for DNOC in the atmosphere or precipitation in Canada were identified. Monitoring data from other countries are summarized in Table 9.

Table 9. Concentrations of DNOC in the atmosphere and precipitation

Location	Sampling period	No. of samples[1]	Mean concentration (µg/L)[2]	Reference
Denmark	October–November 2001	5	[0.07–3.2 ng/m^3]	Bossi and Andersen 2003
Netherlands	2000–2001	18	> 0.1	Duyzer and Vonk 2002
Italy, Milan	November 1998	12	[600–7200], rainwater	Belloli et al. 2000
Germany, Bavaria	1995–1998	ns	[0.1–2.4], rainwater (approximated from graph)	Schüssler and Nitschke 2001
Germany, Bavaria	July 1998 – March 1999	> 100	3.4 [0.5–4.2], fogwater	Römpp et al. 2001
Germany, Hanover	1988	ns	Qualitatively identified in rain and snow	Alber et al. 1989
England, Great Dun Fell	April–May 1993	6	0.7 [0.26–2.13], cloudwater	Lüttke and Levsen 1997
Germany, Mount Brocken	June 1994	6	4.2 [0.1–10], cloudwater	Lüttke et al. 1999
Switzerland, Dübendorf	March–November 1985	3	0.05 µg/m^3, ambient air [0.95–1.6 µg/L], rain	Leuenberger et al. 1988

[1] ns = not specified.
[2] Unless otherwise specified. The range of values is indicated in square brackets, if available (e.g., [minimum–maximum]).

DNOC has been detected in atmospheric air and precipitation at a number of locations in Europe, and the presence of nitrated phenols in rain is not explained solely by input from pesticide applications (Leuenberger et al. 1988). DNOC has been shown to partition favourably from the gas phase to the aqueous phase, and its presence in rainwater would therefore be expected (Schwarzenbach et al. 2003). DNOC was detected in Denmark, even though the substance had not been used there in the previous 10 years (Danish Environmental Protection Agency 2001). The concentrations found in rain in Denmark are of the same order of magnitude as have been detected in England, Germany and Switzerland.

As no atmospheric or precipitation monitoring data for DNOC in Canada could be located, a series of release scenarios was developed to estimate the amount of DNOC that could be released into receiving waters in Canada as a result of rainfall scavenging of DNOC in the atmosphere. The scenarios incorporated precipitation data for 12 Canadian cities, an estimate of the amount of DNOC in rainwater, and a calculation of runoff from built-up and natural areas into the receiving STPs. It was assumed that the rain event that would result in DNOC being removed from the atmosphere would be a heavy rainfall and that DNOC would be washed out in the early stages of the rain event and not over the length of the rainfall. The concentration of DNOC used in the scenario is based on precipitation values from Europe that were considered realistic possible levels of DNOC in air in Canada. The mean concentration of DNOC in cloudwater from northern Germany (4.2 µg/L) was selected. It was assumed that rainwater would be released as a point source from an STP but that it would not undergo STP treatment, as STP removal efficiency during a storm event is likely to be poor. The highest concentrations of DNOC were

estimated in receiving waters from the STPs in London, Ontario (0.0023 mg/L), Guelph, Ontario (0.0023 mg/L), and Granby, Quebec (0.0025 mg/L).

Aquatic Concentrations

No recent aquatic monitoring data for DNOC in Canada were identified. Older data on levels of DNOC in Canadian waters as well as in other countries are summarized in Table 10.

Table 10. Concentrations of DNOC in surface water

Location	Sampling period[1]	No. of samples[1]	Detection limit[1] (µg/L)	Mean concentration[1,2] (µg/L)	Reference
Italy, River Po	January 1994 – December 1996	ns (samples were taken at 15-day intervals during the sampling period)	0.1	nd	Davi and Gnudi 1999
Germany, Elbe River	1994	ns	0.05	[ns–0.06]	Pietsch et al. 1995
Denmark, Hølvads Rende area, soil water, drainage water, stream water	October 1989 – December 1991	ns	ns	0.005 (soil water) nd (drainage water) [0.02–0.16] (stream water)	Mogensen and Spliid 1995
Denmark, Bolbo Bæk area, soil water, stream water	April 1990 – December 1991	ns	ns	0.005 (soil water) 0.16 (stream water)	Mogensen and Spliid 1995
Denmark, four ponds	November 1989 – December 1990	ns	ns	[nd–0.64]	Mogensen and Spliid 1995
Netherlands, Meuse River and Rhine River; Slovakia, Danube River and Nitra River	ns	4	0.4	nd	Brouwer and Brinkman 1994
Germany, Bavaria, Mount Ochsenkopf and University of Bayreuth campus	Fall 1988	ns	1.98	[nd–12.5]	Richartz et al. 1990
Point source					
Ontario, St. Clair River near Sarnia (industrial area)	1979	24	1	[nd–10]	Munro et al. 1985
Ontario, St. Clair River near Sarnia (industrial area)	1980	25	1	nd	Munro et al. 1985
Ontario, St. Clair River near Sarnia, industrial effluent, process/sewer water, township ditch water[3]	1979	119	1	[nd–10 000]	Munro et al. 1985

Location	Sampling period[1]	No. of samples[1]	Detection limit[1] (µg/L)	Mean concentration[1,2] (µg/L)	Reference
Ontario, St. Clair River near Sarnia, industrial effluent, process/sewer water, township ditch water[3]	1980	61	1	nd	Munro et al. 1985
United States, California, groundwater	ns	ns	ns	ns–35	Hallberg 1989
Italy, Taranto, surface seawater contaminated by oil refinery or iron and steel factory wastes	ns	2	0.017	[0.030–0.065]	Cardellicchio et al. 1997
Unspecified location, oil refinery effluent, paper mill effluent	ns	ns	0.5	nd	Paterson et al. 1996

[1] ns = not specified; nd = not detected.
[2] The range of values is indicated in square brackets, if available (e.g., [minimum–maximum]).
[3] Mean concentration in effluent is presented as an indication of resulting exposure. This value was not included in the section on releases of DNOC, as details on effluent quantities and release rate were not provided.

As no recent Canadian surface water monitoring data were identified, aquatic exposure estimates were modelled. The scenario uses the ChemSim model (Environment Canada 2003c) to predict estimated exposure values. ChemSim model runs were done for three river flow estimates and two loading rates (calculated in the section on releases of DNOC), for a total of six model runs. As indicated in the release scenario, it is assumed that DNOC is in use throughout the year and that there is continuous release (24 hours per day) over the year (350 operating days). Two estimates of low river flow (2.5th and 10th percentiles) were selected to derive predicted environmental concentrations (PECs) under low-flow conditions. The 50th-percentile flow value was also selected to estimate PECs under more typical conditions. The maximum concentration of DNOC at 20 m downstream of the reporting facility with a worst-case scenario release of 5.7 kg/day and a 2.5th-percentile river flow is estimated to be less than 0.006 mg/L. If STP treatment is considered, a PEC of 0.0014 mg/L is estimated.

Concentrations in Sediment, Sewage Sludge and Soil

Monitored soil, sediment and sludge concentrations of DNOC are summarized in Table 11. The high flow and velocity of the St. Clair River would rapidly dilute and disperse the substance, and only a minor amount of DNOC is expected to partition to sediments (1%). Based on the results of modelling, at a release rate of 5.7 kg/day, 0.057 kg/day (or 1%) would be available to be adsorbed onto sediments.

Table 11. Concentrations of DNOC in soil, sediment and sludge

Location	Sampling period[1]	No. of samples[1]	Detection limit[1] (ng/g)	Mean concentration[1,2] (ng/g)	Reference
Ontario, old urban parkland soil	ns	60	100	Ontario typical range < W[3]	OMEE 1994

Location	Sampling period[1]	No. of samples[1]	Detection limit[1] (ng/g)	Mean concentration[1,2] (ng/g)	Reference
Ontario, rural parkland soil	ns	101	100	Ontario typical range < W[3]	OMEE 1994
Canada, agricultural soil	ns	30	50	nd	Webber 1994
11 sites across Canada, sludge samples	September 1993 – February 1994	12 samples/site	ns	nd	Webber and Nichols 1995
Sediment, artificial islands, Beaufort Sea	ns	ns	ns	< 10 (dry weight)	Fowler and Hope 1984
Canadian municipal sludges	1980–1985	15	ns	[1200–1500] (dry weight)	Webber and Lesage 1989
Poland, Holy Cross mountains, soil	July 3–6, 1996	8	1	nd	Migaszewski 1999
Italy, Taranto, sediment contaminated by oil refinery or iron and steel factory wastes	ns	2	ns	nd	Cardellicchio et al. 1997

[1] ns = not specified; nd = not detected.
[2] The range of values is indicated in square brackets, if available (e.g., [minimum–maximum]).
[3] < W is a qualifier, given to indicate that the sample may contain the analyte but the level would probably not exceed the laboratory method detection limit (MDL). W is approximately one-third to one-fifth of the MDL (OMEE 1994).

