

Canadian Environmental Protection Act

Priority Substances List
Assessment Report

Benzidine

Government of Canada
Health Canada
Environment Canada

Publié également en français sous le titre de :
Loi canadienne sur la protection de l'environnement
Liste des substances d'intérêt prioritaire
Rapport d'évaluation : Benzidine

CANADIAN CATALOGUING PUBLICATION DATA

Main entry under title:

Benzidine

(Priority substances list assessment report)

Issued also in French under title: Benzidine.

At head of title: *Canadian Environmental
Protection Act.*

Includes bibliographical references.

ISBN 0-662-20895-1

Cat. No. En40-215/20E

1. Benzidine — Toxicity testing — Canada.
2. Benzidine — Environmental aspects.
3. Benzidine dyes — Environmental aspects.
 - I. Canada. Environment Canada.
 - II. Canada. Health Canada.
 - III. Series.

TD195.T48B56 1993 363.73'84 C93-099644-5



Canada	Groupe
Communication	Communication
Group	Canada
Publishing	Édition

©Minister of Supply and Services Canada 1993
Canada Communication Group — Publishing
Ottawa, Canada K1A 0S9
Cat. No. En40-215/20E
ISBN 0-662-20895-1



Printed on Recycled Paper

Table of Contents

Synopsis	v
1.0 Introduction	1
2.0 Summary of Information Critical to Assessment of "Toxic"	3
2.1 Identity, Properties, Production and Uses	3
2.2 Entry into the Environment	3
2.3 Exposure-related Information	4
2.3.1 Fate	4
2.3.2 Concentrations	5
2.4 Effects-related Information	5
2.4.1 Experimental Animals and <i>In Vitro</i>	5
2.4.2 Humans	7
2.4.3 Ecotoxicology	8
3.0 Assessment of "Toxic" under CEPA	9
3.1 CEPA 11(a): Environment	9
3.2 CEPA 11(b): Environment on Which Human Life Depends	9
3.3 CEPA 11(c): Human Life or Health	9
3.4 Conclusion	11
4.0 Recommendations for Research	12
5.0 References	13

Synopsis

Benzidine has been used primarily as an intermediate in the manufacture of dyes and pigments. It is not produced in Canada, and although it may have been imported in small amounts between 1980 and 1987, there no longer appears to be any commercial activity in Canada involving this substance. No conclusive information on the release or occurrence of benzidine in the Canadian environment was identified. Based on available data, benzidine is not expected to persist in the environment.

Available data on the toxic effects of benzidine on aquatic organisms indicate that surface water concentrations in the range of 0.1 mg/L would be required before adverse effects on fish would be expected. Since benzidine is not currently produced in or imported into Canada, and since its half-life in environmental media is less than a few weeks, concentrations of benzidine in surface water in the range of the estimated effect threshold are considered very unlikely. No information on the toxicity of benzidine to wildlife was identified. However, due to the low accumulation of benzidine in aquatic organisms, adverse effects on aquatic-based wildlife due to decreased availability of prey are considered unlikely.

Due to its low volatility, and because it is expected to photooxidize rapidly in air, benzidine is not expected to contribute to ozone depletion, global warming or the formation of ground-level ozone.

Benzidine has been shown to cause cancer in occupationally exposed workers and experimental animals and is therefore considered to be a "non-threshold toxicant" (i.e., a substance for which there is believed to be some chance of adverse effect at any level of exposure). For such substances, where data permit, estimated exposure is compared to quantitative estimates of cancer potency to characterize risk and provide guidance for further action (i.e., analysis of options to reduce exposure). For benzidine, such values would be expected to be low, owing to the lack of confirmed sources of exposure of the general population of Canada to this substance.

Based on these considerations, the Ministers of the Environment and of Health have concluded that benzidine is not entering the environment in a quantity or concentration or under conditions that constitute a danger to the environment or to the environment upon which human life depends. If benzidine were to enter the Canadian environment (as a consequence of its commercial use), however, it might constitute a danger to human life and health. Therefore, benzidine is considered to be "toxic" as defined under section 11 of the *Canadian Environmental Protection Act*.

1.0 Introduction

The *Canadian Environmental Protection Act* (CEPA) requires the Ministers of the Environment and of Health to prepare and publish a Priority Substances List that identifies substances, including chemicals, groups of chemicals, effluents and wastes, that may be harmful to the environment or constitute a danger to human health. The Act also requires both Ministers to assess these substances and determine whether they are "toxic" as interpreted in section 11 of the Act, which states:

"... a substance is toxic if it is entering or may enter the environment in a quantity or concentration or under conditions

- (a) having or that may have an immediate or long-term harmful effect on the environment;
- (b) constituting or that may constitute a danger to the environment on which human life depends; or
- (c) constituting or that may constitute a danger in Canada to human life or health."

Substances assessed as "toxic" according to section 11 may be placed on the List of Toxic Substances (Schedule I of the Act). Consideration can then be given to developing guidelines, codes of practice, or regulations to control any aspect of their life cycle, from the research and development stage through manufacture, use, storage, transport and ultimate disposal.

The assessment of whether benzidine is "toxic", as interpreted under CEPA, was based on the determination of whether it **enters** or is likely to enter the Canadian environment in a concentration or quantities or under conditions that could lead to **exposure** of humans or other biota to levels that could cause harmful **effects**.

