

Screening Assessment for the Challenge

Formamide

**Chemical Abstracts Service Registry Number
75-12-7**

**Environment Canada
Health Canada**

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Synopsis

The Ministers of the Environment and of Health have conducted a screening assessment of formamide, Chemical Abstracts Service Registry Number 75-12-7. The substance formamide was identified in the categorization of the *Domestic Substances List* as a high priority for action under the Ministerial Challenge. Formamide was identified as a high priority as it was considered to pose intermediate potential for exposure of individuals in Canada and had been classified by the European Commission on the basis of reproductive and developmental toxicity. The substance did not meet the ecological categorization criteria for persistence, bioaccumulation potential or inherent toxicity to aquatic organisms. Therefore, the focus of this assessment of formamide relates to human health risks.

Formamide may be emitted to the environment as a result of its use as an intermediate and solvent. It is used in the crystallization of pharmaceuticals, in soil stabilization, as a solvent in inks and as a component of liquid fertilizers. It is a monomer in the production of polymers, such as heat-resistant coatings and some personal care products.

Due to its primary use in industrial settings, the general population is not expected to be exposed to formamide. Exposure is possible from ink in marking pens, where it has been used as a solvent. However, the extent of use of formamide in marking pens in Canada is not known.

Based principally on the weight of evidence-based assessments of international or other national agencies, a critical effect for the characterization of risk to human health for formamide is carcinogenicity. In the standard 2-year carcinogenicity studies with rats and mice, induced tumours were observed in only one organ (liver), one sex (male) and one species (mice). Based on the weight of evidence of the available genotoxicity data, formamide is not considered to be mutagenic. Although the mode of induction of tumours has not been developed and elucidated, the tumours observed in the experimental animals are unlikely to have resulted from direct interaction with genetic material.

The non-cancer critical effects for characterization of risk to human health for formamide are reproductive, developmental and hematological toxicity. The exposures of the general population to formamide through environment media or consumer products are expected to be low. Comparison of the lowest effect levels for these non-cancer critical effects with the upper-bounding estimate of intake of formamide yields margins of exposure that are considered to be adequately protective for non-cancer effects.

On the basis of consideration of the existence of a practical threshold for non-mutagenic carcinogenicity of formamide in the animal studies and the magnitude of the margins of exposure for non-cancer effects, it is concluded that formamide should be considered as a substance that 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.

On the basis of ecological hazard and low reported releases of formamide, it is concluded that this substance 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. Formamide meets the criteria for persistence in air, but not in water, soils or sediment, and does not meet the criteria for bioaccumulation as set out in the *Persistence and Bioaccumulation Regulations*.

This substance will be included in the upcoming *Domestic Substances List* inventory update initiative. In addition and where relevant, research and monitoring will be undertaken to confirm assumptions used during the screening assessment.

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

Introduction

The *Canadian Environmental Protection Act, 1999* (CEPA 1999) (Canada 1999) requires the Minister of the Environment and the Minister of Health to conduct screening assessments of substances that have met the categorization criteria set out in the Act to determine whether these substances present or may present a risk to the environment or to human health. Based on the results of a screening assessment, the Ministers can propose to take no further action with respect to the substance, to add the substance to the Priority Substances List for further assessment or to recommend that the substance be added to the List of Toxic Substances in Schedule 1 of the Act and, where applicable, the implementation of virtual elimination.

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

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

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

The substance formamide was identified as a high priority for assessment of human health risk because it was considered to present IPE and had been classified by another agency on the basis of reproductive toxicity and developmental toxicity. The Challenge for formamide was published in the *Canada Gazette* on February 16, 2008 (Canada 2008). A substance profile was released at the same time. The substance profile presented the technical information available prior to December 2005 that formed the basis for categorization of this substance. As a result of the Challenge, submissions of information were received.

Although formamide was determined to be a high priority for assessment with respect to human health, it did not meet the criteria persistence in water, sediments or soil, or potential for bioaccumulation or inherent toxicity to aquatic organisms. Therefore, this assessment focuses principally on information relevant to the evaluation of risks to human health.

Under CEPA 1999, 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 the Act, where

64. [...] a substance is toxic if it is entering or may enter the environment in a quantity or concentration or under conditions that
- (a) have or may have an immediate or long-term harmful effect on the environment or its biological diversity;
 - (b) constitute or may constitute a danger to the environment on which life depends; or
 - (c) constitute or may constitute a danger in Canada to human life or health.

Screening assessments examine scientific information and develop conclusions by incorporating a weight of evidence approach and precaution.

This screening assessment includes consideration of information on chemical properties, hazards, uses and exposure, including the additional information submitted under the Challenge. Data relevant to the screening assessment of this substance were identified in original literature, review and assessment documents and stakeholder research reports and from recent literature searches, up to May 2009 for health effects and exposure. Key studies were critically evaluated; modelling results may have been used to reach conclusions. 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 (based principally on the weight of evidence assessments of other agencies that were used for prioritizing the substance). 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 existing critical information upon which the conclusion is based.

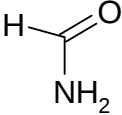
This screening assessment was prepared by staff in the Risk Assessment Bureau at Health Canada and the Existing Substances Program at Environment Canada and incorporates input from other programs within these departments. This assessment has undergone external written peer review/consultation. Comments on the technical portions relevant to human health were received from scientific experts selected and directed by Toxicology Excellence for Risk Assessment (TERA), including Ms. Joan Strawson (TERA), Dr. Donna Vorhees (Science Collaborative – North Shore) and Dr. John Christopher (California Department of Toxic Substances). 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 assessment was subject to a 60-day public comment period.

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

Substance Identity

Formamide is a colourless, hygroscopic, oily liquid (Sax and Lewis 1987). The substance identity is presented in Table 1.

Table 1. Substance identity of formamide

CAS RN	75-12-7
NCI names	Formamide
Other names	Carbamaldehyde; Formimidic acid; Methanamide
Chemical group	Discrete organics
Chemical subgroup	Aliphatic amide, aliphatic carboxamide, alkanolic acid
Chemical formula	CH ₃ NO
Chemical structure	
SMILES	O=CN
Molecular mass	45.04 g/mol

Abbreviations: CAS RN, Chemical Abstracts Service Registry Number; NCI, National Chemical Inventories; SMILES, simplified molecular input line entry specification.

Source: NCI 2006

Physical and Chemical Properties

The physical and chemical properties of formamide are presented in Table 2.

Table 2. Physical and chemical properties of formamide

Property	Type	Value	Temperature (°C)	Reference
Melting point (°C)	Experimental	2.55		Budavari 1996; PhysProp 2008
Boiling point (°C)	Experimental	210.5		Budavari 1996
Density (kg/m ³)		1.1338		Körösi and Kováts 1981
Vapour pressure (Pa)	Experimental	8.13 (0.061 mmHg) ¹	25	Daubert and Danner 1989
Henry's Law constant (Pa·m ³ /mol)	Modelled	1.55 × 10 ⁻³	25	HENRYWIN 2000
Log K _{ow} (dimensionless)	Experimental	-1.51	(no temperature provided)	Hansch et al. 1995
	Experimental	-0.82	25	BASF AG 1988a
Log K _{oc} (dimensionless)	Modelled	0.176		PCKOCWIN 2000

Property	Type	Value	Temperature (°C)	Reference
Water solubility (mg/L)	Experimental	1 000 000 (miscible)	25	PhysProp 2008
pK _a (dimensionless)	Experimental	−0.48 (for base form)	25	PhysProp 2008

Abbreviations: K_{oc}, organic carbon partition coefficient; K_{ow}, octanol–water partition coefficient; pK_a, acid dissociation constant.

¹ The value in parentheses is the value originally reported in the reference.