DNOC was detected in 13% of Canadian municipal sludges sampled during the period 1980–1985 at concentrations ranging from 1200 to 1500 ng/g dry weight, with a median concentration of 1300 ng/g dry weight (Webber and Lesage 1989). It was not detected (detection limit not stated) in sludge or sludge compost from various locations in Canada sampled in 1993–1994 (Webber and Nichols 1995).

DNOC was not detected (method detection limit = 100 ng/g) in 101 samples of "rural parkland" soil or in 60 samples of "old urban parkland" soil in Ontario (OMEE 1994). Similarly, DNOC was not detected (detection limit = 50 ng/g) in agricultural soil from various locations across Canada (Webber 1994).

Concentrations in Biota

DNOC was not detected in fish composite samples (detection limit not stated) from the United States (DeVault 1985).

As indicated in the section on environmental fate and partitioning, DNOC has a relatively low bioaccumulation potential. However, as will be seen in the section on effects characterization, results of repeated oral dose toxicity studies indicate that mammals may be fairly sensitive to DNOC. Therefore, wildlife exposure to DNOC from food and water has been estimated.

A PEC for wildlife was estimated based on a calculation of the total daily intake of the substance by mink and otter. An energetics model based on the general exposure model for wildlife from the U.S. Environmental Protection Agency's (EPA) Exposure Factors Handbook (US EPA 1993) was used.

$$TDI = \left[FMR\left(\frac{C_i \cdot P_i}{GE_i \cdot AE_i} \right) \right] \cdot Pt$$

where:

TDI = total daily intake (mg/kg-bw per day)
FMR = normalized free metabolic rate of wildlife receptor of interest (250 kcal/kg-bw per day for mink and river otter)
C_i = concentration of contaminant in the ith prey species (mg/kg-bw) (see below)
P_i = proportion of the ith prey species in the diet (unitless) (default = 35% for mink; 100% for otter)
GE_i = gross energy of the ith prey species (default = 850 kcal/kg-bw prey)
AE_i = assimilation efficiency of the ith prey species by the wildlife receptor (default = 0.91)
Pt = proportion of the time the receptor spends in the contaminated area (= 9% for mink and 0.06% for otter).

The model incorporated the metabolic rate of the wildlife receptors of interest (mink and otter), the proportion of food uptake by the receptors and the amount of time the animals spend in the contaminated area, which is based on the typical habitat range of the wildlife receptors.

The concentration of the substance in a fish (C_i) must be estimated based on the highest PEC_{water} and a BAF. The BAF was estimated using the Modified Gobas Model (Gobas and Arnot 2003). The BAF represents a benthic/pelagic food chain and estimates the accumulation from all sources in a mid-trophic-level fish that would typically be eaten by a mammalian piscivore.

$$C_i = PEC_{water} \cdot BAF$$

where:

C_i = concentration in a prey fish (mg/kg-bw)
PEC_{water} = PEC calculated for surface water (mg/L) (see section on aquatic concentrations)
BAF = bioaccumulation factor for substance (L/kg) (see section on environmental fate and partitioning).

$$C_i = 0.0014 \cdot 25 = 0.035$$

The model estimated PECs of 0.0004 mg/kg-bw per day and 0.000 007 mg/kg-bw per day for mink and otter, respectively.

Characterization of Ecological Risk

As part of risk characterization, one line of evidence includes consideration of risk quotients to identify potential for ecological effects. Other factors that affect current or potential risks, such as persistence, bioaccumulation and trends in ambient concentrations, are also considered.

Risk Quotient Analysis

Critical exposure and effects results and risk quotients are summarized in Table 12 and described in more detail below.

Table 12. Summary of data used in risk quotient (RQ) analysis of DNOC

Scenario	PEC	CTV	AF[1]	PNEC	RQ (PEC/ PNEC)
Pelagic organisms					
Industrial release; rainbow trout	0.0014 mg/L	0.26 mg/L	100	0.0026 mg/L	0.54
Rainfall; rainbow trout	0.0025 mg/L	0.26 mg/L	10	0.026 mg/L	0.096
Soil organisms					
Earthworm	0.1 mg/kg	15 mg/kg dry weight	100	0.15 mg/kg dry weight	0.67
Wildlife consumers					
Mink	0.0004 mg/kg-bw per day	0.35	10	0.035 mg/kg-bw per day	0.011
River otter	0.000 007 mg/kg-bw per day	0.047	10	0.0047 mg/kg-bw per day	0.0015

[1] AF = application factor.

Pelagic Organisms

For pelagic organisms, a risk quotient was developed using the average 96-hour LC_{50} values of rainbow trout reported by Mayer and Ellersieck (1986) (0.066 mg/L) and Sewell et al. (1995c) (0.45 mg/L). The average of the two studies, which is the CTV, is 0.26 mg/L.

For the *industrial release scenario*, if STP treatment is considered (27% removal efficiency), the PEC will be 0.0014 mg/L. Using an application factor of 100 on the CTV to account for acute to chronic extrapolation and intra- and interspecies variations, differently sensitive biological endpoints and laboratory to field extrapolations, the PNEC is calculated to be 0.0026 mg/L.

The risk quotient is therefore calculated as:

$$\frac{\text{PEC}}{\text{PNEC}} = \frac{0.0014 \text{ mg/L}}{0.0026 \text{ mg/L}} = 0.54$$

Even with STP removal considered, this represents a conservative scenario due largely to the very high quantity of DNOC assumed to be used by a single facility.

The maximum PEC under the defined *rainfall scenario* was determined to be 0.0025 mg/L with no STP treatment due to the assumption of a heavy rainfall. As rainfall represents an acute exposure scenario, the application factor does not need to account for acute to chronic extrapolation. Therefore, using an application factor of 10 and the same CTV of 0.26 mg/L for rainbow trout, a PNEC of 0.026 mg/L is calculated. The risk quotient is therefore:

$$\frac{\text{PEC}}{\text{PNEC}} = \frac{0.0025 \text{ mg/L}}{0.026 \text{ mg/L}} = 0.096$$

Soil Organisms

There are no quantified amounts of DNOC concentrations in Canadian soils. OMEE (1994) did not detect DNOC in 161 soil samples collected from soils in Ontario. The method detection limit of 0.1 mg/kg (100 ng/g) will be used as a surrogate for the level of DNOC in Canadian soil and is selected as the PEC.

One study was located in the literature on the effects of DNOC on terrestrial organisms. The LC_{50} from a 14-day acute toxicity study on the earthworm is 15 mg/kg of soil. This value is selected as the CTV for exposures of soil organisms to DNOC. Dividing the value by a factor of 100 to account for extrapolation from laboratory to field conditions, acute to chronic ratio and interspecies and intraspecies variations in sensitivity gives a PNEC of 0.15 mg/kg.

The risk quotient for soil organisms is therefore:

$$\frac{\text{PEC}}{\text{PNEC}} = \frac{0.1 \text{ mg/kg}}{0.15 \text{ mg/kg}} = 0.67$$

Aquatic Wildlife

The PECs for the mink and river otter were estimated to be 0.0004 mg/kg-bw per day and 0.000 007 mg/kg-bw per day, respectively. The PNEC for the mink was estimated to be 0.035 mg/kg-bw per day, and the PNEC for the river otter was calculated to be 0.0047 mg/kg-bw per day.

The risk quotients for aquatic wildlife are thus calculated to be:

$$\frac{\text{PEC}_{\text{mink}}}{\text{PNEC}_{\text{mink}}} = \frac{0.0004 \text{ mg/kg-bw per day}}{0.035 \text{ mg/kg-bw per day}} = 0.011$$

$$\frac{\text{PEC}_{\text{otter}}}{\text{PNEC}_{\text{otter}}} = \frac{0.000\,007 \text{ mg/kg-bw per day}}{0.0047 \text{ mg/kg-bw per day}} = 0.0015$$

Benthic Organisms

No monitoring data for DNOC in sediments in Canada were identified. Level III multimedia fate simulation estimated that only about 1% of DNOC is expected to partition to sediments. It is therefore believed that there will be minimal exposure of benthic organisms to DNOC.

Weight-of-Evidence Analysis

The risk quotient analyses for pelagic and soil organisms and wildlife have shown that it is unlikely that organisms are currently exposed to concentrations of DNOC above known effect thresholds. This conclusion is based on import levels and locations where DNOC was used industrially in the year 2000, and the current state of knowledge of its atmospheric chemistry.

A conservative scenario based on concentrations of DNOC in precipitation that could be expected to enter Canadian receiving water indicated that the potential for risk to aquatic organisms from this source is low.