Data relevant to the assessment of whether benzidine is "toxic" under CEPA were identified through evaluation of existing review documents (ATSDR, 1989; U.S. EPA, 1980, 1986, 1987; IARC, 1982, 1987), as well as an unpublished review of the environmental behaviour and health effects of this substance prepared under contract by Cambridge Environmental Inc. (Croy and DeVoto, 1990), supplemented with information from published reference texts and literature identified through on-line searches (from 1965 to 1992) of various databases (HSDB, RTECS, IRIS, CCRIS, TOXLINE, TOXLIT, MEDLINE, ENVIROLINE, CHEMICAL ABSTRACTS, BIOLOGICAL ABSTRACTS, ELIAS, AQUAREF, MICROLOG, CODOC). In addition, a number of

provincial authorities were requested to provide any available information on the levels of benzidine in the drinking water in their provinces. The Quebec Ministry of the Environment was requested to provide available quantitative data on potential release of this substance from petrochemical facilities. Data relevant to the assessment of the effects of benzidine on the environment and human health obtained after November 1992 and February 1993, respectively, were not considered for inclusion.

Review articles were consulted where appropriate. However, all original studies that form the basis for determining whether benzidine is "toxic" under CEPA have been critically evaluated by the following staff of Health Canada (human exposure and effects on human health) and Environment Canada (entry and environmental exposure and effects):

R.G. Liteplo (Health Canada)
R.J. Maguire (Environment Canada)
M.E. Meek (Health Canada)

In this report, a summary of technical information critical to the assessment is presented in section 2. The assessment of whether benzidine is "toxic" is presented in section 3. Supporting documentation that discusses the technical information in greater detail has also been prepared and is available upon request.

The environmental sections of this report were reviewed by Drs. C.M. Auer and W.H. Farland of the U.S. Environmental Protection Agency. Sections related to the assessment of effects on human health were approved by the Standards and Guidelines Rulings Committee of the Bureau of Chemical Hazards of Health Canada. The entire Assessment Report was reviewed and approved by the Environment Canada/Health Canada CEPA Management Committee.

Copies of this Assessment Report and the supporting documentation are available upon request from:

Environmental Health Centre
Health Canada
Room 104
Tunney's Pasture
Ottawa, Ontario, Canada
K1A 0L2

Commercial Chemicals
Branch
Environment Canada
14th Floor
Place Vincent Massey
351 Saint-Joseph Boulevard
Hull, Quebec, Canada
K1A 0H3

2.0 Summary of Information Critical to Assessment of "Toxic"

2.1 Identity, Properties, Production and Uses

Benzidine (Chemical Abstracts Service Registry Number 92-87-5) is a primary aromatic amine with the molecular formula $C_{12}H_{12}N_2$. Synonyms for benzidine include 4,4'-biphenyldiamine, 4,4'-diaminodiphenyl and 4,4'-diphenylenediamine (ATSDR, 1989; Croy and DeVoto, 1990). At room temperature, benzidine is white or slightly red, and in the form of either crystals, powder or leaflets (Ferber, 1978). Benzidine has a vapour pressure of 6.6×10^{-2} Pa at 25°C (Mabey *et al.*, 1982), a water solubility of 500 mg/L at 25°C (Bowman *et al.*, 1976) and a log n-octanol/water partition coefficient of 1.34 (Lu *et al.*, 1977).

The commercial manufacture of benzidine involves the alkaline reduction of nitrobenzene (Ferber, 1978). Currently, benzidine does not appear to be produced in or imported into Canada, since no company in Canada reported commercial activity involving more than 10 kilograms of this substance in 1990 (Environment Canada, 1991a, 1991b). Available data indicate that benzidine has been sporadically imported into Canada since 1980: 1980, 4 tonnes; 1981, 12 tonnes; 1982, 0.1 tonne; 1983, 0 tonne; 1984, 0 tonne; 1985, 0.3 tonne; 1986, 0 tonne; and 1987, 1.9 tonnes (Statistics Canada, 1990).

Benzidine has been used primarily as an intermediate in the manufacture of dyes and pigments, and may also be used in the analytical determination of various inorganic cations and anions, in various organic analyses, in the determination of blood in forensic and clinical medicine and as a stain in microscopy (Ferber, 1978; Budavari, 1989).

2.2 Entry into the Environment

No conclusive data on the environmental release of benzidine in Canada were identified. It can enter the environment from any stage in the production, storage, transport, use and disposal of benzidine itself or benzidine-containing materials (such as dyes and pigments), or possibly by atmospheric and water-borne transport from other countries. In water, benzidine can be produced by the photodegradation of 3,3'-dichlorobenzidine (Banerjee *et al.*, 1978). No information on the extent to which benzidine may be formed and released into the environment by this mechanism was identified. Approximately 100 tonnes of 3,3'-dichlorobenzidine were imported into Canada in 1989 (Statistics Canada, 1990). 3,3'-Dichlorobenzidine is on the CEPA Priority Substances List.

2.3 Exposure-related Information

2.3.1 Fate

Oxidation, photochemical transformation, partitioning to sediment or soil, and microbial degradation are expected to be the main pathways of distribution and transformation of benzidine in the environment. Benzidine is not expected to persist in the environment, with overall half-lives in water, soil and air of less than a few weeks. The products formed by the degradation of this substance have not been well characterized.

