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Abstract

The following aspects of benzothiazoles are reviewed: (1) physical and chemical properties, (2) analysis and occurrence, (3) biological properties and (4) mammalian toxicity and human health. Six reports were found in the literature on the occurrence of benzothiazoles in the following areas of the aquatic environment: river water (1-2 ppb), drinking water, and tire plant wastewater (20-60 ppb). Benzothiazoles display a wide range of biological activity and are used commercially as fungicides. No data were found for toxicity to aquatic organisms, although acute mammalian toxicity is low. Future research areas are identified, the most important of which are (1) physical properties, (2) persistence and (3) aquatic toxicity.

Résumé

Le présent rapport étudie les benzothiazoles des points de vue suivants: (1) propriétés physico-chimiques, (2) analyse et concentration, (3) propriétés biologiques et (4) toxicité pour les mammifères et répercussions sur la santé. Six travaux ont été trouvés qui traitaient des concentrations des benzothiazoles dans le milieu aquatique, c'est-à-dire dans les cours d'eau (1 à 2 parties par milliard), l'eau potable, et les eaux résiduaires des usines de fabrication de pneus (20 à 60 parties par milliard). Les benzothiazoles montrent de nombreuses activités biologiques et servent industriellement en tant que fongicides. Aucunes données n'ont été trouvées sur leur toxicité pour les organismes aquatiques, bien que leur toxicité aiguë à l'égard des mammifères soit faible. Les domaines de la recherche à effectuer sont déterminés, dont les plus importants concernent: (1) les propriétés physiques, (2) la persistance et (3) la toxicité dans le milieu aquatique.

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INTRODUCTION

In 1978, a study was begun on the fate of organic contaminants in Canagagigue Creek below Elmira, Ontario. When the authors found that benzothiazoles were the major components in the neutral fraction of water extracts, they reviewed the literature on these substances in the aquatic environment. The UNIROYAL CHEMICALS plant in Elmira manufactures benzothiazole derivatives for the rubber industry. Seven compounds were chosen on the basis of preliminary analyses and a product list (Canada Department of Industry, Trade and Commerce, 1971) for this plant. The survey included *Chemical Abstracts* for 1967–80 and *Biological Abstracts* for 1972–80. The structural formulae for these compounds are presented in Figure 1. Ring numbering is shown for benzothiazole, the parent compound. Listed below are the abbreviations used in this report along with their chemical names, synonyms and *Chemical Abstracts* registry numbers:

Abbreviation	Chemical name	Registry No.
BT	Benzothiazole	[95-16-9]
MBT	2-Mercaptobenzothiazole or 2-benzothiazolethiol	[149-30-4]
MMBT	2-Methylmercaptobenzothiazole or 2-(methylthio)benzothiazole	[615-22-5]
DBBT	2,2′-Dithiobisbenzothiazole	[120-78-5]
TBBT	2,2′-Thiobisbenzothiazole	[4074-77-5]
BBTS	*N-tert.*-Butyl-2-benzothiazolesulfenamide	[95-31-8]
CBTS	*N*-Cyclohexyl-2-benzothiazolesulfenamide	[95-33-0]

Judging from the number of citations in *Chemical Abstracts*, MBT is by far the most widely used compound of the seven. The sulphide TBBT and the two sulfenamides BBTS and CBTS had no pertinent references and are not discussed further.

Although the literature on benzothiazoles is extensive, very little pertains to their occurrence, effects and fate in the environment. This review attempts to gather what is known about these compounds in aquatic systems and to point out where further work is needed to model their behaviour as environmental contaminants.

Benzothiazoles have many uses, e.g., as vulcanization accelerators in the rubber industry, as biocides, as photosensitizers in photography, and as corrosion inhibitors. Possible mechanisms of entry into the environment are wastewater and air emissions from their manufacture; cooling water in which they have been used as corrosion inhibitors; applications as biocides; and from rubber tires and other rubber goods. Some rubber contains as much as 2% MBT (Guess and O'Leary, 1969).