In addition, modelling estimates of industrial releases to the St. Clair River indicate that DNOC is not likely to have adverse effects on pelagic or benthic organisms. This is based on a conservative release scenario developed for a facility located in the same region as the one company that reported use of DNOC in 2000 in response to a notice published under section 71 of CEPA 1999 and that reported to the NPRI. It is noted that the reporting facility ceased use of DNOC in late 2002.

Although sorption is low at environmentally relevant pHs, little leaching to groundwater has been found, likely due to biodegradation.

Potential sources of release of DNOC to the environment are to air and water. Based on its properties, DNOC is persistent in air and water but is not bioaccumulative. Long-range transport modelling estimates that it will be transported over moderate distances, and a decreasing concentration with increasing latitude is expected.

Uncertainties in Evaluation of Ecological Risk

There are uncertainties associated with development of the PNECs used in this assessment. However, a moderate number of empirical studies from different sources were identified, and this increases confidence in the values. Application factors of 10–100 were used to account for information gaps relating to chronic toxicity, effects in the field, and effects on potentially more sensitive species.

Very few Canadian monitoring data are available for DNOC, and those that were identified were fairly old. To both support the limited amount of empirical data and provide greater insight into the potential range of levels of DNOC in the environment, releases were estimated and fate and exposure were modelled. Entry of DNOC into the environment from two sources was considered—industrial releases and precipitation containing DNOC scavenged from the atmosphere. To address the significant uncertainty in these estimations, conservative assumptions were used to ensure that errors would be protective of the environment.

Although there have been no reports of direct releases of DNOC to water from industrial facilities, a conservative scenario was developed to estimate possible releases from an industrial source. This conservatively assumed an upper-limit estimate of the quantity of DNOC potentially

used by a single facility; a slightly conservative estimate of the fraction of substance typically released due to handling practices for a substance used in bulk; and a low-percentile estimate of river flow for the receiving water body used in the scenario. Flow characteristics of the St. Clair River were used in the exposure scenario, as the only facility that had reported use of DNOC was located close to this water body. This river is extremely fast flowing and consequently disperses effluents very rapidly. Were there to be facilities with substantive releases to smaller water bodies, then the assumptions used in this scenario might not be sufficiently protective. However, it is believed that there are currently no large users of DNOC in Canada, and it is possible that the substance is no longer in commercial use in Canada.

Estimation of possible exposure from atmospherically generated DNOC in precipitation conservatively assumed that the concentration in the atmosphere in Canada would be similar to that in more heavily populated regions of Europe; that the rainfall event would be particularly heavy; that a high percentage of precipitation from a census subdivision would be released to the receiving river body through a single discharge point; and that there would be no removal of DNOC by the municipal STP. In particular, the assumption that atmospheric concentrations in Canada would be the same as average to high concentrations in Germany, which is much more heavily populated and industrialized, is uncertain. While it is believed that use of monitoring data from Germany in the scenario is conservative, the origins of atmospherically generated DNOC are at present not well understood, and no Canadian atmospheric monitoring data were identified for comparison.

Potential to Cause Harm to Human Health

Exposure Assessment

The upper-bounding estimate of exposure to DNOC for the general population is 0.06 µg/kg-bw per day for the 0- to 6-month (formula-fed) age group, based on very limited data from Canadian surveys of drinking water and soil (OMEE 1994; City of Toronto Water and Wastewater Services Division 2002a, 2002b, 2002c, 2002d) and an estimated concentration of DNOC in air in Switzerland (Leuenberger et al. 1988) (see Appendix 1). No quantitative data on levels of DNOC in food were identified. Confidence in the database for estimating exposure is considered moderate, since there is information for conservative estimation of exposure through drinking water and air, the likely principal media of exposure. The levels of DNOC in drinking water were below the detection limit; thus, estimates based on the detection limit likely overestimate exposure. The concentration of DNOC in air was estimated from rain samples but is considered to be conservative, as it is higher than levels measured in automobile exhaust, a source of DNOC (Tremp et al. 1993).

Health Effects Assessment

A health assessment of DNOC was published by the International Programme on Chemical Safety (IPCS) in 2000 (see Appendix 2 for an overview of the toxicological database, in which confidence is considered to be high, in view of the wide range of toxicity studies available). Although the IPCS did not select a critical study for use as a basis of a tolerable intake or guidance value, the lowest-observed-effect level (LOEL) identified in that review that is

considered to be the critical effect level is 2.5 mg/kg-bw per day in a 90-day rat dietary exposure study, with resulting dose-related decreases in blood pyruvate and triiodothyronine levels (Den Tonkelaar et al. 1983). Although several lower effect levels were reported in the IPCS assessment, there was less confidence in these studies due to the fact that insufficient details were available; however, these lower values were generally within an order of magnitude of the effect level considered to be critical. Similarly, in very early clinical investigations of the potential application of DNOC in the treatment of obesity, effects associated with increases in basal metabolic rate were observed in individuals administered doses in the range of this critical value. DNOC was not carcinogenic in the only long-term study identified (Broadmeadow 1991), and the weight of evidence for genotoxicity was considered to be equivocal by the IPCS (2000), as positive results were observed in some but not all *in vivo* assays in which rodents were administered doses generally greater than the critical effect level for non-neoplastic effects. Similarly, the results of modelling of *in vivo* and *in vitro* genotoxicity endpoints are also equivocal.

Confidence in the database upon which the critical effect level is based is considered to be high in view of the wide range of toxicity studies available (i.e., acute toxicity, repeated dose toxicity, chronic toxicity/carcinogenicity, genotoxicity, reproductive and developmental toxicity and immunotoxicity). There is some uncertainty concerning lower effect levels reported in secondary accounts of studies for which original reports could not be obtained; however, since these values are generally within an order of magnitude of the effect level considered to be critical, they would not alter the conclusion of the screening assessment. There is also uncertainty with regards to the potential genotoxicity of DNOC, as the IPCS (2000) concluded it to be equivocal.

Characterization of Risk to Human Health

Comparison of a conservatively selected lowest effect level (i.e., 2.5 mg/kg-bw per day) for slight changes in biochemical parameters in a 90-day study in rats to the highest of the upper-bounding estimates of exposure for all age groups in the population (i.e., 0.06 µg/kg-bw per day) for the 0- to 6-month (formula-fed) age group resulted in a margin of exposure of approximately 41 700. In light of the moderate to high confidence in the databases on exposure and effects upon which this assessment is based and the conservative nature of this evaluation, including the use of an upper-bounding exposure estimate and lowest effect level, this margin is considered adequate to address elements of uncertainty associated with limitations of the database for health effects and population exposure and intraspecies and interspecies variations in sensitivity, as well as the biological adversity or severity of the effects deemed critical.

Conclusion

Based on the information presented in this screening assessment, it is concluded that DNOC 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. In addition, it is concluded that DNOC is 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 DNOC does not meet the criteria in section 64 of the *Canadian Environmental Protection Act, 1999*. Additionally, DNOC meets the criteria for persistence but does not meet the criteria for bioaccumulation set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

References

Adler B, Braun R, Schoneich J, Bohme H. 1976. Repair-defective mutants of *Proteus mirabilis* as a pre-screening system for the detection of potential carcinogens. Biol Zentralbl 95:463–469 [cited in IPCS 2000].

Alber M, Böhm HB, Brodesser J, Feltes J, Levsen K, Schöler HF. 1989. Determination of nitrophenols in rain and snow. Fresenius Z Anal Chem 334:540–545.

Allen PA, Biedermann K, Terrier C. 1990a. Embryotoxicity study (including teratogenicity) with DNOC Technical in the rabbit (dermal application). Unpublished report prepared for Pennwalt Holland by RCC, Itingen, Switzerland (RCC Study No. 215638) [cited in IPCS 2000].

Allen PA, Biedermann K, Terrier C. 1990b. Embryotoxicity study (including teratogenicity) with DNOC Technical in the rabbit (oral administration). Unpublished report prepared for Pennwalt Holland by RCC, Itingen, Switzerland (RCC Study No. 215651) [cited in IPCS 2000].

Ambrose AM. 1942. Some toxicological and pharmacological studies on 3,5-dinitro-*o*-cresol. J Pharm Exp Ther 76:245–251 [cited in IPCS 2000].

[AOPWIN] Atmospheric Oxidation Program for Windows [Estimation Model]. 2000. Version 1.91. 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

Arustamyn AN. 1972. [The toxicity of dinitro-*ortho*-cresols for warm-blooded animals and problems of industrial hygiene in its application.] Tr Inst Vet Sanit 45:166–169 (in Russian) [cited in IPCS 2000].

Atkinson R. 1987. Kinetics and mechanisms of the gas-phase reactions of the hydroxyl radical with organic compounds. Int J Chem Kinet 19:799–828.

[ATSDR] Agency for Toxic Substances and Disease Registry. 1995. Toxicological profile for dinitrocresols. Atlanta (GA): Public Health Service, United States Department of Health and Human Services. 204 pp.