Benzidine is expected to be slightly volatile (from water), based on its low Henry's law constant of 2.2×10^{-2} Pa m³/mol (Smith *et al.*, 1980). In water, although oxidation (by hydroperoxyl radical or molecular oxygen), biodegradation (Baird *et al.*, 1977; Tabak and Barth, 1978) and photolysis (Bilbo and Wyman, 1953; Larson and Zepp, 1988; Lu *et al.*, 1977; Freitag *et al.*, 1985) may be significant processes, the most important process controlling the fate of benzidine appears to be oxidation by naturally occurring metal cations; the half-life is approximately a few hours (Callahan *et al.*, 1979). Benzidine is quickly absorbed into clays and subsequently oxidized. Although the environmental fate of such complexes is not known with certainty, it is assumed that further oxidation would be facile (Callahan *et al.*, 1979). Estimated half-lives for the biodegradation of benzidine in surface water and groundwater are 31 to 192 h and 96 to 384 h, respectively (Syracuse Research Corp., 1989).

Benzidine is quickly bound in soils and sediments; however, information on the bioavailability of such bound residues was not identified. Zierath *et al.* (1980) noted that benzidine adsorption to soil or sediment was favoured by low pH, and highly correlated with the surface area of the soil or sediment. In soil, benzidine is degraded microbially (Graveel *et al.*, 1985, 1986; Lu *et al.*, 1977). The half-life of benzidine was estimated to be 48 to 192 h for aerobic degradation (Lu *et al.*, 1977).

In air, benzidine is expected to photooxidize moderately rapidly, with an estimated half-life ranging from 0.3 to 3.2 h (Syracuse Research Corp., 1989).

2.3.2 Concentrations

Benzidine was not detected (detection limit = 2 µg/L) in 34 samples of raw and 1 015 samples of treated drinking water obtained in the province of Alberta between 1987 and 1991 (Alberta Environment, 1992). No other data on the concentrations of benzidine within Canada in drinking water, surface water, groundwater, air, biota, soil or sediment, foodstuffs or products containing dyes derived from this substance were identified.

In the United States, benzidine was not detected in a survey of biota and sediment; however, it was detected (but not quantitated) in 1.1% of 1 235 samples of industrial effluent and 0.1% of 879 samples of natural water collected between 1980 and 1982 (Staples *et al.*, 1985).

Benzidine accumulates only moderately in aquatic biota. Bioconcentration factors (after 3 days) were 55 for mosquito fish (*Gambusia affinis*), 293 for *Daphnia magna*, 456 for mosquito larva (*Culex pipiens quinquefasciatus*), 645 for snail (*Physa* sp.) and 2 617 for a filamentous green alga (*Oedogonium cardiacum*) [Lu *et al.*, 1977]. Freitag *et al.* (1985) reported a 5-day bioaccumulation factor in activated sludge of 1 200, a 1-day bioaccumulation factor in algae (*Chlorella fusca*) of 850, and a 3-day bioaccumulation factor in fish (golden orfe, *Leuciscus idus melanotus*) of 83. While some of the results may suggest some potential for the bioaccumulation of benzidine by predator organisms, none has been observed, nor would it be expected for a chemical with a log octanol-water partition coefficient of 1.34.

2.4 Effects-related Information

2.4.1 Experimental Animals and In Vitro

Based on data derived from studies involving predominantly experimental animals, it is apparent that benzidine may be metabolized via a number of metabolic routes (reviewed in Hein, 1988; and Weber and Hein, 1985). One metabolic pathway involves the acetylation of benzidine by cytosolic (acetyl-coenzyme A-dependent) N-acetyltransferase enzymes, which are present in many tissues. Humans (as well as some animal species) may be classified as either "fast" or "slow" acetylators, based on the extent to which they are able to acetylate a variety of chemical substances (Hein, 1988). Based on results of studies on individuals with and without bladder tumours, it has been proposed that this "acetylation polymorphism" may be associated with the development of bladder cancer in individuals exposed to aromatic amines—individuals with a "slow acetylator phenotype" may be more predisposed to develop bladder cancer than individuals with a "fast acetylator phenotype" (Weber and Hein, 1985;

Hein, 1988; Peters *et al.*, 1990 and references therein). Humans are capable of metabolizing benzidine-based azo dyes to benzidine (Martin and Kennelly, 1985; Gregory, 1984).

The carcinogenicity of benzidine has been assessed in a number of animal species. An increased incidence of hepatocellular tumours (carcinomas, adenomas) has been observed in mice exposed to benzidine (in drinking water or in the diet) compared to unexposed controls (Littlefield *et al.*, 1983, 1984; Nelson *et al.*, 1982; Osanai, 1976; Vesselinovitch *et al.*, 1975). Rats administered benzidine (by gastric intubation of the substance dissolved in sesame oil) had a greater incidence of mammary lesions (i.e., carcinomas, adenomas, fibromas and hyperplasia) compared to controls administered vehicle alone (Griswold *et al.*, 1968). The incidence of liver tumours ("hepatomas and cholangiomas") was increased in Syrian hamsters administered benzidine (in the diet), compared to unexposed controls (Saffiotti *et al.*, 1967). In a limited study, Spitz *et al.* (1950, cited in ATSDR, 1989; and U.S. EPA, 1986) reported the development of bladder carcinomas in 3 of 7 dogs administered (orally) benzidine for a period of 5 years. Benzidine is carcinogenic following injection (intraperitoneally; subcutaneously) in rodents (i.e., rats, mice), although such routes of exposure are considered less relevant to the assessment of risk than those by which humans are generally exposed (i.e., oral; inhalation). Results of a limited study in mice indicate that benzidine may induce tumours transplacentally (Vesselinovitch, 1983).

