3,3′-Dimethylbenzidine and Dyes Metabolized to 3,3′-Dimethylbenzidine

Introduction

3,3′-Dimethylbenzidine was first listed in the *Third Annual Report on Carcinogens* (1983) and was re-reviewed for the *Tenth Report on Carcinogens* (2002). 3,3′-Dimethylbenzidine-based dyes that are metabolized to 3,3′-dimethylbenzidine (3,3′-dimethylbenzidine dyes class) were first listed in the *Tenth Report on Carcinogens* (2002). The profiles for 3,3′-dimethylbenzidine and dyes metabolized to 3,3′-dimethylbenzidine, which are listed (separately) as *reasonably anticipated to be human carcinogens*, follow this introduction.

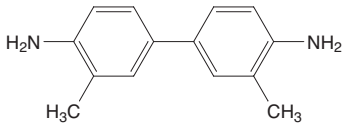
3,3′-Dimethylbenzidine

CAS No. 119-93-7

Reasonably anticipated to be a human carcinogen

First listed in the *Third Annual Report on Carcinogens* (1983)

Also known as *o*-tolidine



Carcinogenicity

3,3′-Dimethylbenzidine is *reasonably anticipated to be a human carcinogen* based on sufficient evidence of carcinogenicity from studies in experimental animals.

Cancer Studies in Experimental Animals

3,3′-Dimethylbenzidine caused tumors in rats at numerous tissue sites and by two different routes of exposure. Administration of the dihydrochloride salt of 3,3′-dimethylbenzidine in the drinking water caused benign and/or malignant tumors of the Zymbal gland (adenoma or carcinoma), liver (hepatocellular adenoma or carcinoma), skin (basal-cell adenoma or squamous-cell papilloma or carcinoma), preputial and clitoral glands (adenoma or carcinoma), and large intestine (adenomatous polyps) in rats of both sexes. In males, it also caused cancer of the small intestine (adenocarcinoma) and benign lung tumors (adenoma), and in females, it also caused mammary-gland cancer (adenocarcinoma) and benign or malignant oral-cavity tumors (squamous-cell papilloma or carcinoma) (NTP 1991). Exposure of rats (of unspecified sex) to commercial-grade or purified 3,3′-dimethylbenzidine by subcutaneous injection primarily caused Zymbal-gland cancer (carcinoma) (IARC 1972).

Cancer Studies in Humans

No epidemiological studies were identified that evaluated the relationship between human cancer and exposure specifically to 3,3′-dimethylbenzidine. However, one study found that occupational exposure to a mixture of arylamine amine products, including 3,3′-dimethylbenzidine, was associated with an increased incidence of urinary-bladder cancer (Ouellet-Hellstrom and Rench 1996).

Properties

3,3′-Dimethylbenzidine is an aromatic amine that exists at room temperature as a white-to-reddish powder or crystals. It is slightly soluble in water and soluble in ethanol, ether, and dilute acids. It is stable at normal temperatures and pressures (Akron 2009, HSDB 2009). Physical and chemical properties of 3,3′-dimethylbenzidine are listed in the following table

Property	Information
Molecular weight	212.3[a]
Specific gravity	1.234 g/mL[b]
Melting point	129°C to 131°C[a]
Boiling point	300°C[a]
Log K_{ow}	2.34[a]
Water solubility	1.3 g/L at 25°C[a]
Vapor pressure	6.9×10^{-7} mm Hg at 25°C[a]
Dissociation constant (pK_a)	4.5 at 25°C[a]

Sources: [a]HSDB 2009, [b]Akron 2009.

Use

Dimethylbenzidine is used as a dye or as an intermediate for producing dyestuffs and pigments; it is used to color textiles, leather, plastic, rubber, and paper (see Dyes Metabolized to 3,3′-Dimethylbenzidine, below). 3,3′-Dimethylbenzidine is also used as a laboratory reagent for the detection of blood, gold, and chlorine and as a curing agent for urethane resins (IARC 1972, NTP 1991, HSDB 2009).