Belloli R, Bolzacchini E, Clerici L, Rindone B, Sesana G. 2000. Nitrophenols in air and rainwater. Division of Environmental Chemistry Preprints of Extended Abstracts 40(1):378–381.

Ben Dyke R, Sanderson DM, Noakes DN. 1970. Acute toxicity data for pesticides. World Rev Pest Control 9(3):119–127 [cited in IPCS 2000].

Beran F, Neururer J. 1955. About understanding of the effects for pesticides on the honey bee (*Apis mellifera* L.). Hazards of pesticides to honey bees. Pfanzenschutzberichte 15(8/12):97–147 [cited in IPCS 2000].

[BIOWIN] Biodegradation Probability Program for Windows [Estimation Model]. 2001. Version 4.02. Washington (DC): U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics; Syracuse (NY): Syracuse Research Corporation.

Bossi R, Andersen HV. 2003. A multiresidue method for the determination of pesticides and selected nitrophenols in the atmosphere. In: Del Re AAM, Capri E, Padovani L, Trevisan M (editors). Pesticides in air, plant, soil and water systems. Proceedings of the 12th Symposium on Pesticide Chemistry, Piacenza, Italy, June 4–6, 2003. La Goliardica Pavese s.r.l, Pavia, Italy. p. 781–788.

Bringmann G, Kühn R. 1978. Grenzwerte der schadwirkung wassergefährdender stoffe gegen blaualgen (*Microcystis aeruginosa*) und Grünalgen (*Scenedesmus quadricauda*) im Zellvermehrungshemmtest. Vom Wasser 50:45–60.

Bringmann G, Kühn R. 1980. Comparison of the toxicity thresholds of water pollutants to bacteria, algae, and protozoa in the cell multiplication inhibition test. Water Res 14:231–241.

Bringmann G, Kühn R. 1981. Vergleich der wirkung von schadstoffen auf flagellate sowie ciliate bzw auf holozoische bakterienfressende sowie saprozoische protozoen. Gwf-Wasser/Abwasser 122:308–313.

Broadmeadow A. 1988. Technical DNOC: Preliminary toxicity study by dietary administration to F-344 rats for six weeks. Unpublished report prepared for Pennwalt Corporation Agrichemicals Division by Life Science Research, Eye, U.K. (Life Science Research Study No. 87/PTN 001/433) [cited in IPCS 2000].

Broadmeadow A. 1991. Technical DNOC: Combined oncogenicity and toxicity study by dietary administration to F-344 rats for 104 weeks. Unpublished report prepared for Pennwalt Corporation by Life Science Research, Eye, U.K. (Life Science Research Study No. PTN/003/DNOC) [cited in IPCS 2000].

Brouwer ET, Brinkman UA. 1994. Determination of phenolic compounds in surface water using on-line liquid chromatographic precolumn-based column switching techniques. J Chromatogr 678:223–231.

Buccafusco RJ, Ells SJ, LeBlanc GA. 1981. Acute toxicity of priority pollutants to bluegill (*Lepomis macrochirus*). Bull Environ Contam Toxicol 26(4):446–452 [cited in US EPA 1986a].

Burkatskaya EN. 1965a. [Maximum permissible concentration of dinitro-*o*-cresol in air.] Gig Sanit 30:34–37 (in Russian) [cited in IPCS 2000].

Burkatskaya EN. 1965b. The toxicity of dinitro-*ortho*-cresols for warm-blooded animals and problems of industrial hygiene in its application. Gig Tr Prof Zabol 9(4):56–57 [cited in IPCS 2000].

Callahan MA, Slimak MW, Gabel NW, May IP, Fowler CF, Freed JR, Jennings P, Durfee RL, Whitmore FC, Maestri B, et al. 1979. Water-related environmental fate of 129 priority pollutants. Vol. 2. Washington (DC): Office of Water and Waste Management and Office of Water Planning and Standards, United States Environmental Protection Agency. (EPA-440/4-79-029b; PB-204381).

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://canadagazette.gc.ca/partIII/1999/g3-02203.pdf

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. 143, no. 7, p. 607-612. Ottawa: Queen's Printer. Available from: http://canadagazette.gc.ca/partII/2000/20000329/pdf/g2-13407.pdf

Capel PD, Larson SJ. 1995. A chemodynamic approach for estimating losses of target organic chemicals from water during sample holding time. Chemosphere 30(6):1097-1107.

Cardellicchio N, Cavalli S, Piangerelli V, Giandomenico S, Ragone P. 1997. Determination of phenols in environmental samples by liquid chromatography–electrochemistry. Fresenius J Anal Chem 358(6):749–754.

[CEMC] Canadian Environmental Modelling Centre. 2002. Level III fugacity model version 2.7. Version dated March 2002. Peterborough (ON): Trent University. [cited 2004 March]. Available from: http://www.trentu.ca/cemc/models/L3270.html

ChemFinder. [cited 2003 Sept 17]. Available from: http://www.chemfinder.com

City of Toronto. 1990. The quality of drinking water in Toronto: A review of tap water, bottled water and water treated by a point-of-use device. Summary report. Toronto (ON): City of Toronto, Department of Public Health.

City of Toronto Water and Wastewater Services Division. 2002a. Water quality quarterly report—October–December 2002 [Internet]. City of Toronto. Available from: http://www.city.toronto.on.ca/water/quality_report/index.htm

City of Toronto Water and Wastewater Services Division. 2002b. Water quality quarterly report—July–September 2002 [Internet]. City of Toronto. Available from: http://www.city.toronto.on.ca/water/quality_report/index.htm

City of Toronto Water and Wastewater Services Division. 2002c. Water quality quarterly report—April–June 2002 [Internet]. City of Toronto. Available from: http://www.city.toronto.on.ca/water/quality_report/index.htm

City of Toronto Water and Wastewater Services Division. 2002d. Water quality quarterly report—January–March 2002 [Internet]. City of Toronto. Available from: http://www.city.toronto.on.ca/water/quality_report/index.htm

Coles RJ, Brooks PN. 1997. Technical DNOC: dietary two generations reproduction study in the rat. Unpublished report prepared for Elf Atochem Agri SA by Safepharm Laboratory, Derby, U.K. (Safepharm Laboratory Project No. 764/010) [cited in IPCS 2000].

Danish Environmental Protection Agency. 2001. Pesticides in air and in precipitation and effects on plant communities [Internet]. Pesticides Research No. 57. Available from: http://www.mst.dk/udgiv/publications/2001/87-7944-929-8/html/helepubl_eng.htm.

Davi ML, Gnudi F. 1999. Phenolic compounds in surface water. Water Res 14:3213–3219.

Den Tonkelaar EM, Van Leeuwen FZR, Kuiper C. 1983. Semichronic toxicity of DNOC in the rat. Meded Fac Landbouwwet Rijksuniv Gent 48(4):1015–1022 [cited in Health Canada 2004].

DeVault DS. 1985. Contaminants in fish from Great Lakes harbors and tributary mouths. Arch Environ Contam Toxicol 14:587–594 [cited in Health Canada 2004].

Dey-Hazra A, Heisler E. 1981. [Acute toxicity of trifocide 50% flowable, 50% DNOC ammonium salt in water by inhalation in the rat.] Unpublished report prepared for Elf Atochem Agri bv by Pharmatox, Hamburg, Germany (Pharmatox Study No. 1-4-246-81) (in German) [cited in IPCS 2000].

Dickhaus S, Heisler E. 1980. Acute toxicity of technical active substance DNOC (99 ± 1%) after oral administration to quail. Pharmatox, Hanover, Germany (Pharmatox Study No. 1-8-239-80; unpublished report prepared for Ruhr-Stickstof AG) [cited in IPCS 2000].

Dickhaus S, Heisler E. 1984. [Teratogenic/embryotoxic study with the product "trifocide liquid 50%" following oral administration in the rat.] Unpublished report prepared for Ruhr-Stickstoff AG by Pharmatox, Hamburg, Germany (Pharmatox Study No. 2-4-240-83) (in German) [cited in IPCS 2000].

Dodds EC, Robertson JD. 1933. The clinical applications of dinitro-o-cresol. Lancet ii:1137-1139.

Dow Chemical Co. 1940. Initial submission: Toxicity and health hazards of 2,4-dinitrophenol, dinitro-*ortho*-cresol and dinitro-*ortho*-cyclohexyl phenol (final report) with letter dated 18 March 1992 (sanitized). Submitted (1992) to United States Environmental Protection Agency (microfiche no. OTS0536148; document identification no. 88-920001472S).

Dow Chemical Co. 1950. Initial submission: The comparative acute oral toxicity of several dinitrophenols used in agriculture (final report) with cover letter dated 18 March 1992 (sanitized). Submitted (1992) to United States Environmental Protection Agency (microfiche no. OTS0536145; document identification no. 88-920001469S).