Though benzidine was not mutagenic nor did it bind covalently to DNA in some mammalian cells *in vitro* (Phillips *et al.*, 1990; Oglesby *et al.*, 1983; O'Brien *et al.*, 1990), the weight of evidence convincingly indicates that benzidine is mutagenic and genotoxic (reviewed in ATSDR, 1989; IARC, 1982; U.S. EPA, 1980, 1986; Beland *et al.*, 1983; Beland and Kadlubar, 1985). It is mutagenic in prokaryotic and eukaryotic cells, has transformed a variety of rodent cells in *in vitro* assays, and increased sister chromatid exchange, unscheduled DNA synthesis and induced chromosomal aberrations in eukaryotic cells in *in vivo* and *in vitro* assays. Benzidine induced DNA damage in eukaryotic cells following *in vitro* or *in vivo* exposure, and the covalent binding of benzidine (i.e., its metabolites) to DNA has been observed following the *in vivo* exposure of experimental animals to this substance.

Mice administered drinking water containing benzidine dihydrochloride (20 to 160 mg/L) for their entire lifespan had vacuolation in the brain (Littlefield *et al.*, 1983, 1984). Mice administered (by gavage) benzidine hydrochloride (10.8 to 43.2 mg/kg bw/day) for 5 consecutive days had diminished immunological function (i.e., reduced B- and T-cell mitogenic responses, reduced natural killer cell activity, delayed hypersensitivity responses and reduced resistance to infection) [Luster *et al.*, 1985]. Data on the reproductive and developmental effects of benzidine on experimental animals were limited, and of little significance in assessing the toxicological effects of this substance.

2.4.2 Humans

In case reports and series published since 1927 (cited in IARC, 1982, 1987), the occurrence of bladder cancer in workers in Germany, Switzerland, Italy, England, Japan, France and the United States who had been occupationally exposed to benzidine has been reported.

You *et al.* (1990) reported a significant ($p < 0.01$) standardized incidence ratio (SIR = 19.2) for bladder cancer (14 observed cases) in a group of males ($n = 550$) employed for at least 6 months between 1946 and 1976 in 7 factories in Shanghai producing benzidine-based dyes. The "standardized rate" for bladder cancer increased with increasing duration of exposure to benzidine. The average periods of exposure to benzidine and latency were 8 and 20 years, respectively.

Meigs *et al.* (1986) reported a significant ($p < 0.01$) SIR (3.4, 95% confidence limit (CL) = 1.5 to 6.8) for cancer of the urinary bladder (8 observed cases/2.3 expected cases) for a group of males ($n = 830$) employed for at least 1 day between 1945 and 1965 at a chemical plant in Connecticut producing benzidine and substituted benzidine compounds. SIRs for bladder cancer of 1.8 (95% CL = 0.05 to 10.1; 1 observed/0.55 expected), 0 (95% CL = 0 to 4.7; 0 observed/0.79 expected), 1.9 (95% CL = 0.05 to 10.7; 1 observed/0.52 expected) and 13 (95% CL = 4.8 to 28.4; 6 observed/0.46 expected) were reported for males in the unexposed, low-, medium- and high-exposure groups, (classified based on the duration of exposure to benzidine), respectively; however, a similar trend was not observed for "non-bladder" tumours. SIRs for bladder cancer of 0 (95% CL = 0 to 3.2; 0 observed/1.15 expected), 3.4 (95% CL = 0.4 to 12.4; 2 observed/0.58 expected) and 10 (95% CL = 3.6 to 21.7; 6 observed/0.6 expected) were reported for males employed at the plant from 0 to 1, 1 to 5 and more than 5 years, respectively. The SIR for bladder cancer (4 observed cases) for males occupationally exposed to benzidine between 1945 and 1949, was 9.8 (95% CL = 2.7 to 25), while the SIR based on 1 observed case was 2.1 for workers employed between 1950 and 1954 (95% CL = 0.05 to 11.9). Measures to reduce the exposure of workers to benzidine were introduced in 1950. The average latency period was approximately 20.9 years.

You *et al.* (1990) reported a significant ($p < 0.01$) standardized mortality ratio (SMR = 14.7) for deaths due to bladder cancer (5 observed cases) in a group of males ($n = 550$) employed for at least 6 months between 1946 and 1976 in 7 factories in Shanghai producing benzidine-based dyes.

Morinaga *et al.* (1990) reported a significant ($p < 0.01$) SMR (14.3) for deaths due to cancer of the "urinary organ" (3 observed deaths) in a group of males ($n = 155$) occupationally exposed (between 1945 and 1971) to benzidine at two chemical plants in Osaka, Japan.