Sources

No natural occurrence of formamide was identified (Howard 1993; OECD 2007). Formamide may be emitted to the environment as a result of its manufacture and use as an intermediate and solvent (OECD 2007).

Uses

Formamide is a solvent used in the manufacture and processing of plastics, non-aqueous electrolysis, crystallization of pharmaceuticals and separation of chlorosilanes (Howard 1993). It is used in soil stabilization and as an ink solvent in fibre and felt-tip pens and markers. It has been used or has the potential to be used as an additive to lubricating oil and hydraulic fluid, a component of de-icing fluids for airport runways, a curing agent for epoxy resins, a plasticizer, an affinity enhancer for dyes and a component of liquid fertilizers. It is an intermediate in the production of formic acid and in the synthesis of hydrogen cyanide, imidazoles, pyrimidine and 1,3,5-triazines. It is also used as a monomer in the production of polymers, such as heat-resistant coatings.

Formamide is applied as a softener in the production of pastes and paper (OECD 2007). Although it can be used as an intermediate in the production of fungicides (OECD 2007), it has never been registered for use in pesticides in Canada (2008 personal communication from Pest Management Regulatory Agency; unreferenced).

Formamide is an analytical reagent for, for example, moisture content analysis (Corn Refiners Association, Inc. 2006) and the detection of *Listeria monocytogenes* (Health Canada 2005).

Formamide has not been notified in cosmetics in Canada, nor is it reported in the International Cosmetic Ingredient Dictionary (2008 personal communication from Consumer Product Safety, Cosmetics Division, Health Canada; unreferenced). Two skin cleansing products in Canada contain greater than 30% polyvinyl formamide.

In Canada, formamide is used as an emulsifier in some invert emulsion fluids (CAPP 2008).

Previously reported uses in Canada in 1984–1986 were as an analytical reagent, a chemical intermediate, a drilling mud additive/oil recovery agent/oil well treating agent and a solvent/carrier. The industrial sectors identified at that time were biotechnology, specialty organic chemicals, petroleum and natural gas, photographic/photocopier and pigment, dye and printing ink (Environment Canada 1988).

According to submissions made under section 71 of CEPA 1999, formamide is used in conventional oil and gas extraction and as a corrosion inhibitor (Environment Canada 2008c). With respect to amounts of formamide in use in Canada, in 2006, three companies reported importing formamide in quantities between 1000 and 10 000 kg, and fewer than four companies reported using between 1 and 1200 kg (Environment Canada 2008c).

Release to the Environment

Formamide may be emitted to the environment as a result of its manufacture and use as an intermediate and solvent (OECD 2007). It was detected at 2.0 mg/L in condensate retort water of an oil shale retort but was not detected in the process retort water. It was detected in wastewater from a polyacrylamide production plant and also in the waste streams from an acrylonitrile plant as a result of a detoxification process for cyanide-containing wastewaters. It may also be detected in waste streams due to chemical hydrolysis of cyanide.

Environmental Fate

Based on its physical and chemical properties (Table 2), the results of Level III fugacity modelling (Table 3) suggest that formamide will reside predominantly in water and soil, depending on the compartment of release.

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

Substance released to:	Fraction of substance partitioning to each medium (%)			
	Air	Water	Soil	Sediment
Air (100%)	0.161	25.0	74.8	0.0416
Water (100%)	5.34E-6	99.8	0.00248	0.167
Soil (100%)	0.00128	22.1	77.9	0.0369

Persistence and Bioaccumulation Potential

Environmental Persistence

Results of biodegradability tests (Table 4) show degradation ranging from 22.6% to 100% after 14 days in a Japanese Ministry of International Trade & Industry (MITI) test and 28 days in a dissolved organic carbon (DOC) die-away test, respectively. According to MITI criteria, formamide is classified as “well biodegradable” (Sasaki 1978). These test data indicate that the ultimate degradation half-life in water is shorter than 182 days (6 months) and that the substance is considered to not persist in that environmental compartment.

Table 4. Empirical data for persistence of formamide

Medium	Fate process	Degradation value	Degradation endpoint / units	Reference
Water	Biodegradation (Japanese MITI test)	>30	% BOD; 14 days; inoculum; activated sludge	Sasaki 1978
Water	Biodegradation (Japanese MITI test)	>30	% BOD; 14 days; inoculum; activated sludge	Kawasaki 1980
Water	Biodegradation (Japanese MITI test)	22.6; 57.7	% BOD; 14 days; inoculum; activated sludge	Kitano 1978
Water	Biodegradation (DOC die-away test)	90–100	% DOC; 28 days; inoculum; activated sludge from a wastewater treatment plant	BASF AG 2003b

Abbreviations: BOD, biochemical oxygen demand; DOC, dissolved organic carbon; MITI, Ministry of International Trade & Industry, Japan.

Although experimental data on the degradation of formamide are available, a quantitative structure–activity relationship (QSAR)-based weight of evidence approach (Environment Canada 2007) was also applied using the degradation models shown in Table 5.

Table 5. Modelled data for degradation of formamide

Fate process	Model and model basis	Result	Interpretation	Extrapolated half-life (days)	Extrapolation reference and/or source
Air					
Atmospheric oxidation	AOPWIN 2000 (days)	5.348	Degrades slowly in air	n/a	n/a
Water					
Hydrolysis	HYDROWIN 2000		Hydrolysis rate extremely slow	>365	n/a
Biodegradation (aerobic)	BIOWIN 2000 Submodel 1: Linear probability	0.9363	Biodegrades quickly in water	n/a	n/a
Biodegradation	BIOWIN 2000	0.9937	Biodegrades	n/a	n/a

Fate process	Model and model basis	Result	Interpretation	Extrapolated half-life (days)	Extrapolation reference and/or source
(aerobic)	Submodel 2: Non-linear probability		quickly in water		
Biodegradation (aerobic)	BIOWIN 2000 Submodel 3: Expert Survey (ultimate biodegradation) (days)	3.0454	Ultimate degradation in weeks in water	15	US EPA 2002
				<60	Aronson et al. 2006
Biodegradation (aerobic)	BIOWIN 2000 Submodel 4: Expert Survey (primary biodegradation) (days)	3.9882	Primary biodegradation in days in water	2	US EPA 2002
				<60	Aronson et al. 2006
Biodegradation (aerobic)	BIOWIN 2000 Submodel 5: MITI linear probability	0.7048	Readily biodegradable in water	<60	Aronson et al. 2006
Biodegradation (aerobic)	BIOWIN 2000 Submodel 6: MITI non-linear probability	0.8920	Readily biodegradable in water	<60	Aronson et al. 2006
Biodegradation (anaerobic)	BIOWIN 2000 Submodel 7: Linear probability	0.2682	Does not biodegrade quickly	n/a	n/a
Biodegradation	BIOWIN 2000 Overall conclusion	Yes	Readily biodegradable in water	n/a	n/a
Biodegradation (aerobic)	CATABOL ©2004–2008 % BOD (OECD TG 301C)	100.0	Not persistent in water	<182	Calculated from BOD assuming first-order rate kinetics

Abbreviations: BOD, biochemical oxygen demand; MITI, Ministry of International Trade & Industry, Japan; n/a, not available; OECD TG, Organisation for Economic Co-operation and Development Test Guideline.

According to a model of gas/particle partitioning of semivolatile organic compounds in the atmosphere (Bidleman 1988), formamide is expected to exist solely as a vapour in the ambient atmosphere (SRC 1988). Vapour-phase formamide is degraded in the atmosphere by reaction with photochemically produced hydroxyl radicals (SRC 1988). According to AOPWIN (2000), formamide has a calculated half-life in air of 5.348 days, assuming a hydroxyl radical concentration of 1.5×10^6 molecules/cm³ (Leifer 1993).