Production

In 2009, 3,3′-dimethylbenzidine was produced by seven manufacturers worldwide, including one in the United States, one each in Europe and East Asia, and two each in China and India (SRI 2009), and was available from 34 suppliers, including 20 U.S. suppliers (ChemSources 2009).

Exposure

The routes of potential human exposure to 3,3′-dimethylbenzidine are inhalation, dermal contact, and ingestion (IARC 1972, HSDB 2009). The general population may be exposed through contact with dyes or pigments in final consumer products that may contain small residual amounts of 3,3′-dimethylbenzidine (NTP 1991). Indirect exposure may occur through dimethylbenzidine-based dyes that can be metabolized to 3,3′-dimethylbenzidine by the liver or bacteria in the gastrointestinal tract (NTP 1991) or on the skin. Although dimethylbenzidine-based dyes may not be absorbed dermally to any substantial degree, 3,3′-dimethylbenzidine itself is known to easily penetrate the skin, and it was demonstrated that an azo dye (C.I. direct blue 14) could be metabolized by numerous strains of common skin bacteria to yield 3,3′-dimethylbenzidine (Platzek *et al.* 1999).

According to the U.S. Environmental Protection Agency's Toxics Release Inventory, no more than 270 lb of 3,3′-dimethylbenzidine has been released to the environment in any year since 1989, and no more than 17 lb has been released to air. In 2007, one facility released 10 lb of 3,3′-dimethylbenzidine (TRI 2009). If released to air, 3,3′-dimethylbenzidine will exist as both a vapor and particulate. The vapor fraction will be degraded by photochemically produced hydroxyl radicals, with a half-life of 3 hours, and it may be sensitive to direct photolysis. If released to water, 3,3′-dimethylbenzidine is expected to adsorb to suspended solids and sediment, and not to volatilize or undergo hydrolysis. If released to soil, it is expected to bind to humic materials with moderate mobility at neutral pH; however, mobility may decrease under acidic conditions. Limited data suggest that 3,3′-dimethylbenzidine may slowly biodegrade in the environment (HSDB 2009).

Workers potentially exposed to 3,3′-dimethylbenzidine include dye makers, repackagers of 3,3′-dimethylbenzidine and dimethylbenzidine-based dyes, and analytical chemistry laboratory workers (NIOSH 1978). It has been recommended that analytical methods

for determining chlorine in water using 3,3′-dimethylbenzidine be replaced with the methyl-orange method, which uses less hazardous reagents (IPCS 1982). 3,3′-Dimethylbenzidine was found in the urine of dye-manufacturing workers who had been indirectly exposed through contact with 3,3′-dimethylbenzidine-based dyes (NTP 1991). The National Occupational Exposure Survey (conducted from 1981 to 1983) estimated that 9,640 workers including 6,005 women (mostly in the health-services sector), potentially were exposed to 3,3′-dimethylbenzidine (NIOSH 1990).

Regulations

Department of Transportation (DOT)

Toxic dyes and toxic dye intermediates are considered hazardous materials, and special requirements have been set for marking, labeling, and transporting these materials.

Environmental Protection Agency (EPA)

Clean Air Act

National Emission Standards for Hazardous Air Pollutants: Listed as a hazardous air pollutant.

Comprehensive Environmental Response, Compensation, and Liability Act

Reportable quantity (RQ) = 10 lb.

Emergency Planning and Community Right-To-Know Act

Toxics Release Inventory: Listed substance subject to reporting requirements.

Resource Conservation and Recovery Act

Listed Hazardous Waste: Waste code for which the listing is based wholly or partly on the presence of 3,3′-dimethylbenzidine = U095.
Listed as a hazardous constituent of waste.

Guidelines

American Conference of Governmental Industrial Hygienists (ACGIH)

Potential for dermal absorption.