Delzell *et al.* (1989) reported a significant ($p < 0.05$) SMR for bladder cancer (SMR = 12.5; 2 observed/0.16 expected)¹ in a group ($n = 379$) of hourly paid "azo-dye" employees exposed to benzidine (in addition to other chemical compounds), although the observed cases of bladder cancer occurred in men who had been previously exposed to benzidine and β -naphthylamine (former workers at the Cincinnati Chemical Works). The azo-dye workers had been employed for at least 12 months (between 1952 and 1985) at a chemical plant in New Jersey. Mortality in a subgroup ($n = 89$) of males previously employed at the Cincinnati Chemical Works was also assessed, and there was a significant ($p < 0.05$) increase in SMRs for deaths due to cancer of the bladder (SMR = 12; 3 observed/0.25 expected), kidney (SMR = 9.5; 2 observed/0.21 expected) and central nervous system (SMR = 9.1; 2 observed/0.22 expected) [Delzell *et al.*, 1989].

Rubino *et al.* (1982) reported a significant ($p < 0.001$) SMR (83.3; 5 observed/0.06 expected) for deaths due to bladder cancer in a group of males ($n = 65$) employed for at least 1 month between 1922 and 1970 at a dyestuff factory in Northern Italy, who had been exposed to benzidine during its manufacture. The mean latency period was 23.4 years.

Case *et al.* (1954) identified 10 deaths due to bladder cancer from 1921 to 1952 in a group (number not specified) of male workers employed in the chemical industry in Britain who had been occupationally exposed to benzidine; the expected number of deaths due to bladder cancer was 0.72.

2.4.3 Ecotoxicology

Limited data on the acute toxicity of benzidine in aquatic organisms were identified. For the red shiner (*Notropis lutrensis*), a 72- and 96-h LC₅₀ of 2.5 mg/L has been reported (Jones, 1980), while for the sheepshead minnow (*Cyprinodon variegatus*), the 96-h LC₅₀ was 64 mg/L (Martin, 1982).

Baird *et al.* (1977) reported that benzidine (20 mg/L) had some (unquantified) inhibitory effect on the respiration of organisms in activated sludge while this substance was being degraded, suggesting that a metabolite or metabolites may be responsible for the observed toxicity.

No data on the toxicity of benzidine to wild mammals, birds, sediment or soil biota were identified. Because of the low accumulation of benzidine by aquatic organisms, adverse effects on aquatic-based wildlife due to decreased availability of prey are considered unlikely.

1. SMRs for death due to all cancers (SMR = 1.9; 16 observed/8.3 expected), cancer of the stomach (SMR = 9.7; 3 observed/0.31 expected), and central nervous system (SMR = 9.1; 3 observed/0.33 expected) were also significantly ($p < 0.05$) increased.

3.0 Assessment of "Toxic" under CEPA

3.1 CEPA 11(a): Environment

The most sensitive species of fish identified is the red shiner (*Notropis lutrensis*) with a 72- and 96-hour LC₅₀ of 2.5 mg/L. This concentration was divided by a factor of 20 to convert it to a chronic no-observed-effect-level, to account for interspecies differences and to extrapolate laboratory results to the field. This yielded an estimated effect threshold of 0.13 mg/L. Since benzidine is not currently produced in or imported into Canada, and since its half-life in environmental media is less than a few weeks, concentrations of benzidine in surface water in the range of the estimated effect threshold are considered very unlikely.

Therefore, on the basis of the limited available data, benzidine is not considered to be "toxic" as interpreted under paragraph 11(a) of CEPA.

3.2 CEPA 11(b): Environment on Which Human Life Depends

Benzidine is expected to be slightly volatile and to photooxidize rapidly in air. Therefore, this substance is not expected to contribute to ozone depletion, global warming or the formation of ground-level ozone.

Therefore, on the basis of available data, benzidine is not considered to be "toxic" as interpreted under paragraph 11(b) of CEPA.

3.3 CEPA 11(c): Human Life or Health

Population Exposure

Quantitative data on the concentrations of benzidine in air, drinking water, soil or food-stuffs within Canada (or elsewhere) were not identified. Consequently, the available data are inadequate to estimate the exposure of the general population of Canada to benzidine.

Effects

The results of a number of analytical epidemiological studies as well as supporting data from case reports and series of workers occupationally exposed to benzidine have provided clear evidence for the carcinogenicity of this substance in humans. Indeed, the observed association between the occurrence of bladder carcinoma and occupational exposure to benzidine fulfils the traditional criteria (consistency, strength, specificity, temporal relationship, exposure-response relationship and plausibility) for assessment of causality in epidemiological studies.

The observed associations have been very specific, in that occupational exposure to benzidine has been associated with an increased incidence of, or death due to, cancer of the bladder—almost exclusively, transitional cell carcinoma. The results have been remarkably consistent, with an association between occupational exposure to benzidine and an increased incidence of, or mortality due to, bladder cancer observed in all the analytical epidemiological studies (Meigs *et al.*, 1986; You *et al.*, 1990; Morinaga *et al.*, 1990; Delzell *et al.*, 1989; Rubino *et al.*, 1982; Case *et al.*, 1954) in which these relationships were examined.

The association between the increased incidence of, or mortality due to, bladder carcinoma is strong. Reported standardized incidence ratios (SIRs) for bladder cancer in occupationally exposed workers are 3.4 (Meigs *et al.*, 1986) and 19.2 (You *et al.*, 1990). Reported standardized mortality ratios (SMRs) for death due to bladder cancer in occupationally exposed workers range from 12 (Delzell *et al.*, 1989) to 83.3 (Rubino *et al.*, 1982).