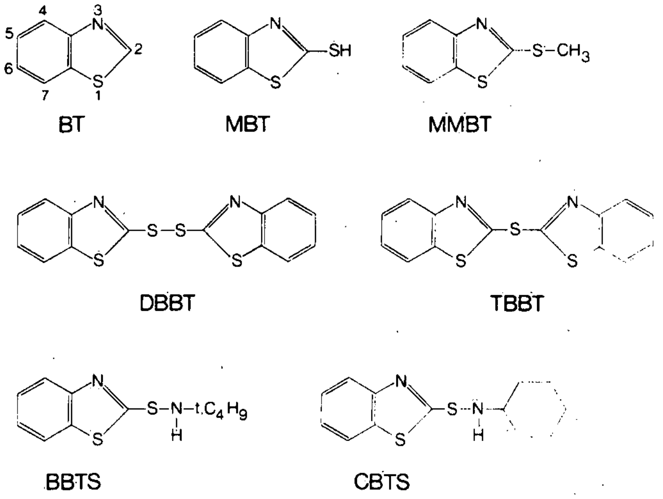

Figure 1. Structural formulae and abbreviations for the benzothiazoles reviewed. Ring numbering is shown for benzothiazole (BT).

PHYSICAL AND CHEMICAL PROPERTIES

The physical properties are listed in Table 1. Water solubility and partition coefficient are of prime importance in determining behaviour and fate in the aquatic environment. These data, however, are incomplete for these compounds.

The 2-position of benzothiazole is quite reactive. Benzothiazole adds to substituted phenyl isocyanates to give *N*-arylbenzothiazole-2-carboxamides (Papadopoulos and Schupbach, 1979), as shown in Figure 2a. The 2-acylbenzothiazoles undergo homolytic substitution by 1-adamantyl (Fiorentino *et al.*, 1977a,b; Fig. 2b). Benzothiazole-2-sulfonic acid gives 2-hydroxybenzothiazole on acid hydrolysis (Mainprize *et al.*, 1976; Fig. 2c). This may be important in determining the products of biological and abiotic degradation.

Table 1. Physical Data for Benzothiazoles

	BT	MBT	MMBT	DBBT
Boiling point (°C)	227-228* 231†	NA	NA	NA
Melting point (°C)	NA	180.2-181.7* 179†	52†	180* 180†
Water solubility	Slight*	Moderate*	100 ppm‡	Insoluble
Partition coefficient (log P)	2.04§ 2.03§	1.61§ 2.20** 0.90‡‡	2.00** 3.0††	–

NA – Not applicable.
* *Merck Index* (1968).
† Chemical Rubber Company (1967).
‡ R.F. Platford, unpub. results.
§ Hansch and Leo (1979); octanol-water.
** Hansch and Leo (1979); chloroform-water.
†† R.F. Platford, unpub. results; octanol-water.
‡‡ Hansch and Leo (1979); methyl isobutyl ketone-water.

MBT can exist in two tautomeric forms, which are the thiol or thione form (Fig. 2d). The thione form is favoured in the solid state for several metal complexes of MBT (Yoshida *et al.*, 1979). Bonding is due to the exocyclic sulphur and probably the heterocyclic nitrogen. Coordination is through the heterocyclic nitrogen in the platinum, palladium, osmium, and gold complexes of MBT and MMBT (Doadrio *et al.*, 1978). Benzothiazoles and 2-thiobenzothiazoles appear to be very good ligands for several metals, and this may affect their behaviour in aquatic systems, such as partitioning between the liquid and solid phase.

The high resolution mass spectra (EI) of some substituted benzothiazoles have been reported (Millard and Temple, 1968). Benzothiazole itself loses CS and HCN, with the C-2 H becoming the H of HCN. The 2-substituted benzothiazoles (no C-2 H) fragment by different pathways, including HCN loss by a mechanism different from the one above. Abbreviated fragmentation patterns for BT, MBT and MMBT are given in Figure 3.