National Institute for Occupational Safety and Health (NIOSH, CDC, HHS)

Ceiling recommended exposure limit = 0.02 mg/m^3 (60-min exposure).
Potential for dermal absorption.
Listed as a potential occupational carcinogen.

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Dyes Metabolized to 3,3′-Dimethylbenzidine (3,3′-Dimethylbenzidine Dye Class)

CAS No.: none assigned

Reasonably anticipated to be human carcinogens

First listed in the *Tenth Report on Carcinogens* (2002)

Carcinogenicity

3,3′-Dimethylbenzidine-based dyes that are metabolized to 3,3′-dimethylbenzidine are *reasonably anticipated to be human carcinogens* based cancer studies in experimental animals and studies on mechanisms of carcinogenesis, which have provided the following evidence:

- 3,3′-Dimethylbenzidine is carcinogenic in rats of both sexes.
- Metabolism of 3,3′-dimethylbenzidine-based dyes to release free 3,3′-dimethylbenzidine is a generalized phenomenon that occurs in all animal species studied (Lynn *et al.* 1980, Bowman *et al.* 1982).
- A representative 3,3′-dimethylbenzidine-based dye, C.I. acid red 114, is carcinogenic in experimental animals.
- The profile of tumors caused by 3,3′-dimethylbenzidine and acid red 114 is similar to that caused by structurally related chemicals.

Cancer Studies in Experimental Animals

Oral exposure to a representative 3,3′-dimethylbenzidine-based dye, acid red 114, caused tumors in rats at numerous tissue sites (NTP 1991a). Acid red 114 administered in the drinking water caused benign and/or malignant tumors of the liver, skin, and Zymbal gland in rats of both sexes and of the clitoral gland, oral-cavity epithelium, small and large intestines, and lung in female rats. Increased incidences of tumors of the oral-cavity epithelium, adrenal gland, and lung in male rats and of mononuclear-cell leukemia and cancer of the mammary gland (adenomcarcinoma) and adrenal gland (pheochromocytoma) in female rats also may have been related to exposure to acid red 114.

Cancer Studies in Humans

No epidemiological studies were identified that evaluated the relationship between human cancer and exposure specifically to 3,3′-dimethylbenzidine-based dyes.

Studies on Mechanisms of Carcinogenesis

3,3′-Dimethylbenzidine is structurally similar to benzidine, which is listed in the Report on Carcinogens as *known to be a human carcinogen*, and 3,3′-dimethoxybenzidine, which is listed as *reasonably anticipated to be a human carcinogen*. The pattern of tumors observed for acid red 114 (NTP 1991a) and 3,3′-dimethylbenzidine (NTP 1991b) in rats is similar to that observed for the structurally similar chemical 3,3′-dimethoxybenzidine (NTP 1990) and the 3,3′-dimethoxybenzidine-based dye C.I. direct blue 15 (NTP 1992). In rats, these four chemicals caused tumors of the skin, Zymbal gland, liver, oral cavity, gastrointestinal tract, preputial gland, and clitoral gland, as well as at other tissue sites.

Like benzidine and 3,3′-dimethoxybenzidine, 3,3′-dimethylbenzidine is used to synthesize many dyes through azo linking of various chromophores to the base chemical. The azo bonds of 3,3′-dimethylbenzidine-based dyes are chemically similar regardless of the chromophore used, and they are easily broken via reduction by chemicals or enzymes to form free 3,3′-dimethylbenzidine and free chromophore(s). A number of bacteria catalyze this process, in-

cluding *Escherichia coli*, which is found in the human gastrointestinal tract (Cerniglia *et al.* 1982, Morgan *et al.* 1994). This reductive process has also been found in rats, hamsters, and dogs (Lynn *et al.* 1980, Bowman *et al.* 1983, Nony *et al.* 1983). Bacteria in the animals' gastrointestinal tract are thought to be the primary agents of this metabolism (Cerniglia *et al.* 1982, Morgan *et al.* 1994). 3,3′-Dimethylbenzidine-based dyes are mutagenic in bacteria when tested with mammalian metabolic activation and an azo-reductive preincubation protocol (NTP 1991a). It is assumed that the breakdown process results in the formation of 3,3′-dimethylbenzidine, which is known to cause mutations in bacteria (Haworth *et al.* 1983).