Although quantitative information on exposure to benzidine was not assessed in any of the available analytical epidemiological studies, a relationship between qualitative measures of exposure and an increased incidence of bladder cancer was reported in two studies (You *et al.*, 1990; Meigs *et al.*, 1986). Although the data are limited, there is evidence indicating that a reduction in the (occupational) exposure to benzidine was associated with a decrease in the incidence of bladder carcinoma (Meigs *et al.*, 1986).

The carcinogenicity of benzidine in humans is plausible, based on the overwhelming evidence of the genotoxicity of this substance. Moreover, the carcinogenicity of benzidine in experimental animals (i.e., rats, mice, hamsters) has been well documented.

Since the observed association of bladder cancer (predominantly transitional cell carcinoma) with occupational exposure to benzidine fulfils the traditional criteria for assessment of causality in epidemiological studies, on the basis of the available data,

benzidine has been classified in Group I (Carcinogenic to Humans) of the classification scheme developed for the determination of "toxic" under paragraph 11(c) of CEPA (EHD, 1992).

For such substances, where possible, estimated total daily intake by the general population in Canada is compared to quantitative estimates of carcinogenic potency to characterize risk and provide guidance for further action (i.e., analysis of options to reduce exposure). Owing to the lack of available information on concentrations of benzidine in environmental media to which humans are exposed, it is not possible to quantitatively estimate the total daily intake of this substance by the general population of Canada. Consequently, estimates of total daily intake have not been compared to quantitative estimates of cancer potency, although such values would be expected to be low owing to the lack of reported use of this substance in Canada.

Benzidine has been classified as being "Carcinogenic to Humans", and is therefore considered to be "toxic" under paragraph 11(c) of CEPA.

This approach is consistent with the objective that exposure to non-threshold toxicants should be reduced wherever possible, and obviates the need to establish an arbitrary *de minimis* level of risk for determination of "toxic" under CEPA.

3.4 Conclusion

Based on the available data, benzidine is not considered to be "toxic" as defined under paragraphs 11(a) or 11(b) of CEPA. Benzidine is considered to be "toxic" as defined under paragraph 11(c) of CEPA.

4.0 Recommendations for Research

Although a number of data gaps on the effects of benzidine on the environment and human health were identified, because of the negligible exposure of biota and the general population of Canada to this substance the priority for additional research is considered to be low.

5.0 References

Alberta Environment. 1992. Personal communication with G.P. Halina. Environmental Protection Services, Alberta Environment.

ATSDR (Agency for Toxic Substances and Disease Registry). 1989. Toxicological Profile for Benzidine. U.S. Public Health Service. 113 pp.

Baird, R., L. Carmona, and R.L. Jenkins. 1977. Behaviour of benzidine and other aromatic amines in aerobic wastewater treatment. *J. Water Pollut. Contr. Fed.* 49: 1609-1615.

Banerjee, S., H.C. Sikka, R. Gray, and C.M. Kelly. 1978. Photodegradation of 3,3'-dichlorobenzidine. *Environ. Sci. Technol.* 12: 1425-1427.

Béland, F.A., D.T. Beranek, K.L. Dooley, R.H. Heflich, and F.F. Kadlubar. 1983. Arylamine-DNA adducts in vitro and in vivo: Their role in bacterial mutagenesis and bladder carcinogenesis. *Environ. Health Perspec.* 49: 125-134.

Béland, F.A., and F.F. Kadlubar. 1985. Formation and persistence of arylamine DNA adducts in vivo. *Environ. Health Perspec.* 62: 19-30.

Bilbo, A.J., and G.M. Wyman. 1953. Steric hindrance to coplanarity in o-fluorobenzidines. *J. Am. Chem. Soc.* 75: 5312-5314.

Bowman, M.C., J.R. King, and C.L. Holder. 1976. Benzidine and congeners: analytical chemical properties and trace analysis in five substrates. *Inter. J. Environ. Anal. Chem.* 4: 205-223.

Budavari, S., ed. 1989. *The Merck Index*, Merck and Co., Rahway, New Jersey.

Callahan, M.A., M.W. Slimak, N.W. Gabel, I.P. May, C.F. Fowler, J.R. Freed, P. Jennings, R.L. Durfee, F.C. Whitmore, B. Maestri, W.R. Mabey, B.R. Holt, and C. Gould. 1979. Water-related environmental fate of 129 priority pollutants, Volume II: halogenated aliphatic hydrocarbons, halogenated ethers, monocyclic aromatics, phthalate esters, polycyclic aromatic hydrocarbons, nitrosamines and miscellaneous compounds. U.S. Environmental Protection Agency, Washington. EPA 440/4-79-029b.

Case, R.A.M., M.E. Hosker, D.B. McDonald, and J.T. Pearson. 1954. Tumours of the urinary bladder in workmen engaged in the manufacture and use of certain dyestuff intermediates in the British chemical industry. *Br. J. Ind. Med.* 11: 75-104.

Croy, R.G., and E. DeVoto. 1990. Benzidine: A Review of Environmental Behavior and Health Effects. Prepared for Priority Substances Section, Health Protection Branch, Health Canada, Ottawa.