Dow Chemical Co. 1992. Letter submitting multiple studies on multiple chemicals required for docket OPTS-82036 with attachments (sanitized). Submitted (1992) to United States Environmental Protection Agency (Microfiche no. OTS0535413; Document Identification no. 88-920000855S).

Driscoll R. 1995a. Technical DNOC: Acute oral toxicity test in the rat. Unpublished report prepared for Elf Atochem Agri SA by Safepharm Laboratories, Derby, U.K. (Safepharm Laboratories Study No. 765/4) [cited in IPCS 2000].

Driscoll R. 1995b. Technical DNOC: Acute dermal toxicity (limit test) in the rat. Unpublished report prepared for Elf Atochem Agri SA by Safepharm Laboratories, Derby, U.K. (Safepharm Laboratories Study No. 764/5) [cited in IPCS 2000].

Duyzer J, Vonk VW. 2002. Deposition of persistent organic compounds and pesticides to water surfaces in the Netherlands. In: Mindgley PM, Reuther M (editors). Transport and chemical transformation in the troposphere: Proceedings of EUROTRAC Symposium 2002, Garmisch-Partenkirchen, Germany, March 11–15, 2002. p. 1–4.

Environment Canada. 1990. Quantity and use pattern information relating to the Domestic Substances List 1984–1986. Data collected with respect to subsection 25(1) (CEPA 1988) and following procedures stated in Environment Canada. 1988. Reporting for the Domestic Substances List. Ministry of Supply and Services (DSS Cat. No. En 40-364/1988E).

Environment Canada, 2001. *Canadian Environmental Protection Act, 1999*: *Notice with respect to certain substances on the Domestic Substances List (DSL).*Canada Gazette, Part I, vol. 135, no. 46, p. 4194–4210. Available from: http://canadagazette.gc.ca/partI/2001/20011117/pdf/g1-13546.pdf

Environment Canada. 2003a. Phenol, 2-methyl-4,6-dinitro- (CAS RN 534-52-1). Preliminary report of section 71 (CEPA 1999). Notice with respect to certain substances on the Domestic Substances List (DSL). February 2003. Existing Substances Branch, Environment Canada. p. 13.

Environment Canada. 2003b. National Pollution Release Inventory. Multi-year summary.

[Environment Canada. 2003c. ChemSim. Chemical release and dispersion analysis application. Developed by Canadian Hydraulics Centre, National Research Council of Canada, Ottawa, Ontario.

[EQC] Equilibrium Criterion Model. 2003. Version 2.02. Peterborough (ON): Trent University, Canadian Environmental Modelling Centre. [cited 2003 Oct 17]. Available from: http://www.trentu.ca/academic/aminss/envmodel/models/EQC2.html.

Fellows M. 1998. DNOC technical measurement of unscheduled DNA synthesis in rat liver using an *in vivo/in vitro* procedure. Unpublished report prepared for Elf Atochem Agri SA by Covance Laboratories Ltd., North Yorkshire, U.K. (Report No. 160711-DS140) [cited in IPCS 2000].

Fowler BR, Hope D. 1984. Detailed organic analysis of surficial sediment from abandoned artificial petroleum exploration islands in the Beaufort Sea. Prepared for the Environmental Protection Service, Environment Canada, Yellowknife, N.W.T., by Arctic Laboratories Ltd., Inuvik, N.W.T. 52 pp. + appendices.

Garner RC. 1984. Study to evaluate the chromosome damaging potential of DNOC by its effects on cultured Chinese hamster ovary cells using an *in vitro* cytogenetics assay. Unpublished report prepared for Pennwalt Holland bv by Microtest Research, York, U.K. (Microtest Research Study No. PHARM 1/CYT/RCG 2) [cited in IPCS 2000].

Ghillebaert F, Chaillou C, Deschanps F, Roubaud P. 1995. Toxic effects, at three pH levels, of two reference molecules on common carp embryo. Ecotoxicol Environ Saf 32:19–28.

Gobas FAPC, Arnot J. 2003. Categorization of organic substances on the Domestic Substances List for bioaccumulation potential. Prepared for Existing Substances Branch, Environment Canada. June 15, 2003. 109 pp.

Grilli S, Ancora G, Valenti AM, Mazzullo M, Colacci A. 1991. *In vivo* unwinding fluorometric assay as evidence of the damage induced by fenarimol and DNOC in rat liver DNA. J Toxicol Environ Health 34:485–494 [cited in IPCS 2000].

Hallberg GR. 1989. Pesticide pollution of groundwater in the humid United States. Agric Ecosyst Environ 26:299–367 [cited in IPCS 2000].

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.

Health Canada. 2004. Screening health assessment supporting working document, dinitro-*o*-cresol (CAS 534-52-1). Draft. April 22, 2004.

Howard P, Boethling R, Jarvis W. 1991. Handbook of environmental degradation rates. Boca Raton (FL): Lewis Publishers.

Hrelia P, Vigagni F, Maffei F, Morotti M, Colacci A, Perocco P, Griili S, Cantelli-Forti G. 1994. Genetic safety evaluation of pesticides in different short-term tests. Mutat Res 321:219–228 [cited in IPCS 2000].

[HSDB] Hazardous Substances Data Bank [database on the Internet]. 2000. [cited 2003 Oct 17] Bethesda (MD): U.S. National Library of Medicine, National Center for Biotechnology Information: Available from: http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen?HSDB

[IPCS] International Programme on Chemical Safety. 2000. Dinitro-*ortho*-cresol. World Health Organization, Geneva. 70 pp. (Environmental Health Criteria 220).

Janda J. 1970. On the toxicity of DNOC to pheasants, partridges and hares. Sci Agric Bohem 2(4): 301-312 [cited in IPCS 2000]

Jongerius O and Jongeneelen FJ. 1991. Criteria document for an occupational exposure limit value of 4,6-dinitro-*o*-cresol (CAS 534-52-1). Industrial Medicine and Hygiene Unit, Health and Safety Directorate, Directorate General Employment, Industrial Relations and Social Affairs, Commission of the European Communities, Luxembourg (SEG/CDO/29.1992) [cited in IPCS 2000].

Kelly J. 1995. DNOC: 13 week oral (dietary administration) range-finding study in the mouse. Unpublished report prepared for Elf Atochem Agri SA by Corning Hazelton, Harrogate, UK (Corning Hazelton Project No. CHE 1151/8) [cited in IPCS 2000].

King E and Harvey DG. 1953a. Some observations on the absorption and excretion of 4,6-dinitro-*o*-cresol (DNOC). I. Blood dinitro-*o*-cresol levels in the rat and the rabbit following different methods of absorption. Biochem J 53:185–195 [cited in IPCS 2000].

King E and Harvey DG. 1953b. Some observations on the absorption and excretion of 4,6-dinitro-*o*-cresol. Biochem J 53:196–200 [cited in IPCS 2000].

Kirkland DJ. 1984. Study to evaluate the chromosome damaging potential of DNOC by its effects on the bone marrow cells of treated rats. Unpublished report prepared for Pennwalt Holland bv by Pharmatox, Hanover, Germany (Pharmatox Study No. PHM 6/RBM/AR/KF6) [cited in IPCS 2000].

Kirkland DJ. 1986. Study to evaluate the chromosome damaging potential of 4,6-dinitro-*o*-cresol (DNOC) by its effects on the bone marrow cells of treated mice. Unpublished report prepared for Pennwalt Holland bv by Microtest Research, York, UK (Microtest Research Study No. PEN 1/MBM/KF27/MB1) [cited in IPCS 2000].

Kühn R, Pattard M, Pernak K-D, Winter A. 1989. Results of the harmful effects of water pollutants to *Daphnia magna* in the 21 day reproduction test. Water Res 23(4):501–510.

Leuenberger D, Czuczwa J, Tremp J, Giger W. 1988. Nitrated phenols in rain: atmospheric occurrence of phytotoxic pollutants. Chemosphere 17(3):511–515.

Lüttke J, Levsen K. 1997. Phase partitioning of phenol and nitrophenols in clouds. Atmos Environ 31(16):2649–2655.

Lüttke J, Levsen K, Acker K, Wieprecht W, Möller D. 1999. Phenols and nitrated phenols in clouds at Mount Brocken. Int J Environ Anal Chem 74(1–4):69–89.

Mabey WR, Smith JH, Podoll RT. 1981. Aquatic fate process data for organic priority pollutants. Washington (DC): United States Environmental Protection Agency. EPA 440/4-81-014.

Martin CN. 1981. Study to determine the ability of DNOC to induce mutations to ouabain and 6-thioguanine resistance in mouse lymphoma L5178Y cells. Unpublished report prepared for Pennwalt Holland bv by Microtest Research, York, UK (Microtest Research Study No. PHARM 2/ML/JC/JGL) [cited in IPCS 2000].