There is no evidence to suggest that mechanisms by which these substances cause tumors in experimental animals would not also operate in humans.

Properties

All dimethylbenzidine-based dyes have the characteristic diazotized dimethylbenzidine nucleus, but they differ with respect to the chemical groups attached at the diazo linkages. They all exist as colored powders (in a wide range of hues) at room temperature and have negligible vapor pressures. Their water solubility varies, but it is sufficient for dyeing in aqueous solution. There are no rigid chemical specifications for dyes, including dimethylbenzidine-based dyes; therefore, their composition varies according to the shade and intensity requirements of the customer. Also, because various dyes are mixed together to produce particular colors, the final products are more accurately described as mixtures of substances than as specific chemical compounds (NIOSH 1980).

Use

3,3′-Dimethylbenzidine-based dyes and pigments are used in printing textiles, as biological stains, and in color photography (HSDB 2009). In 1972, the Colour Index listed nearly 100 dyes derived from 3,3′-dimethylbenzidine (IARC 1972).

Production

In 2009, no information on production of dyes previously identified as derived from 3,3′-dimethylbenzidine was found; however, the dyes were available from numerous suppliers worldwide, including at least ten U.S. suppliers (ChemSources 2009). All synthetic organic dyes and dye intermediates are included in a single category for reporting of U.S. imports and exports; no data were available on imports or exports of specific dye products.

Exposure

The general population may be exposed via contact with textiles and papers containing 3,3′-dimethylbenzidine-based dyes (NTP 1991a). Occupational exposure to 3,3′-dimethylbenzidine-based dyes may occur by inhalation of dust or mist, accidental ingestion, or dermal contact. Most occupational exposure is of workers in dye-manufacturing and -processing plants. In 1986–87, the U.S. Environmental Protection Agency, the American Textile Manufacturers Institute, and the Toxicological Association of the Dyestuffs Manufacturing Industry conducted a joint survey to estimate airborne concentrations of dye dust in dye-weighing rooms of facilities where powdered dyes were used to dye and print textiles. The estimated mean airborne concentration of total dye in 24 randomly monitored plants was 0.085 mg/m^3 (EPA 1990). Workers in other occupations may be exposed to small quantities of 3,3′-dimethylbenzidine-based dyes, including water and sewage plant attendants, chemical test tape or kit makers, and swimming-pool service representatives (NIOSH 1978)

Regulations

Department of Transportation (DOT)

Toxic dyes and toxic dye intermediates are considered hazardous materials, and special requirements have been set for marking, labeling, and transporting these materials.

Environmental Protection Agency (EPA)

Comprehensive Environmental Response, Compensation, and Liability Act

Reportable quantity (RQ) for trypan blue = 10 lb.

Emergency Planning and Community Right-To-Know Act

Toxics Release Inventory: C.I. acid red 114 and trypan blue are listed substances subject to reporting requirements.

Resource Conservation and Recovery Act

Listed Hazardous Waste: Waste code for which the listing is based wholly or partly on the presence of trypan blue = U236.

Trypan blue is listed as a hazardous constituent of waste.

Occupational Safety and Health Administration (OSHA, Dept. of Labor)

3,3′-Dimethylbenzidine-based dyes are listed as potential occupational carcinogens.

Guidelines

National Institute for Occupational Safety and Health (NIOSH, CDC, HHS)

3,3′-Dimethylbenzidine-based dyes are listed as potential occupational carcinogens.

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