Delzell, E., M. Macaluso, and P. Cole. 1989. A follow-up study of workers at a dye and resin manufacturing plant. *J. Occup. Med.* 31: 273–278.

EHD (Environmental Health Directorate). 1992, unpublished. Determination of "Toxic" Under Paragraph 11(c) of the Canadian Environmental Protection Act 1st edition, 20 November 1992, Bureau of Chemical Hazards.

Environment Canada. 1991a. Canadian Environmental Protection Act notice with respect to certain aromatic amine substances and their salts. *Canada Gazette*, Part I, Queen's Printer for Canada, Ottawa. August 10: 2580–2583.

Environment Canada. 1991b. Canadian Environmental Protection Act notice with respect to certain aromatic amine substances and their salts. Preliminary results of the aromatic amines notice. Use Patterns Section, Commercial Chemicals Branch, Environmental Protection, Ottawa.

Ferber, K.H. 1978. Benzidine and related biphenyldiamines. In: H.F. Mark, D.F. Othmer, C.G. Overberger, and G.T. Seaborg, eds. *Kirk-Othmer Encyclopaedia of Chemical Technology*, 3rd ed., Volume 3. John Wiley and Sons, Toronto: 772–777.

Freitag, D., L. Ballhorn, H. Geyer, and F. Korte. 1985. Environmental hazard profile for organic chemicals. An experimental method for the assessment of the behaviour of organic chemicals in the ecosphere by means of simple laboratory tests with ¹⁴C-labelled chemicals. *Chemosphere* 14: 1589–1616.

Graveel, J.G., L.E. Sommers, and D.W. Nelson. 1986. Decomposition of benzidine, α -naphthylamine and p-toluidine in soils. *J. Environ. Qual.* 15: 53–59.

Graveel, J.G., L.E. Sommers, and D.W. Nelson. 1985. Sites of benzidine, α -naphthylamine and p-toluidine retention in soils. *Environ. Toxicol. Chem.* 4: 607–613.

Gregory, A.R. 1984. The carcinogenic potential of benzidine-based dyes. *J. Environ. Pathol. Toxicol. Oncol.* 5: 243–259.

Griswold, D.P., A.E. Casey, E.K. Weisburger, and J.H. Weisburger. 1968. The carcinogenicity of multiple intragastric doses of aromatic and heterocyclic nitro or amino derivatives in young female Sprague-Dawley rats. *Cancer Res.* 28: 924–933.

Hein, D.W. 1988. Acetylator genotype and arylamine-induced carcinogenesis. *Biochim. Biophys. Acta* 948: 37–66.

IARC (International Agency for Research on Cancer). 1982. *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. Some Industrial Chemicals and Dyestuffs*, Volume 29. Lyon, France: 149–183.

IARC (International Agency for Research on Cancer). 1987. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. Overall Evaluation of Carcinogenicity: An Updating of IARC Monographs Volumes 1 to 42, Supplement 7. Lyon, France: 123-125.

Jones, T.C. 1980. Risk assessment, phase 1: benzidine, its congeners and their derivative dyes and pigments. U.S. Environmental Protection Agency, Washington. EPA 560/11-80-019.

Larson, R.A., and R.G. Zepp. 1988. Reactivity of the carbonate radical with aniline derivatives. *Environ. Toxicol. Chem.* 7: 265-274.

Littlefield, N.A., C.J. Nelson, and C.H. Frith. 1983. Benzidine dihydrochloride: Toxicological assessment in mice during chronic exposures. *J. Toxicol. Environ. Health* 12: 671-685.

Littlefield, N.A., C.J. Nelson, and D.W. Gaylor. 1984. Benzidine dihydrochloride: Risk assessment. *Fund. Appl. Toxicol.* 4: 69-80.

Lu, P.-Y., R.L. Metcalf, N. Plummer, and D. Mandel. 1977. The environmental fate of three carcinogens: benzo(a)pyrene, benzidine and vinyl chloride evaluated in laboratory model ecosystems. *Arch. Environ. Contam. Toxicol.* 6: 129-142.

Luster, M.I., A.N. Tucker, H.T. Hayes, O.J. Pung, T. Burka, R. McMillan, and T. Eling. 1985. Immunosuppressive effects of benzidine in mice: evidence of alterations in arachidonic acid metabolism. *J. Immunol.* 135: 2754-2761.

Mabey, W.R., J.H. Smith, R.T. Podoll, H.L. Johnson, T. Mill, T.-W. Chou, J. Gates, I.W. Partridge, H. Jaber, and D. Vandenberg. 1982. Aquatic fate process data for organic priority pollutants. U.S. Environmental Protection Agency, Washington. EPA 440/4-81-014.

Martin, B.J. 1982. Development of a carcinogen assay system utilizing estuarine fishes. U.S. Environmental Protection Agency, Gulf Breeze, Florida. EPA 600/3-82-091. 50+ pp.

Martin, C.N., and J.C. Kennelly. 1985. Metabolism, mutagenicity, and DNA binding of biphenyl-based azodyes. *Drug Metabol. Rev.* 16: 89-117.

Meigs, J.W., L.D. Marrett, F.U. Ulrich, and J.T. Flannery. 1986. Bladder tumor incidence among workers exposed to benzidine: A thirty-year follow-up. *J. Natl. Cancer Inst.* 76: 1-8.