Marzin D. 1991a. Recherche de mutagénicité sur *Salmonella typhimurium his* — selon la technique de B.N. Ames sur le produit dinitro-*ortho*-crésol. Unpublished report prepared for Elf Atochem Agri SA by Institut Pasteur de Lille, Lille, France (Institut Pasteur de Lille Study No. IPL-R 910901) [cited in IPCS 2000].

Marzin D. 1991b. Etude de mutagenèse au locus HPRT sur cellules V79 de hamster chinois (résistance à la 6-thioguanine) sur le produit dinitro-*ortho*-crésol (2-méthyl-4,6-dinitrophénol). Unpublished report prepared for Elf Atochem Agri SA by Institut Pasteur de Lille, Lille, France (Institut Pasteur de Lille Study No. IPL-R910904) [cited in IPCS 2000].

Marzin D. 1991c. Etude de l'activité génotoxique par la technique du micronucléus chez la souris sur le produit dinitro-*ortho*-crésol. Unpublished report prepared for Elf Atochem Agri SA by Institut Pasteur de Lille, Lille, France (Institut Pasteur de Lille Study No. IPL-R 910804) [cited in IPCS 2000].

Marzin D. 1991d. Etude de l'activité génotoxique du produit dinitro-*ortho*-crésol par la recherche d'aberrations chromosomiques par analyse de métaphases sur lymphocytes humains en culture. Unpublished report prepared for Elf Atochem Agri SA by Institut Pasteur de Lille, Lille, France (Institut Pasteur de Lille Study No. IPL-R 911010) [cited in IPCS 2000].

Mayer RL Jr, Ellersieck MR. 1986. Manual of acute toxicity: interpretation and data base for 410 chemicals and 66 species of freshwater animals. Washington (DC): Fish and Wildlife Service, United States Department of the Interior. 508 pp. (Resource Publication 160).

Migaszewski ZM. 1999. Determining organic compounds in soils and vegetation of the Holy Cross Mtns, Poland. Water Air Soil Pollut 111(1–4):123–138.

McGirr JL and Papworth DS. 1953. Toxic hazards of the newer insecticides and herbicides. Vet Rec 65(48):857–862 [cited in IPCS 2000].

Mogensen BB, Spliid NH. 1995. Pesticides in Danish watercourses: occurrence and effects. Chemosphere 31(8):3977–3990.

Muller J and Haberzetti R. 1980. Mutagenicity of DNOC in *Drosophila melanogaster*. Arch Toxicol Suppl 4:59–61 [cited in IPCS 2000].

Munro JR, Foster MG, Pawson T, Stelzig A, Tseng T, King L. 1985. St. Clair River point source survey 1979–1980. Toronto (ON): Ontario Ministry of the Environment; and Ottawa (ON): Environment Canada. 194 pp.

[NCI] National Chemical Inventories [database on CD-ROM]. 2006. Columbus (OH): American Chemical Society. Available from: http://www.cas.org/products/cd/nci/index.html

Nehéz M, Selypes A and Paldy A. 1977. [Examination of dinitro-*o*-cresol-containing fertilizer for mutagenic effects.] Egeszsegtudomany 21: 237–243 (in Hungarian) [cited in IPCS 2000].

Nehéz M, Selypes A, Paldy A and Berencsi G. 1978. The mutagenic effect of a dinitro-*o*-cresol containing pesticide on mice germ cells. Ecotoxicol Environ Saf 2:401–405 [cited in IPCS 2000].

Nehéz M, Paldy A and Selypes A. 1981. The teratogenicity and mutagenicity effects of dinitro-*o*-cresol-containing herbicide on laboratory mouse. Ecotoxicol Environ Saf 5:38–44 [cited in IPCS 2000].

Nehéz M, Selypes A, Mazzag E and Berencsi G. 1984. Additional data on the mutagenic effect of dinitro-*o*-cresol containing herbicides. Ecotoxicol Environ Saf 8: 75–79 [cited in IPCS 2000].

Nishimura N, Nishimura H and Oshima H. 1982. Survey on mutagenicity of pesticides by the *Salmonella* microsome test. J Aichi Med Univ Assoc 10(4):305–312 [cited in IPCS 2000].

Nojima K, Fukaya K, Fukui S. 1976. Studies of photochemistry of aromatic compounds. Chemosphere 2:25–30.

NOVA Chemicals Corporation. 2007. Comments on publication after screening assessment of a substance - 4,6-dinitro-*o*-cresol (CAS No. 534-52-1) [subsection 77(1) of the *Canadian Environmental Protection Act, 1999*]. 2007-06-23 - Canada Gazette Part I, Vol. 141 No. 25.

[NPRI] National Pollutant Release Inventory [database on the Internet]. 2006. Gatineau (QC); Environment Canada. [cited 2003 10 20] Available from: http://www.ec.gc.ca/pdh/querysire/query_e;cfm

[OMEE] Ontario Ministry of Environment and Energy. 1994. Ontario typical range of chemical parameters in soil, vegetation, moss bags and snow. Toronto (ON): Queen's Printer for Ontario. (ISBN 0-7778-1979-1).

PALLAS. Version 4.0. for Windows. CompuDrug Chemistry Ltd.

[PAN] Pesticide Action Network North America Pesticides Database [database on the Internet]. 2004. DNOC, sodium salt — Aquatic ecotoxicity. [cited 2004 April 6]. Available from: http://www.pesticideinfo.org/Detail_Chemical.jsp?Rec_Id=PC33294

Paterson B, Cowie CE, Jackson PE. 1996. Determination of phenols in environmental waters using liquid chromatography with electrochemical detection. J Chromatogr 731(1–2):95–102.

Pietsch J, Schmidt W, Sacher F, Fichtner S, Brauch H-J. 1995. Pesticides and other organic micro pollutants in the river Elbe. Fresenius J Anal Chem 353:75–82.

Plotz M. 1936. Dinitro-ortho-cresol, A metabolic stimulator and its toxic side-actions. NY State J Medicine 41:266-268.

[PMRA] Pest Management Regulatory Agency. 2009. Communication from Robert Martin, Regulatory Information Officer, Ottawa, Ont. October 7, 2009.

[PhysProp] Interactive PhysProp Database [databse on the Internet]. 2006. Syracuse (NY): Syracuse Research Corporation.. [cited 2003 Oct 17]. Available from: http://www.syrres.com/esc/physdemo.htm.

Quinto I, DeMarinis E, Mallardo M, Arcucci A, Della Morte R and Staiano N. 1989. Effect of DNOC, ferbam and imidan exposure on mouse sperm morphology. Mutat Res 224:405–408 [cited in IPCS 2000].

Richartz H, Reischl A, Trautner F, Hutzinger O. 1990. Nitrated phenols in fog. Atmos Environ 24A(12):3067–3071.

Römpp A, Klemm O, Frank H. 2001. Haloacetates and nitrophenols in fog and rain. In: Midgley PM, Reuther M, Williams M (editors). Transport and chemical transformation in the troposphere. Proceedings of the 6th EUROTRAC Symposium, Garmisch-Partenkirchen, Germany, March 27–31, 2000. Berlin (DE): Springer. p. 908–911.

Sample BE, Opresko DM, Suter GW II. 1996. Toxicological benchmarks for wildlife: 1996 revision [Internet]. Prepared by the Risk Assessment Program, Health Sciences Research Division, United States Department of Energy, Oak Ridge, Tennessee (ES/ER/TM-86/R3). Available from: http://www.esd.ornl.gov/programs/ecorisk/documents/tm86r3.pdf.

Schmidt,M. 1970. [Studies of DNOC residues in potatoes after destruction with Hedolit concentrate.] Nachrichtenbl Dtsch Pflanzenschutzdienstes (Braunschweig) 24(11):222–224 (in German) [cited in NLM 2000].

Schüssler W, Nitschke L. 2001. Nitrophenols in precipitation. Chemosphere 42:277–283.

Schwarzenbach RP, Stierli R, Folsom BR, Zeyer J. 1988. Compound properties relevant for assessing the environmental partitioning of nitrophenols. Environ Sci Technol 22(1):83–92.

Schwarzenbach RP, Gschwend PM, Imboden DM. 2003. Environmental organic chemistry. 2nd edition. Hoboken (NJ): John Wiley and Sons,.

Sewell IG, Mead C, Bartlett AJ. 1995a. Technical DNOC. Algal inhibition test. Safepharm Laboratories, Derby, U.K. (Study No. 764/15; unpublished report prepared for ELF Atochem Agri SA).

Sewell IG, Mead C, Bartlett AJ. 1995b. Technical DNOC. Acute toxicity to bluegill sunfish (*Lepomis macrochirus*). Safepharm Laboratories, Derby, U.K. (Study No. 764/12; unpublished report prepared for ELF Atochem Agri SA).

Sewell IG, Mead C, Bartlett AJ. 1995c. Technical DNOC. Acute toxicity to rainbow trout (*Onchorhynchus mykiss*). Safepharm Laboratories, Derby, U.K. (Study No. 764/13; unpublished report prepared for ELF Atochem Agri SA).