That concentration assumes an estimated global daylight average of 12 h, which is currently recommended by the US Environmental Protection Agency (EPA).

Formamide is predicted to hydrolyse very slowly at room temperature (Table 5; Hine et al. 1981), although the rate of hydrolysis increases rapidly in the presence of acids or bases and is further accelerated at elevated temperatures (HSDB 2008).

Given the ecological importance of the water compartment and the fact that most of the available models apply to water, biodegradation in water was examined in most detail. The results from Table 5 show that all of the aerobic probability models (BIOWIN submodels 1, 2, 5 and 6) suggest that this substance biodegrades quickly. In fact, all probability results, except BIOWIN submodel 7, are greater than 0.3, the cut-off suggested by Aronson et al. (2006) to identify substances as having a half-life of >60 days (based on the MITI probability models), and are greater than 0.5, the probability suggested by the model developers as indicating “biodegrades quickly” or “readily biodegradable.” BIOWIN submodel 7 results suggest a lower probability of anaerobic degradation—i.e., that formamide does not biodegrade quickly under anaerobic conditions. The half-life result from the primary survey model (BIOWIN submodel 4) of “days” is suggested to mean approximately 2.3 days (US EPA 2002; Aronson et al. 2006), and the ultimate survey model (BIOWIN submodel 3) result of “weeks” is suggested to mean approximately 15 days by the US EPA (2002) and <60 days by Aronson et al. (2006). The overall conclusion from BIOWIN (2000) is that formamide is “readily biodegradable.”

The ultimate degradation model CATABOL (©2004–2008) predicts that formamide will undergo complete mineralization in a 28-day timeframe. That model predicted a 100% rate of biodegradation based on the Organisation for Economic Co-operation and Development (OECD) Test Guideline 301 ready biodegradation test (% BOD), which has been suggested to mean “not persistent” (Aronson and Howard 1999) and having a half-life in water of <182 days, assuming first-order rate kinetics.

Overall, modelled and empirical data indicate fast biodegradation of formamide in water.

Based on the empirical and modelled data and using an extrapolation ratio of 1:1:4 for a water:soil:sediment biodegradation half-life (Boethling et al. 1995), the ultimate degradation half-life in soil and sediments is <60 days. This indicates that formamide is expected to not be persistent in soil and sediment.

Thus, the empirical and modelled data (Tables 5 and 6) demonstrate that formamide does not meet the persistence criteria for water, soil or sediment (half-lives in soil and water ≥ 182 days and/or half-life in sediment ≥ 365 days) as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000). Formamide is considered to be persistent in air (half-life in air ≥ 2 days).

Potential for Bioaccumulation

The modelled log K_{ow} value of -1.15 for formamide (Hansch et al. 1995) suggests that this chemical has low potential to bioaccumulate in the environment (see Table 2 above).

As no experimental bioaccumulation factor (BAF) or bioconcentration factor (BCF) data for formamide were available, a predictive approach was applied using available BAF and BCF models, as shown in Table 6.

Table 6. Fish BAF and BCF predictions for formamide

Test organism	Endpoint	Value (L/kg wet weight)	Reference
Fish	BAF	1.0	Arnot and Gobas 2003 (Gobas BAF Middle Trophic Level) ¹
Fish	BCF	1.0	Arnot and Gobas 2003 (Gobas BCF Middle Trophic Level) ¹
Fish	BCF	8.91	OASIS Forecast 2005
Fish	BCF	3.16	BCFWIN 2000

¹ The default of no metabolism was used for this calculation.

Metabolism information for this substance was not available, nor was it considered in the BAF or BCF models.

The modified Gobas BAF middle trophic level model for fish predicted a BAF of 1.0 L/kg, indicating that formamide does not have the potential to bioconcentrate and biomagnify in the environment. The results of BCF model calculations provide additional evidence supporting the low bioconcentration potential of this substance. Based on the available kinetic-based and other modelled values, formamide does not meet the bioaccumulation criterion (BCF or BAF ≥ 5000) as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

Potential to Cause Ecological Harm

As indicated previously, formamide does not meet the bioaccumulation criteria as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000). It also does not meet the persistence criteria for soil or water as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000), although it does meet the criterion for persistence in air.

Ecological Effects Assessment

Aquatic Compartment

Although no measured aquatic toxicity data were available for use in the categorization of formamide, further research has revealed a number of such studies, as summarized in Table 7.

Table 7. Measured data for aquatic toxicity of formamide

Test organism	Type of test	Endpoint	Value (mg/L)	Reference
<i>Leuciscus idus</i>	Acute (96 h)	LC ₅₀	6 569	BASF AG 1989
<i>Danio rerio</i>	Acute (96 h)	LC ₅₀	9 135	Groth et al. 1994
<i>Chaetogammarus marinus</i>	Acute (96 h)	EC ₅₀	19 031	Adema 1982
<i>Daphnia magna</i>	Acute (48 h)	EC ₅₀	>500	BASF AG 1988b
<i>Scenedesmus subspicatus</i> (renamed <i>Desmodesmus subspicatus</i>)	Acute (72 h)	EC ₅₀	>500	BASF AG 1988c
<i>Scenedesmus subspicatus</i> (renamed <i>Desmodesmus subspicatus</i>)	Acute (24 h)	EC ₅₀	>2 000	Eich et al. 1997
<i>Lemna minor</i>	Acute (24 h)	EC ₅₀	81.2	Eich et al. 1997
<i>Xenopus laevis</i> (embryos)	Acute (96 h)	LC ₅₀	11 400	Dresser et al. 1992
		EC ₅₀	12 800	

Abbreviations: EC₅₀, concentration of a substance that is estimated to cause some toxic sublethal effect on 50% of the test organisms; LC₅₀, concentration of a substance that is estimated to be lethal to 50% of the test organisms.

A number of published and unpublished acute toxicity studies were included in the European Commission assessment of formamide (ECB 2000). The acute toxicity of formamide to the golden orfe (*Leuciscus idus*) was assessed by BASF AG (1989) in a 96-h static test based on the German Industrial Standard DIN 38412, Part 15, resulting in a 96-h median lethal concentration (LC₅₀) of 6569 mg/L. The toxicity of formamide to embryos of the zebra fish *Danio rerio* was assessed by Groth et al. (1994) in a static test. The 96-h LC₅₀ was 9135 mg/L, and the 96-h no-observed-effect concentration (NOEC) for sublethal effects (e.g., skeletal abnormalities, reduction of heart size and blood circulation) was 1080 mg/L.

The acute toxicity of formamide to the water flea (*Daphnia magna*) was investigated in a 48-h static study following Directive 79/831/EEC, Annex V, Part C. Based on nominal concentrations, the 48-h median effective dose (EC₅₀) was >500 mg/L (BASF AG 1988b). The acute toxic effects of formamide on the marine gammarid *Chaetogammarus marinus* were also tested (Adema 1982). The 96-h EC₅₀ was calculated to be 19 031 mg/L, and the NOEC was 1000 mg/L.

The toxicity of formamide to algae was studied in two 96-h static tests, conducted by BASF AG (1988c), with the green alga *Desmodesmus subspicatus*, closely following German Industrial Standard DIN 38412, Part 9. In both studies, the 72- and 96-h EC₅₀ values for growth rate and biomass were above the highest concentration tested (>500 mg/L). In the first test, however, significant differences in cell growth and biomass were observed. The 72-h NOEC values were 15.6 mg/L for biomass integral and 125 mg/L for growth rate. The 72-h NOEC values in the second test were ≥500 mg/L for biomass integral and growth rate. Conductometric measurements of ion leakage were carried out with *Desmodesmus subspicatus* in order to determine membrane damage due

to formamide exposure (Eich et al. 1997). Based on nominal concentrations, the 24-h EC_{50} was >2000 mg/L and the 24-h EC_{10} (effective concentration for 10% effect) was 77.6 mg/L. Additional measurements of ion leakage resulting from membrane damage were carried out with the aquatic macrophyte *Lemna minor*, and a 24-h EC_{50} of 81.2 mg/L was determined (Eich et al. 1997).