Morinaga, K., S. Yutani, and I. Hara. 1990. Cancer mortality of male workers exposed to benzidine and/or β -naphthylamine. *Jpn. J. Hyg.* 45: 909-918. [Japanese]

- Nelson, C.J., K.P. Baetcke, C.H. Frith, R.L. Kodell, and G. Schieferstein. 1982. The influence of sex, dose, time, and cross on neoplasia in mice given benzidine dihydrochloride. *Toxicol. Appl. Pharmacol.* 64: 171-186.
- O'Brien, K.A.F., D.G. Gatehouse, and M. Tiley. 1990. Induction of mutations in TK6 human lymphoblastoid cells by ethyl methanesulphonate, benzo[a]pyrene and benzidine. *Mutagenesis* 5 [Supplement]: 55-60.
- Oglesby, L.A., C. Hix, L. Snow, P. MacNair, M. Seig, and R. Langerbach. 1983. Bovine bladder urothelial cell activation of carcinogens to metabolites mutagenic in Chinese hamster V-79 cells and *Salmonella typhimurium*. *Cancer Res.* 43: 5194-5199.
- Osanai, H. 1976. An experimental study on hepatoma caused by aromatic amines. *Jpn. J. Sci. Labour* 52: 179-201.
- Peters, J.H., G.H. Gordon, E. Lin, C.E. Green, and C.A. Tyson. 1990. Polymorphic N-acetylation of sulfamethazine and benzidine by human liver: Implications for cancer risk. *Anticancer Res.* 10: 225-230.
- Phillips, D.H., M.F. Cross, J.C. Kennelly, P. Wilcox, and M.R. O'Donovan. 1990. Determination of benzidine-DNA adduct formation in CHO, HeLa, L5178Y, TK6 and V79 cells. *Mutagenesis* 5 [Supplement]: 67-69.
- Rubino, G.F., G. Scansetti, G. Piolatto, and E. Pira. 1982. The carcinogenic effect of aromatic amines: An epidemiological study on the role of *o*-toluidine and 4,4'-methylene bis (2-methylaniline) in inducing bladder cancer in man. *Environ. Res.* 27: 241-254.
- Saffiotti, U., F. Cefis, R. Montesano, and A.R. Sellakumar. 1967. Induction of bladder cancer in hamsters fed aromatic amines. In: W.B. Deichmann, K.F. Lampe, R.A. Penalver, and A. Soto eds., *Bladder Cancer, A Symposium*. Aesculapis Publishing Co., Birmingham, Alabama: 129-135.
- Smith, J.H., D.C. Bomberger, Jr., and D.L. Haynes. 1980. Prediction of the volatilization rates of high-volatility chemicals from natural water bodies. *Environ. Sci. Technol.* 14: 1332-1337.
- Staples, C.A., A.F. Werner, and T.J. Hoogheem. 1985. Assessment of priority pollutant concentrations in the United States using STORET data base. *Environ. Toxicol. Chem.* 4: 131-142.
- Statistics Canada. 1990. Imports, merchandise trade commodity detail, various years. Ottawa.

Syracuse Research Corp. 1989. Chemical fate rate constants for SARA Section 313 chemicals and Superfund health evaluation manual chemicals. Report (EPA 68-02-4254, Versar Task 176, SRC F0107-10, EPA 68-C8-0004, SRC F0111-119) to U.S. Environmental Protection Agency, Office of Toxic Substances, Washington: 267-268.

Tabak, H.H., and E.F. Barth. 1978. Biodegradability of benzidine in aerobic suspended growth reactors. *J. Water Pollut. Contr. Fed.* 50: 552-558.

U.S. EPA. 1980. Ambient Water Quality Criteria for Benzidine. Office of Water Regulations and Standards, U.S. Environmental Protection Agency.

U.S. EPA. 1986. Health and Environmental Effects Profile for Benzidine. Environmental Criteria and Assessment Office, U.S. Environmental Protection Agency.

U.S. EPA. 1987. Health Effects Assessment for Benzidine. Environmental Criteria and Assessment Office, U.S. Environmental Protection Agency.

Vesselinovitch, S.D. 1983. Perinatal hepatocarcinogenesis. *Biol. Res. Pregnancy and Perinatol.* 4: 22-25.

Vesselinovitch, S.D., K.V.N. Rao, and N. Mihailovich. 1975. Factors modulating benzidine carcinogenicity bioassay. *Cancer Res.* 35: 2814-2819.

Weber, W.W., and D.W. Hein. 1985. N-Acetylation pharmacogenetics. *Pharmacol. Rev.* 37: 25-79.

You, X.-Y., J.-G. Chen, and Y.-N. Hu. 1990. Studies on the relation between bladder cancer and benzidine or its derived dyes in Shanghai. *Br. J. Ind. Med.* 47: 544-552.

Zierath, D.L., J.P. Hassett, W.L. Banwart, S.G. Wood, and J.C. Means. 1980. Sorption of benzidine by sediments and soils. *Soil Sci.* 129: 277-281.

TD 195 T48 B46 1993 c. 1
Canada. Environment Canada
Benzidine

LIBRARY
CANADA CENTRE FOR INLAND WATERS
867 LAKESHORE ROAD
BURLINGTON, ONTARIO, CANADA
L7R 4A6