Sloof W, Canton JH. 1983. Comparison of the susceptibility of 11 freshwater species to 8 chemical compounds. II. (Semi)chronic toxicity tests. Aquat Toxicol 4(3):271–281.

Somani SM, Schaeffer DJ and Mack JO. 1981. Quantifying the toxic and mutagenic activity of complex mixtures with *Salmonella typhimurium*. J Toxicol Environ Health 7:643–653 [cited in IPCS 2000].

Spencer HC, Rowe VK, Adams EM and Irish DD. 1948. Toxicological studies on laboratory animals of certain alkyldinitrophenols used in agriculture. J Ind Hyg Toxicol 30(1):10–25 [cited in IPCS 2000].

Spliid NH and Koppen B. 1998. Occurrence of pesticides in Danish shallow ground water. Chemosphere 37(7):1307–1316.

STP Model. [sewage treatment plant removal model]. 2001. Version 1.5. Peterborough (ON): Trent University., Canadian Environmental Modelling Centre. [cited 2005 March]. Available from: http://www.trentu.ca/academic/aminss/envmodel/models/VBSTP.html

Strube K, Janke D, Kappler R, Kristen U. 1991. Toxicity of some herbicides to *in vitro* growing tobacco pollen tubes (the pollen test). Environ Exp Bot 31(2):217–222.

Sundvall A, Marklund H and Rannung U. 1984. The mutagenicity of *Salmonella typhimurium* of nitrobenzoic acids and other wastewater components generated in the production of nitrobenzoic acids and nitrotoluenes. Mutat Res 137:71–78 [cited in IPCS 2000].

Takahashi K, Kikuta M, Aoyama H and Teramoto S. 1999. Appearance of morphologically abnormal sperm in the caput, corpus, and cauda epididymis of male rats after treatment with 4,6-dinitro-*o*-cresol. J Toxicol Sci 24(4):350.

Til HP. 1980. Sub-chronic (90-day) oral toxicity study with DNOC in dogs. Unpublished report prepared for Pennwalt Holland bv by CIVO TNO Institute, Zeist, Netherlands (CIVO TNO Institute Study No. B 80/0359) [cited in IPCS 2000].

Til HP, Kengen MTF. 1980. Subacute (8-day) dietary LC_{50} study with DNOC in Japanese quail. CIVO-TNO, Zeist, The Netherlands (Study No. R 6596; unpublished report prepared for Pennwalt Holland bv) [cited in IPCS 2000].

Tremp J, Mattrell P, Fingler S, Giger W. 1993. Phenols and nitrophenols as tropospheric pollutants: emissions from automobile exhausts and phase transfer in the atmosphere. Water Air Soil Pollut 68:113–123.

Tuxen N, Tüchsen PL, Rügge K, Albrechtsen H-J, Bjerg PL. 2000. Fate of seven pesticides in an aerobic aquifer studied in column experiments. Chemosphere 41:1485-1494.

[UNEP/FAO] United Nations Environment Programme / Food and Agriculture Organization of the United Nations. 2002. DNOC (dinitro-*ortho*-crésol). Document d'orientation des décisions. Secrétariat provisoire de la Convention de Rotterdam sur la procédure de consentement préalable en connaissance de cause applicable à certains produits chimiques et pesticides dangereux qui font l'objet d'un commerce international. 32 pp.

[US EPA] United States Environmental Protection Agency. 1986a. Health and environmental effects profile for dinitrocresols. Cincinnati (OH): Environmental Criteria and Assessment Office, Office of Health and Environmental Assessment, Office of Research and Development, United States Environmental Protection Agency. 76 pp. (EPA/600/X-86/197).

[US EPA] United States Environmental Protection Agency. 1986b. Computer printout: Graphic Exposure Modeling System (GEMS) EXAMS model. Washington (DC): Office of Toxic Substances, United States Environmental Protection Agency. [cited in US EPA 1986a].

[[US EPA] United States Environmental Protection Agency. 1993. Wildlife exposure factors handbook. Volume 1 [Internet]. Washington (DC): National Center for Environmental Assessment, United States Environmental Protection Agency. (EPA/600/R-93/187). Available from http://www.epa.gov/ncea/pdfs/toc2-37.pdf.

van der Hoeven JCM. 1984. Assessment of the effects of 4,6-dinitro-*o*-cresol (DNOC) on the reproduction of *Daphnia magna.* Notox Toxicological Research and Consultancy, sHertogenbosch, The Netherlands (unpublished report prepared for Pennwalt Holland).

van der Hoeven JCM. 1992. Acute toxicity of DNOC technical to the worm species *Eisenia fetida*. TNO Institute, Delft, The Netherlands (TNO Study No. R91/324; unpublished report prepared for Elf Atochem Agri SA) [cited in IPCS 2000].

Verschueren K. 2001. Handbook of environmental data on organic chemicals. 4th edition. New York (NY): John Wiley and Sons. pp. 958–961.

Vogue PA, Kerle EA, Jenkins JJ. 1994. OSU extension pesticide properties database. [cited 2004 March 18]. Available from http://npic.orst.edu/ppdmove.htm

Vos JG, Karjnc EL, Beekhof PK and van Logten MJ. 1983. Methods for testing immune effects of toxic chemicals: evaluation of the immunotoxicity of various pesticides in the rat. In: Miyamoto J (ed.), IUPAC pesticide chemistry: human welfare and the environment. Pergamon Press, Kyoto, Japan. pp. 497–504 [cited in IPCS 2000].

Webber MD. 1994. Industrial organic compounds in selected Canadian municipal sludges and agricultural soils. Burlington (ON): Wastewater Technology Centre, Rockcliffe Research Management Inc. 100 pp.

Webber MD, Lesage S. 1989. Organic contaminants in Canadian municipal sludges. Waste Manage Res 7:63–82.

Webber MD, Nichols JA. 1995. Organic and metal contaminants in Canadian municipal sludges and a sludge compost. Burlington (ON): Wastewater Technology Centre, Rockcliffe Research Management Inc. 168 pp.

Zahn R, Wellens H. 1980. Prüfung der biologischen Abbaubarkeit im Standversuch – weitere Erfahrungen und neuere Einsatzmöglichkeiten. Zeitschrift Wasser Abwasser Forschung 13(1):1-7.

Zitko V, McLeese DW, Carson WG, Welch HE. 1976. Toxicity of alkyldinitrophenols to some aquatic organisms. Bull Environ Contam Toxicol 16(5):508–515.

Appendix 1. Upper-bounding estimates of daily intake of DNOC by the general population in Canada

Route of exposure	Estimated intake (µg/kg-bw per day) of DNOC by various age groups						
	0–6 months[1]		0.5–4 years[3]	5–11 years[4]	12–19 years[5]	20–59 years[6]	60+ years[7]
	Formula fed[2]	Not formula fed					
Air[8]	1.4×10^{-2}		3.0×10^{-2}	2.4×10^{-2}	1.4×10^{-2}	1.1×10^{-2}	9.9×10^{-3}
Drinking water[9]	4.3×10^{-2}	1.6×10^{-2}	1.8×10^{-2}	1.4×10^{-2}	8.1×10^{-3}	8.5×10^{-2}	8.9×10^{-3}
Food[10]		NA[11]	NA	NA	NA	NA	NA
Soil[12]	4.0×10^{-4}		6.5×10^{-4}	2.1×10^{-4}	5.1×10^{-5}	4.2×10^{-5}	4.2×10^{-5}
Total intake	5.7×10^{-2}	3.0×10^{-2}	4.9×10^{-2}	3.8×10^{-2}	2.1×10^{-2}	2.0×10^{-2}	1.9×10^{-2}

[1] Assumed to weigh 7.5 kg, to breathe 2.1 m^3 of air per day, to drink 0.8 L of water per day (formula fed) or 0.3 L/day (not formula fed), and to ingest 30 mg of soil per day (Health Canada 1998).

[2] For formula-fed infants, intake from water is synonymous with intake from food. No data on concentrations of DNOC in formula were identified for Canada.

[3] Assumed to weigh 15.5 kg, to breathe 9.3 m^3 of air per day, to drink 0.7 L of water per day, and to ingest 100 mg of soil per day (Health Canada 1998).

[4] Assumed to weigh 31.0 kg, to breathe 14.5 m^3 of air per day, to drink 1.1 L of water per day, and to ingest 65 mg of soil per day (Health Canada 1998).

[5] Assumed to weigh 59.4 kg, to breathe 15.8 m^3 of air per day, to drink 1.2 L of water per day, and to ingest 30 mg of soil per day (Health Canada 1998).

[6] Assumed to weigh 70.9 kg, to breathe 16.2 m^3 of air per day, to drink 1.5 L of water per day, and to ingest 30 mg of soil per day (Health Canada 1998).