Body length and developmental malformations of South African clawed frog (*Xenopus laevis*) embryos were measured in a 96-h test by Dresser et al. (1992). The pooled 96-h LC_{50} and EC_{50} values for mortality and malformation corresponded to 11 400 mg/L and 12 800 mg/L, respectively. The 96-h NOEC for body length ranged from 8500 to 11 300 mg/L, and the 96-h NOEC for malformation ranged from 5700 to 8500 mg/L.

No empirical chronic toxicity data were available.

In addition to available empirical data, a number of modelled toxicity data were produced to add to the weight of evidence on the aquatic toxicity potential of formamide. Those are presented in Table 8. The modelled results were generally consistent with measured data, with the exception of TOPKAT (2004), which predicted an acute fish LC_{50} of 1.3 mg/L. That model uses a fragment method to calculate the toxicity, whereas ECOSAR (2004) calculates toxicity based on regressions of K_{ow} and toxicity.

Table 8. Modelled data for aquatic toxicity of formamide

Test organism	Type of test	Endpoint	Value (mg/L)	Reference
Fish	Acute (96 h)	LC_{50}	1.3 ¹	TOPKAT (2004)
Fish	Acute (96 h)	LC_{50}	82 605	ECOSAR (2004)
Fish	Acute (14 days)	LC_{50}	84 317.9	ECOSAR (2004)
<i>Daphnia</i>	Acute (48 h)	EC_{50}	267.6	TOPKAT (2004)
<i>Daphnia</i>	Acute (48 h)	LC_{50}	68 977	ECOSAR (2004)
<i>Daphnia</i>	Chronic (16 days)	EC_{50}	729.1	ECOSAR (2004)
Shrimp	Acute (96 h)	LC_{50}	313 000	ECOSAR (2004)
Alga	Acute (96 h)	EC_{50}	35 031.6	ECOSAR (2004)

Abbreviations: EC_{50} , concentration of a substance that is estimated to cause some toxic sublethal effect on 50% of the test organisms; LC_{50} , concentration of a substance that is estimated to be lethal to 50% of the test organisms.

¹ Pivotal value for categorization; critical toxicity value (CTV) used for this assessment.

Other Environmental Compartments

In a study by Nicoloff et al. (1980) on dry seeds of two barley varieties, neither shoot length nor number of chlorophyll mutations were found to be affected significantly by exposure to formamide.

Ecological Exposure Assessment

No Canadian release data for formamide are available.

In this screening assessment, a generic conservative exposure scenario (Industrial Generic Exposure Tool – Aquatic) was developed to estimate releases into the aquatic environment from industrial operations using the substance and the resulting aquatic concentrations (Environment Canada 2008a, b). Given the high water solubility of formamide and the lack of release data, environmental concentrations in watercourses were estimated based on use volumes reported. No information was available on current consumer uses in Canada; therefore, releases to the environment as a result of those uses could not be estimated.

Assuming a use quantity of 1200 kg/year, which is the highest use amount reported at one facility under section 71 of CEPA 1999 (Environment Canada 2008c), 5% loss during use, approximately 16% removal during wastewater treatment (STP 2001) and release to a small generic watercourse, the conservative predicted environmental concentration (PEC) in water is 0.0056 mg/L.

Characterization of Ecological Risk

Based on the available information, formamide is not persistent in the principal environmental media into which it tends to partition (water and soil) and is not bioaccumulative based on criteria defined in the *Persistence and Bioaccumulation Regulations* (Canada 2000). While formamide is persistent (i.e., is not rapidly transformed) in air, it will not remain in that medium but will partition into soil and water, where it is not persistent.

Experimental ecotoxicological data indicate that formamide does not cause significant harm to aquatic organisms at low concentrations. The lowest modelled acute ecotoxicity value for fish was 1.3 mg/L (TOPKAT 2004), and measured acute toxicity values ranged from 81.2 mg/L for the duckweed *Lemna minor* (Eich et al. 1997) to 19 031 mg/L for the amphipod *Chaetogammarus marinus* (Adema 1982). Taking the modelled 1.3 mg/L result as the critical toxicity value (CTV) and applying an assessment factor of 100 to account for inter- and intraspecies variability and extrapolation from an estimate of acute effects to a no-effect concentration in the field gives a PNEC of 0.013 mg/L.

Dividing the PEC of 0.0056 mg/L, obtained using the conservative Industrial Generic Exposure Tool – Aquatic (Environment Canada 2008a, b), by the PNEC of 0.013 mg/L yields a conservative risk quotient of 0.43, which indicates that formamide is not anticipated to cause ecological harm at environmental concentrations predicted from existing use pattern information

Based on the information available, formamide is unlikely to cause ecological harm in Canada.

Uncertainties in Evaluation of Ecological Risk

There was significant uncertainty in this assessment with regard to the release of formamide into the Canadian environment. Although a survey was conducted to collect use and release data (Canada 2008), industry response was limited. This screening assessment was based on the information provided through that process, as well as information from the international literature indicating a low potential for release.

No empirical chronic toxicity data were available for this assessment; therefore, risk assessment calculations were based on acute effects, with the incorporation of appropriate assessment factors. Ideally, empirical chronic toxicity results would be used directly.

Also, regarding ecotoxicity, based on the predicted partitioning behaviour of this chemical, the significance of soil as an important medium of exposure is not well addressed by the effects data available. Indeed, most of the effects data identified apply primarily to pelagic aquatic exposures, although the water column may not be the only medium of concern, depending on release patterns.

Potential to Cause Harm to Human Health

Exposure Assessment

An assessment by the California Environmental Protection Agency (1997) concluded that there is a low level of concern over the extent of exposure to formamide, as it appears to be used mainly in the pharmaceutical industry as a chemical intermediate or a solvent. It is miscible in water and has low volatility. Formamide is biodegraded and degraded by hydroxyl radicals in the atmosphere. It is not expected to bioconcentrate or biomagnify.

In Canada, formamide is a Class 2 residual solvent (solvent to be limited) in pharmaceutical products (Health Canada 1998a), with a concentration limit of 220 mg/kg (where the maximum daily dose of the product does not exceed 10 g) or a permitted daily exposure of 2.2 mg/day. An identical concentration limit and permitted daily exposure have been set for residual formamide in both natural health products (Health Canada 2007) and veterinary medicinal products (VICH 2000).

Formamide is used in molecular biology laboratories in denaturing gel electrophoresis (California Environmental Protection Agency 1997). It is also used in some glues.

In France, formamide was detected as an emission from a polyester wall covering backed with polyvinyl chloride (Karpe et al. 1995).

In an early report, formamide was detected in the ink from a felt-tip pen made in France, at a concentration up to 50% (Seemann et al. 1976). In contrast, prefilled pens, jumbo

pens and inks currently sold by Pillar Technologies (2008) were reported to contain no formamide.

The most probable exposure of the general population to formamide is as a result of its use in water-soluble ink formulations (Appendix 1; US EPA 1986). A worst-case intake for a child who had painted the central surface of both hands with ink from a broad-tip marker has been reported by the US EPA (1986) (Appendix 1). The concentration of formamide in the ink was 20%, and 100% absorption was assumed. The estimated intake resulting from this dermal exposure was 0.3 mg/kg body weight (kg-bw). The US EPA concluded that “although a large number of consumers may use writing instruments containing formamide, the levels of individual exposure are likely to be well below those found to cause adverse effects.”

More recent information has been obtained from the Art and Creative Materials Institute of Duke University (2008 personal communication to Environment Canada; unreferenced), which reported that formamide is present in some markers in the United States at concentrations ranging from 10% to 27.7%. It was noted that, among 360 000 art materials evaluated, formamide was present in six marker lines and was used in no other type of art material. The estimated amounts of formamide to which an individual might be exposed following 8 h of use of each of six markers are presented in Appendix 2. Presentation is limited to acute exposures only, as estimates of exposure resulting from daily use more closely approximate occupational exposure. For the six available inks, the amounts of formamide to which an individual might be exposed ranged from 22.2 µg/kg-bw to 56.4 µg/kg-bw per day. Estimates of intake were prepared for children age 6 months to four years, as this age group is considered to be the most highly exposed through the use of markers or pens, particularly in terms of mouthing (US EPA 2008).

Based upon the information identified with respect to use, there is confidence that exposure of the general population to formamide is limited to use of felt-tip pens and markers. However, there is uncertainty with respect to the extent of exposure from this source, as the extent of the use of formamide as a solvent in pens and markers sold in Canada is unknown.

Health Effects Assessment

Appendix 3 contains a summary of the available health effects information for formamide.

The European Commission has classified formamide as a reproductive and developmental Category 2 substance with risk phrase R61 (*may cause harm to the unborn child*) (European Commission 2000, 2001; ESIS 2009).

Formamide was found to be embryotoxic and teratogenic in several oral gavage studies using rabbits, rats and mice. The maternal toxicity caused by formamide included reduced food consumption, reduced body weight gain and decreased gravid uterine

weight; fetal toxicity included reduced fetal weight and increased incidences of fetal death; and teratogenicity included skeletal malformations, cleft palate anencephaly and fused ribs.

In rabbit studies, the lowest lowest-observed-adverse-effect levels (LOAELs) for maternal toxicity, embryo/fetal toxicity and teratogenicity were 79, 79 and 79 mg/kg-bw per day, respectively (Merkle and Zeller 1980); embryotoxicity and teratogenicity were observed at maternally toxic doses (LTP 1974; Merkle and Zeller 1980; George et al. 2002). In rats, embryotoxicity (LOAEL = 100 mg/kg-bw per day) was seen in the absence of maternal toxicity (LOAEL = 200 mg/kg-bw per day for maternal toxicity) (NTP 1998; George et al. 2000). In mice, the lowest LOAELs for maternal toxicity, embryotoxicity and teratogenicity were 396, 198 and 198 mg/kg-bw per day, respectively; embryotoxicity and teratogenicity were seen in the absence of maternal toxicity (BASF AG 1974b; OECD 2007). Among the test animals, the rabbit was the species that was the most sensitive to formamide in terms of developmental toxicity. Thus, the lowest oral LOAEL for developmental toxicity (maternal toxicity, embryo/fetal toxicity and teratogenicity) is identified to be 79 mg/kg-bw per day in rabbits.

Embryotoxicity or teratogenicity was also observed in experimental rats or mice through dermal exposure (Oettel and Frohberg 1964; Von Kreybig et al. 1968; BASF AG 1973a, b, 1974c, 1983; Gleich 1974; Stula and Krauss 1977). In rats, the lowest LOAELs for maternal toxicity, embryotoxicity (early fetal deaths) and teratogenicity (distorted face or subcutaneous hemorrhage) were 600, 600 and 600 mg/kg-bw per day, respectively (Stula and Krauss 1977). In mice, the lowest dermal LOAEL for embryotoxicity was determined to be 300 mg/kg-bw per day based on an increase in early fetal deaths (Gleich 1974). The fetal abnormalities were observed only at the high-dose exposure (>2800 mg/kg-bw per day) in mice (BASF AG 1973a, b; Gleich 1974).

The reproductive toxicity of formamide was evaluated using the Reproductive Assessment by Continuous Breeding protocols in Swiss CD-1 mice treated at concentrations of 0, 100, 350 and 750 mg/L (equivalent to 0, 16–32, 48–110 and 144–226 mg/kg-bw per day, respectively) in drinking water (Fail et al. 1998). Reproductive toxicity was observed at 750 mg/L (144–226 mg/kg-bw per day) in parental F₀ and offspring F₁ generations, and the critical effects included decreases in fertility rate and reduction in live litter size. A crossover mating experiment suggested that the reduced fertility rate may be due to impairment of reproduction in females. In addition, after offspring F₁ mating, reduced offspring F₂ litter size, increased days to litter, reduced relative ovarian weight and lengthened estrous cycles were observed at 750 mg/L. The no-observed-adverse-effect level (NOAEL) for the reproductive toxicity of formamide was 350 mg/L (48–110 mg/kg-bw per day), and the LOAEL for the reproductive toxicity of formamide was 750 mg/L (144–226 mg/kg-bw per day) for both generations (Fail et al. 1998).

Recently, the US National Toxicology Program (NTP) published 2-year toxicology and carcinogenesis studies of formamide in B6C3F₁ mice and F344/N rats (NTP 2008). The

results from the NTP studies showed no evidence of carcinogenic activity of formamide in male or female rats. There was clear evidence of carcinogenic activity of formamide in male mice, based on increased incidences of hemangiosarcoma in the liver; there was equivocal evidence of carcinogenic activity in female mice, based on marginally increased incidences of hepatocellular adenoma or carcinoma (significant only when adenoma and carcinoma were combined) (NTP 2008).

Formamide was carcinogenic in the liver of male mice. In the NTP study, groups of 50 male and 50 female B6C3F1 mice were administered 0, 20, 40 or 80 mg formamide/kg-bw per day in deionized water by gavage, 5 days/week for 104 weeks. The incidences of hemangiosarcoma in liver were increased in a dose-related manner (1/50, 5/50, 7/50 and 8/50 for 0, 20, 40 and 80 mg/kg-bw per day, respectively) in males, with significant increases occurring at 40 and 80 mg/kg-bw per day ($p = 0.032$ and 0.016 , respectively). In female mice, the incidence of hepatocellular adenoma or carcinoma (combined) (14/50, 14/50, 20/50 and 28/50, respectively) showed a marginally significant increase at the 80 mg/kg-bw per day dose. Non-neoplastic effects, including mineralization of arteries or tunic in testis and hematopoietic cell proliferation in the spleen, were observed in male mice at 80 mg/kg-bw per day. No neoplastic lesions were observed in either male or female F344/N rats exposed to formamide at doses up to 80 mg/kg-bw per day. However, an increased incidence of bone marrow hyperplasia occurred in male rats (NTP 2008).

Formamide showed no evidence for mutagenicity in a series of short-term bioassays. Formamide was not mutagenic in Ames tests with several strains of *Salmonella typhimurium* or in a mutagenicity assay with *Escherichia coli* strain WPuvrA pKM101, with or without liver S9 metabolic activation (Arimoto et al. 1982; Mortelmans et al. 1986; NTP 2008). Formamide gave negative results in the sex-linked recessive lethal mutation assay in germ cells of male *Drosophila melanogaster* treated with formamide by either feeding or injection (Foureman et al. 1994; NTP 2008). In *in vivo* micronucleus tests, formamide did not induce increases in micronucleated erythrocytes in male or female mice treated with formamide (0–160 mg/kg-bw per day) by gavage for 3 months (NTP 2008), although in another study, a dose-dependent increase in the number of polychromatic erythrocytes containing micronuclei was seen in bone marrow of mice exposed to formamide via intraperitoneal injection at higher doses (225–1800 mg/kg-bw), with significance at doses of 900 mg/kg-bw or higher (BASF AG 2001). However, at the dose of 160 mg/kg-bw, increased incidences of lesions of several tissues/organs and decreased body weights were seen in mice, suggesting that the observed induction of micronuclei may be attributed to the cell damage. In an *in vitro* cell transformation assay, a negative result was seen in rat embryo cells at low concentrations (0.01–100 µg/mL) (Freeman et al. 1973), although a significant and concentration-related increase in the number of transformed colonies was observed in Syrian hamster embryo cells exposed to formamide at higher concentrations (300–550 µg/mL) (BASF AG 2003a). Based on the weight of evidence, formamide is not considered to be mutagenic.