[7] Assumed to weigh 72.0 kg, to breathe 14.3 m^3 of air per day, to drink 1.6 L of water per day, and to ingest 30 mg of soil per day (Health Canada 1998).

[8] Leuenberger et al. (1988) estimated an ambient air concentration of 0.05 µg/m^3 using measured concentrations of DNOC from a rainwater sample (15 nM) taken at Dübendorf, Switzerland, in 1985 and using a reference rain/air partition coefficient (5.6×10^4). Canadians are assumed to spend 3 hours outdoors each day (Health Canada 1998). Data available from which the critical data were selected included Tremp et al. (1993). In the absence of data, the estimated ambient air concentration (0.05 µg/m^3) was also used for indoor air. Canadians are assumed to spend 21 hours indoors each day (Health Canada 1998). Ambient air was assumed to be representative of exposure to indoor air, since there was no indication of additional sources of DNOC in indoor environments.

[9] The detection limit (0.4 µg/L) for DNOC in 19 samples of tap water from Toronto, Ontario, in 2002 was used as a surrogate for the level of DNOC in Canadian drinking water (City of Toronto Water and Wastewater Services Division, 2002a, 2002b, 2002c, 2002d). Data available from which the critical data were selected included Hallberg (1989), City of Toronto (1990), and Spliid and Koppen (1998).

[10] No quantitative data were identified for concentrations of DNOC in food items. A detection limit of 1000 µg/g was used for a study by Schmidt (1970) that measured DNOC in potatoes. However, this value was not used in the intake estimate due to the age of the study and because DNOC is not expected to contaminate foods based on its application method. Data available from which the critical data were selected included DeVault (1985).

[11] NA = not available.

[12] The Ontario Ministry of Environment and Energy (OMEE 1994) did not detect DNOC in 161 soil samples collected from Ontario. The method detection limit of 100 ng/g was used in the intake estimate as a surrogate for the level of DNOC in Canadian soil. Data available from which the critical data were selected included Webber (1994) and Migaszewski (1999).

Appendix 2. Summary of health effects information for DNOC

Endpoint	Lowest effect levels[1]/Results
Laboratory animals and *in vitro*	
Acute toxicity	Lowest **oral LD_{50}** = 16 mg/kg-bw (Jongerius and Jongeneelen 1991) (range: 16 mg/kg-bw to 100 mg/kg-bw) [Additional studies: Dow Chemical Co. 1940; Ambrose 1942; Spencer et al. 1948; Dow Chemical Co. 1950; King and Harvey 1953a; McGirr and Papworth 1953; Burkatskaya 1965b; Ben Dyke et al. 1970; Dow Chemical Co. 1992; Driscoll 1995a] Lowest **dermal LD_{50}** = 187 mg/kg-bw (Arustamyn 1972) (range: 187 mg/kg-bw to > 2000 mg/kg-bw) [Additional studies: Dow Chemical Co. 1940; Spencer et al. 1948; Burkatskaya 1965b; Ben Dyke et al. 1970; Jongerius and Jongeneelen 1991; Dow Chemical Co. 1992; Driscoll 1995b] Lowest **inhalation LC_{50}** = 40 mg/m^3 (Burkatskaya 1965a) (range: 40 mg/m^3 to 230 mg/m^3) [Additional studies: King and Harvey 1953b; Dey-Hazra and Heisler 1981]
Short-term repeated-dose toxicity	Lowest **oral (diet) LOEL** (rat) = 7.24 mg/kg-bw per day: decreased body weight gain (6-week study) (Broadmeadow 1988) [Additional studies: Dow Chemical Co. 1940,; Spencer et al. 1948; Quinto et al. 1989; Dow Chemical Co. 1992; Takahashi et al. 1999] Lowest **inhalation LOEC** (cat) = 2 mg/m^3: mortality (30-day study) (Burkatskaya 1965a)
Subchronic toxicity	Lowest **oral (diet) LOEL** (rat) = 2.5 mg/kg-bw per day: change in blood pyruvate and thyroid hormone levels (13-week study) (Den Tonkelaar et al. 1983) [Additional studies: Til 1980; Kelly 1995] Lowest **inhalation NOEC** (cat) = 0.2 mg/m^3: "no severe adverse effects" (90-day study) (Burkatskaya 1965a)
Chronic toxicity/ carcinogenicity	Lowest **oral (diet) non-neoplastic LOEL** (male rat) = 4.12 mg/kg-bw per day: increased food consumption (104-week study) (Broadmeadow 1991) No increase in tumour incidence was observed at dose levels up to 5 mg/kg-bw per day in a 104-week study using rats exposed through the diet (Broadmeadow 1991). [N.B.: It is not clear based on the secondary account of this study if the substance was tested up to the maximum tolerated dose.]
Genotoxicity and related endpoints: *in vivo*	**Positive**: mouse, bone marrow (micronuclei; 20 mg/kg-bw or 10 mg/kg-bw intraperitoneally [i.p.] after 1 year); rat, bone marrow (chromosomal aberrations; 7.5–30 mg/kg-bw i.p.); rat, hepatocytes (DNA unwinding; 1–9.3 mg/kg-bw i.p.); mouse (dominant lethal assay; 8–15 mg/kg-bw i.p.; and chromosomal aberration in F_1 embryo; 5–10 mg/kg-bw i.p.) (Nehéz et al.[1] 1978, 1981, 1984; Grilli et al. 1991; Hrelia et al. 1994) **Negative**: rat and mouse, bone marrow (chromosomal aberrations; 4–16 mg/kg-bw oral and 3–12 mg/kg-bw i.p., respectively); mouse, bone marrow (micronuclei; 20 mg/kg-bw i.p.); rat, hepatocytes (unscheduled DNA synthesis; 28–70 mg/kg-bw oral) (Kirkland 1984, 1986; Marzin 1991c; Fellows 1998)

[1] It was indicated in the IPCS (2000) review that studies by Nehéz et al. involved testing of a commercial product (Krezonit E) that contains 50% DNOC; therefore, results of these assays may relate to other components in the product.

Endpoint	Lowest effect levels[1]/Results
Genotoxicity and related endpoints: *in vitro*	**Positive**: *Proteus mirabilis* (DNA repair), *Salmonella typhimurium* TA98, TA100, TA1535, TA1537, TA1538 (mutagenicity), *Drosophila* (sex-linked recessive lethal), mouse lymphoma (mutagenicity), human lymphocytes (chromosome damage), Chinese hamster V79 cells (mutagenicity) (Adler et al. 1976; Nehéz et al. 1977, 1978; Muller and Haberzetti 1980; Martin 1981; Nishimura et al. 1982; Sundvall et al. 1984; Marzin 1991a, 1991b) **Negative**: *S. typhimurium* TA98, TA100, TA100NR, TA1535, TA1537 (mutagenicity), mouse lymphoma (mutagenicity), human lymphocytes (chromosome damage, sister chromatid exchange and unscheduled DNA synthesis), Chinese hamster ovary cells (chromosome damage) (Martin 1981; Somani et al. 1981; Nishimura et al. 1982; Garner 1984; Sundvall et al. 1984; Marzin 1991a, 1991b, 1991d; Hrelia et al. 1994)
Developmental toxicity	Lowest **oral (gavage) LOEL** (rabbit) = 25 mg/kg-bw per day: external or visceral malformations or skeletal variations, including microphthalmia or anophthalmia and hydrocephaly or microcephaly (gestation days 6–18) (Allen et al. 1990a) [Additional studies: Nehéz et al. 1981; Dickhaus and Heisler 1984] Lowest **dermal LOEL** (rabbit) = 30 mg/kg-bw per day: total resorptions in two females (gestation days 6–18) (Allen et al. 1990b)
Reproductive toxicity	Lowest **oral (diet) LOEL** (rat) = 1.73–2.24 mg/kg-bw per day: decreased group mean litter size in F_0 generation on days 14 and 21 of lactation (two-generation reproductive study) (Coles and Brooks 1997)
Immunotoxicity	Highest **oral (diet) NOEL** (rat) = 20 mg/kg-bw per day (3-week study) (Vos et al. 1983)
Humans	
Clinical study	Increase in basal metabolic rate and symptoms of toxicity (sweating, lethargy, headache, altered sleep patterns) at 3 mg/kg-bw for "several" days. Slight increase in basal metabolic rate but no symptoms of toxicity were noted in one patient administered 0.5 and then 1 mg/kg-bw per day for 39 days (data presented for two subjects, total number examined unclear) (Dodds and Robertson 1933) [Additional study: Plotz 1936]

[1] LC_{50} = the concentration estimated to be lethal to 50% of the organisms; LD_{50} = the dose estimated to be lethal to 50% of the organisms; LOEC = lowest-observed-effect concentration; LOEL = lowest-observed-effect level; NOEC = no-observed-effect concentration.