The detailed modes of action for the increased incidence of hemangiosarcoma and other tumours in liver have not been proposed or developed by other regulatory agencies, and the development and analysis of the modes of action are normally outside the scope of a screening-level risk assessment. Although it has been suggested that a significant association exists between Kupffer cell pigmentation associated with red cell hemolysis and the incidence of hemangiosarcomas (Nyska et al. 2004), in the NTP study, no hemosiderin pigmentation was observed in the spleen or in the liver, suggesting that hemolysis may not contribute to the induction of hemangiosarcoma in mice liver (NTP 2008). In a review paper on the biological effects of formamide, the author suggested that formamide caused cancer by a non-genotoxic mode of action (Kennedy 2001). Based on the evidence of carcinogenicity observed in only one organ (liver), one sex (male) and one species (mice) and the conclusion that formamide is not mutagenic, the tumours observed in the experimental animals are unlikely to have resulted from direct interaction with genetic material.

With regard to repeated-dose short-term and subchronic toxicity, the main effects found in rats or mice include changes in hematological parameters, irrespective of route of exposure. In a subchronic study, an oral LOAEL of 40 mg/kg-bw per day was determined based on significant increases in hematocrit values, hemoglobin concentrations and erythrocyte counts in both male and female F344/N rats administered 0, 10, 20, 40, 80 or 160 mg formamide/kg-bw per day by gavage, 5 days/week for 14 weeks. The incidences of degeneration of the germinal epithelium of the testes and epididymis were significantly increased in males at the highest dose (NTP 2008). The same oral LOAEL of 40 mg/kg-bw per day was also obtained based on a significant decrease in body weight gains in male B6C3F1 mice administered 0, 10, 20, 40, 80 or 160 mg formamide/kg-bw per day by gavage, 5 days/week for 14 weeks. Increased incidences of non-neoplastic lesions (hyperplasia and inflammation) were seen in pancreatic ducts at the dose of 80 mg/kg-bw per day (NTP 2008). In a short-term study, a higher oral LOAEL of 113 mg/kg-bw per day was identified based on changes in hematological parameters, body weight loss, failure of reflexes, organ atrophy and tissue disintegration (gastrointestinal tract, testes, adrenal gland and kidney) in rats administered formamide at 0, 34, 113, 340 or 1130 mg/kg-bw per day by gavage (BASF AG 1978).

For dermal exposure, a LOAEL of 300 mg/kg-bw per day was identified based on hematological changes (increases in erythrocyte counts and hemoglobin) in rats treated with dermal applications of formamide at 0, 300, 1000 or 3000 mg/kg-bw per day for 90 days (BASF AG 1985a). At the highest dose level, clinical signs (e.g., erythema), pathological effects and an increased incidence of bilateral testicular tubular atrophy were seen (BASF AG 1985a).

Only one short-term inhalation study was reported. In a 2-week inhalation study, Crl:CD BR male rats were exposed to formamide at concentrations of 0, 190, 930 or 2800 mg/m³ (6 h/day, 5 days/week). At the highest concentration (2800 mg/m³), microscopic lesions in the kidney (necrosis and regeneration of renal tubular epithelial cells) and an increase in kidney weights were observed. A lowest-observed-effect concentration (LOEC) of 930

mg/m³ (500 ppm) was identified, based on a significant decrease in the platelet count (hematological effect) (Warheit et al. 1989).

Toxicokinetic studies with rats or mice following a single oral administration showed that formamide was rapidly and completely absorbed in rats and mice, with peak plasma levels occurring within 2 h. The elimination half-life was about 15 h in rats and 4–6 h in mice (MRI 1998). The metabolism and distribution of formamide were studied in rats and mice treated with ¹⁴C-labelled formamide via intravenous injection or inhalation exposure. The results showed that about 30% of formamide was excreted unchanged in urine within 72 h; about 30% (for rats) or 50% (for mice) was excreted as carbon dioxide in breath, and only a minor quantity (1–3%) was excreted in the feces (RTI 1996). It was suggested that cytochrome P450 2E1 was the primary enzyme of formamide metabolism (RTI 1996).

The confidence in the toxicity database for formamide is considered to be moderate. Oral dosing studies (short-term, subchronic and chronic toxicity, carcinogenicity and genotoxicity, reproductive toxicity and developmental toxicity) are available, although inhalation exposure studies (carcinogenicity, reproductive toxicity and developmental toxicity) are limited. The modes of action for the observed carcinogenicity of formamide have not been elaborated.

Characterization of Risk to Human Health

Based principally on the weight of evidence–based assessments of international or other national agencies, a critical effect for the characterization of risk to human health for formamide is carcinogenicity. In the standard 2-year carcinogenicity studies with rats and mice, induced tumours were observed in only one organ (liver), one sex (male) and one species (mice). Based on the weight of evidence of the available genotoxicity data, formamide is not considered to be mutagenic. Although the mode of induction of tumours has not been developed and elucidated, the tumours observed in the experimental animals are unlikely to have resulted from direct interaction with genetic material.

Based on consideration of the weight of evidence–based classification of formamide by the European Commission as a Category 2 substance for reproductive and developmental effects (European Commission 2000, 2001; ESIS 2009), assessments prepared by other national and international jurisdictions (DECOS 1995; OECD 2007) and consideration of the available relevant data, the non-cancer critical effects for characterization of risk to human health for formamide are reproductive, developmental and hematological toxicity. Therefore, margins of exposure are derived between lowest exposure levels associated with induction of these effects and estimates of population exposure to formamide.

Based upon the information obtained on current use of formamide in Canada, exposure to the general population is expected to be limited to the use of felt-tip pens/markers, where formamide may be used as a solvent. Based upon information obtained on markers sold

in the United States, maximum intakes by a child (aged 0.5–4 years) using such a marker was predicted to range from 22.2 to 56.4 µg/kg-bw per day (Appendix 2).

With respect to consumer product exposure, dermal exposure is the main route for a child exposed to formamide through the use of felt-tip markers. Among the non-cancer critical effects (Appendix 3), the lowest dermal LOAELs were identified to be 300 mg/kg-bw per day in a subchronic study based on hematological toxicity in rats; and 300 mg/kg-bw per day for developmental toxicity in mice. Comparison of the critical effect levels of 300 mg/kg-bw per day (hematological and developmental effects) with the conservative estimate of 22.2 to 56.4 µg/kg-bw per day for a child via dermal exposure results in margins of exposure ranging from 5 300 to 13 500. If a short-term dermal LOAEL of 600 mg/kg-bw per day is used for the margin of exposure calculation, which is more appropriate to the pattern of consumer product exposure, the margin of exposure will be much larger. As exposure from environmental media was not quantifiable, margins of exposure for chronic non-cancer effects could not be calculated. Margins between non-cancer effect levels and exposure from environment media are also expected to be large. The margins of exposure are considered to be adequately protective for non-cancer effects.

On the basis of consideration of the existence of a practical threshold for non-mutagenic carcinogenicity of formamide in the experimental animal studies and the magnitude of the margins of exposure for non-cancer effects, it is concluded that formamide should be considered as a substance that 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.

Uncertainties in Evaluation of Risk to Human Health

The assessment of the carcinogenicity and genotoxicity of formamide is based mainly on a recently published NTP carcinogenicity study and a small genotoxicity database. The Existing Substances Bureau will continue to monitor international developments for the evaluation of the carcinogenicity of formamide. Additionally, the precise modes of induction of mouse liver tumours have not been developed. To elucidate the mode of action for carcinogenicity, future research is warranted. Some confounding factors include treatment-related tumours in male mice but not in female mice and evidence of bone marrow hyperplasia in rats but not in mice.

Based upon current uses, exposure of the general public to formamide is expected to be limited. Although there is potential for dermal exposure to formamide resulting from exposure to formamide-containing inks, the extent of availability of formamide-containing pens/markers in Canada is unknown. Pens and markers containing formamide were not reported to be imported into or used in Canada above the section 71 reporting threshold of 1000 kg/year for use or 100 kg/year for import or manufacture (Environment Canada 2008c).

Conclusion

Based on the available information in this screening assessment, it is concluded that formamide 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.

Based on the available information on its potential to cause harm to human health, it is concluded that formamide 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 formamide does not meet the criteria in section 64 of CEPA 1999. Additionally, formamide meets the criteria for persistence in air but does not meet the criteria for persistence in water, soil and sediments nor for bioaccumulation as set out in the *Persistence and Bioaccumulation Regulations* (Canada 2000).

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Appendix 1. Worst-case estimate of intake of formamide following dermal exposure, prepared by the US EPA (1986)

“... considering the physicochemical properties of formamide and the construction of the porous-tip device, which acts to hold the ink in the pen, EPA believes that the use of those products would not result in substantial exposure. On the basis of the above information, EPA has concluded that although a large number of consumers may use writing instruments containing formamide, the levels of individual exposure are likely to be well below those found to cause adverse effects.

“Formamide has been used in the past or has been suggested for a multitude of other commercial uses, some of which could result in significant worker and/or consumer exposure. Such potential uses include: as an additive to lube oil and hydraulic fluid, as a component in deicing fluids for airport runways, as a curing agent for epoxy resins, as a plasticizer, as an affinity enhancer for dyes, and as a component of liquid fertilizers. The current import volume and predominantly pharmaceutical use of this imported formamide supports the conclusion that the amount of formamide used for these and other uses is small, if any.”

Appendix 2. Estimates of intake of formamide by children (6 months – 4 years)¹ using felt-tip markers

Ink	Ink laydown rate (µg/cm) ²	Ink line / day ³	Amount of ink on skin / day (µg) ⁴	% formamide in ink ²	Body weight ⁵	Intake of formamide ⁶ (µg/kg-bw per day)
A	107	25 cm	2675	21.83	15.5	37.7
B	92		2300	27.7		41.1
C	55		1375	25		22.2
D	200		5000	15		48.4
E	219		5475	10		35.3
F	233		5825	15		56.4

¹ This age group is considered to be the most highly exposed through the use of markers or pens, particularly in terms of mouthing (US EPA 2008).

² Ink laydown rate and concentrations of formamide in ink reported by Art and Creative Materials Institute, Duke University (2008 personal communication to Environment Canada; unreferenceed; referred to as ACMI below).

³ ACMI reported that an individual may be exposed to an estimated 25 cm of ink line per day, through skin contact or incidental mouthing. Absorption of 100% is assumed.

⁴ Amount of ink on skin = ink laydown rate × 25 cm.

⁵ Body weight reported in Health Canada (1998b).

⁶ Intake = (amount of ink on skin) × (% formamide in ink) / (body weight).

Appendix 3. Summary of health effects information for formamide

Endpoint	Lowest effect levels ¹ /Results
Experimental animals and cells	
Acute toxicity	Oral LD₅₀ (mouse) = 3150 mg/kg-bw (Zaeva et al. 1967) Oral LD₅₀ (rat) = 5325 mg/kg-bw (BASF AG 1963) Oral LD₅₀ (rat) = 5577–6100 mg/kg-bw (Thiersch 1962; BASF AG 1963; Zaeva et al. 1967) Inhalation LC₅₀ (rat) = >21 mg/L in a 4-h exposure (Warheit et al. 1989) Dermal LD₅₀ (rat) = >3000 mg/kg-bw (BASF AG 1985b) Dermal LD₅₀ (rat) = >4000 mg/kg-bw (Von Kreybig et al. 1968) Dermal LD₅₀ (rat) = >13 500 mg/kg-bw (Stula and Krauss 1977) Dermal LD₅₀ (rabbit) = 6000–17 000 mg/kg-bw (Du Pont 1982) Intraperitoneal LD₅₀ (rat) = 5700–5900 mg/kg-bw (Azum-Gelade 1974) Intraperitoneal LD₅₀ (mouse) = 2060–7400 mg/kg-bw (Azum-Gelade 1974) Intraperitoneal LD₅₀ (guinea pig) = 1250 mg/kg-bw (Zaeva et al. 1967) Intravenous LD₅₀ (rat, mouse) = 5100–6000 mg/kg-bw (Azum-Gelade 1974)
Short-term repeated-dose toxicity	Lowest oral LOAEL: 113 mg/kg-bw per day was identified based on body weight loss, failure of reflexes, organ atrophy, tissue disintegration (gastrointestinal tract, testes, adrenal gland and kidney) and changes in hematological parameters in rats (20 per sex per group) administered formamide at 0, 34, 113, 340 or 1130 mg/kg-bw per day by gavage, 5 days/week for 4 weeks (BASF AG 1978). The NOAEL was 34 mg/kg-bw per day. Lowest inhalation LOEC: 930 mg/m³ (500 ppm) was identified based on significant decrease in the platelet count (hematological effect) in CrI:CD BR male rats (10 per group) exposed to formamide at concentrations of 0, 190, 930 or 2800 mg/m³ for 2 weeks (6 h/day, 5 days/week) (Warheit et al. 1989). The NOEC was 190 mg/m³ (100 ppm) based upon changes in the hematological and clinical chemistry parameters. At the highest concentration, microscopic lesions in the kidney and an increase in kidney weights were observed. Lowest dermal LOAEL: 600 mg/kg-bw per day in pregnant rats exposed to formamide on one or two gestation days, based on a reduction in body weight gain (Stula and Krauss 1977) Other dermal studies: A reduction in muscle tone and loss of coordination occurred in mice when tails were exposed to neat formamide 4 h/day for 12 days (Zaeva et al. 1967)

Endpoint	Lowest effect levels ¹ /Results
Subchronic toxicity	Lowest oral LOAEL: 40 mg/kg-bw per day was identified, based on decreased body weight in female F344/N rats and dose-related increases in hematocrit values, hemoglobin concentrations and erythrocyte counts in rats (10 per sex per group) with formamide treatment at 0, 10, 20, 40, 80 or 160 mg/kg-bw per day in deionized water by gavage, 5 days/week for 14 weeks. The incidences of degeneration of the germinal epithelium of the testes and epididymis were significantly increased in males at the highest dose (NTP 2008). Other oral studies: LOAEL of 40 mg/kg-bw per day, based on significant decrease in body weight gain in male B6C3F1 mice (10 per sex per group) administered 0, 10, 20, 40, 80 or 160 mg formamide/kg-bw per day in deionized water by gavage, 5 days/week for 14 weeks. The incidences of non-neoplastic lesions (hyperplasia and inflammation) in duct of pancreas were significantly increased at higher doses (NTP 2008). Inhalation LOAEC: Effects on blood pressure and on the spleen and lungs were seen in rats exposed to formamide at 4 mg/m³, 4 h/day, 5 days/week, for 4 months (Zaeva et al. 1967). Lowest dermal LOAEL: 300 mg/kg-bw per day was identified based on hematological changes (increases in erythrocyte counts and hemoglobin) in rats (10 per group) treated with dermal applications of formamide at 0, 300, 1000 or 3000 mg/kg-bw per day for 90 days (5 days/week, 6 h/day) (BASF AG 1985a). The clinical signs (erythema, apathy, reduced food consumption, decreased body weight) and pathological effects (decreased absolute weights of liver, kidney, spleen, adrenal glands and testes; increased relative weights of liver and kidneys) and an increased incidence of bilateral testicular tubular atrophy were observed at the highest dose level (BASF AG 1985a). A follow-up study was conducted at lower dose levels (0, 30, 100 and 3000 mg/kg-bw per day) for 90 days (5 days/week, 6 h/day) (BASF AG 1985b), and the NOAEL was identified as 100 mg/kg-bw per day.
Chronic toxicity/carcinogenicity	Oral carcinogenicity in rats: Groups of 50 male and 50 female F344/N rats were administered 0, 20, 40 or 80 mg formamide/kg-bw per day in deionized water by gavage, 5 days/week for 104 weeks. Decreased mean body weights were observed at 40 and 80 mg/kg-bw per day in females during the 2nd year. No neoplastic lesions were attributed to exposure to formamide in both male and female rats (NTP 2008). Oral carcinogenicity in mice: Groups of 50 male and 50 female B6C3F1 mice were administered 0, 20, 40 or 80 mg formamide/kg-bw per day in deionized water by gavage, 5 days/week for 104 weeks. The incidences of hemangiosarcoma in liver were increased in a dose-related manner (1/50, 5/50, 7/50 and 8/50 for 0, 20, 40 and 80 mg/kg-bw per day, respectively) in males. Significant increases occurred at 40 and 80 mg/kg-bw per day (NTP 2008). The incidence of hepatocellular adenoma or carcinoma (combined) was marginally increased in female mice at 80 mg/kg-bw per day (NTP 2008). Non-neoplastic effects: In the same study described above for mice and rats, the incidences of mineralization of the testicular arteries (0/50, 2/50, 5/50 and 35/50, respectively) and testicular tunic (1/50, 0/50, 5/50 and 27/50, respectively) and hematopoietic cell proliferation in the spleen (14/50, 14/50, 20/50 and 28/50, respectively) were significantly increased in male mice at 80 mg/kg-bw per day. In addition, an increased incidence of hyperplasia in bone marrow was observed in male rats (NTP 2008). No inhalation or dermal studies identified

Endpoint	Lowest effect levels ¹ /Results
Reproductive toxicity	Oral reproductive toxicity LOAEL: 750 mg/L (144–226 mg/kg-bw per day) in a continuous breeding study with CD-1 mice treated with formamide at a concentration of 0, 100, 350 or 750 mg/L (equivalent to 0, 16–32, 48–110 and 144–226 mg/kg-bw per day, respectively) in drinking water, based on decreased fertility rate and reduction in live litter size. Reduced offspring F₂ litter size, increased days to litter, reduced relative ovarian weight and lengthened estrous cycles were also observed at 750 mg/L after offspring F₁ mating. NOAEL for the reproductive toxicity of formamide was 350 mg/L (48–110 mg/kg-bw per day) (Fail et al. 1998). No inhalation or dermal studies identified
Developmental toxicity	Lowest oral LOAEL for developmental toxicity in rabbits: 79 mg/kg-bw per day in Chbb:HM rabbits treated with 0, 23, 79 or 226 mg/kg-bw per day by gavage on gestational days 6–18, based on the observed critical effects of decreased maternal body weight gain, decreased fetal body weight and increased fetal malformations (Merkle and Zeller 1980) Lowest oral LOAEL for developmental toxicity in rats: 100 mg/kg-bw per day in Sprague-Dawley rats treated with 0, 50, 100 or 200 mg/kg-bw per day by gavage on gestational days 6–19, based on the observed critical effect of decreased fetal body weight (George et al. 2000) [additional oral studies: BASF AG 1974a, b; LTP 1974; NTP 2001; George et al. 2002] Lowest dermal LOAEL for developmental toxicity in mice: 300 mg/kg-bw per day in mice (species not reported) treated with 0 or 300 mg/kg-bw per day on gestational days 10–11, based on the observed critical effect of increase in early fetal deaths (Gleich 1974) Lowest dermal LOAEL for developmental toxicity in rats: 600 mg/kg-bw per day in Sprague-Dawley rats treated with 0 or 600 mg/kg-bw per day on gestational days 11–12, based on the observed critical effect of increase in early fetal deaths (Stula and Krauss 1977) [additional dermal studies: Oettel and Frohberg 1964; Von Kreybig et al. 1968; BASF AG 1973a, b, 1974c] Inhalation: No effects on reproductive parameters observed in experimental animals chronically exposed to 6 mg formamide/m³ (Zaeva et al. 1967)
Genotoxicity and related endpoints: <i>in vitro</i>	Mutagenicity: Negative: <i>Salmonella typhimurium</i> TA97, TA98, TA100, TA1535 or TA1537 in the presence or absence of metabolic activation by induced rat or hamster liver S9; the doses ranged from 0 to 10 mg/plate (Mortelmans et al. 1986; NTP 2008) Negative: <i>S. typhimurium</i> TA98 up to 75 µL (~75 mg)/plate in the presence of S9 mixture (Arimoto et al. 1982) Negative: <i>Escherichia coli</i> strain WPuvrA pKM101 at concentrations up to 10 mg/plate with or without 10% rat liver S9 metabolic activation (NTP 2008) Cell transformation assay: Negative: In rat embryo cells at test concentrations of 0, 0.01, 0.1, 0.5, 1.0, 10 or 100 µg/ml (Freeman et al. 1973) Positive: Dose-related significant increase in the number of transformed colonies in Syrian hamster embryo (SHE) cells exposed to formamide at concentrations of 0, 300, 350, 400, 450, 500 or 550 µg/ml for 7 days (BASF AG 2003); limited evidence of cell transformation in hamster cells (Sanders 1972)
Genotoxicity and related endpoints:	Micronucleus tests: Negative: No increases in micronucleated normochromatic erythrocytes in

Endpoint	Lowest effect levels ¹ /Results
<i>in vivo</i>	peripheral blood were observed in male or female B6C3F1 mice treated with formamide at concentrations of 0, 10, 20, 40, 80 or 160 mg/kg-bw per day by gavage for 90 days (NTP 2008) Positive: A dose-dependent increase in the number of polychromatic erythrocytes containing micronuclei was seen in bone marrow of mice exposed to formamide at concentrations of 225, 450, 900, 1350 or 1800 mg/kg-bw via intraperitoneal injection after a harvesting interval of 48 h. The increases in micronucleated polychromatic erythrocytes were significant ($p < 0.01$) at doses of 900 mg/kg-bw or higher (BASF AG 2001). Sex-linked recessive lethal mutation assay: Negative: No significant increase in sex-linked recessive lethal mutations was observed in germ cells of male <i>Drosophila melanogaster</i> treated with formamide either in feed (2500 or 5000 mg/kg) or by abdominal injection (21 570 mg/kg) (Foureman et al. 1994; NTP 2008)
Humans	
	No epidemiological data available

¹ LC₅₀, median lethal concentration; LD₅₀, median lethal dose; LOAEC, lowest-observed-adverse-effect concentration; LOAEL, lowest-observed-adverse-effect level; NOAEL, no-observed-adverse-effect level; NOEC, no-observed-effect concentration.