The authors have found that 2-thiobenzothiazoles give a strong response in negative ion mass spectrometry and on the electron capture detector in gas chromatography. The possibility of developing very sensitive and selective methods of analysis therefore exists, should this be necessary.

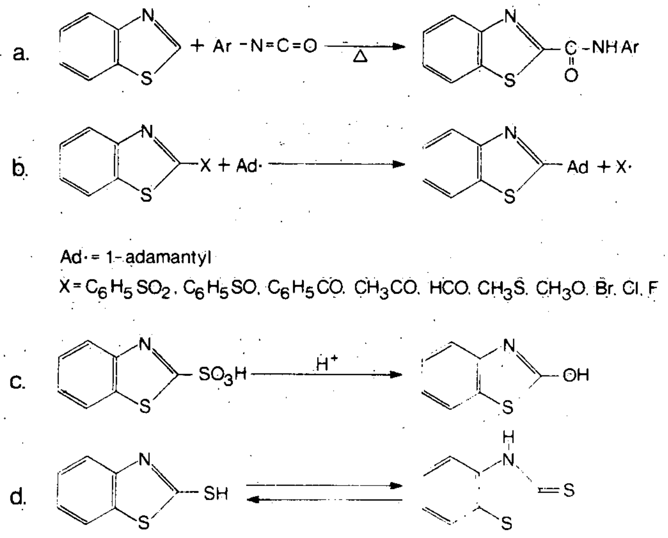

Figure 2. Chemical properties of benzothiazoles.

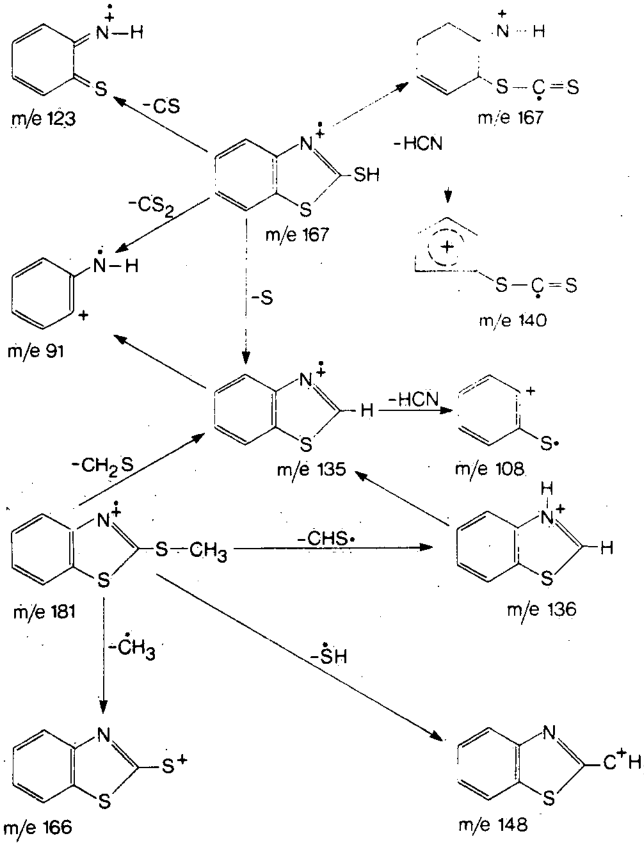

Figure 3. Mass spectral fragmentation patterns for BT, MBT and MMBT.

ANALYSIS AND OCCURRENCE

Three general approaches have been used for extraction and pre-concentration of organic compounds from environmental samples. These are: solvent extraction, adsorption on macroreticular resins with elution by an organic solvent such as ether, and gas stripping. The first two have been used for samples containing benzothiazoles; the latter method would only be suitable for BT and MMBT, which have sufficient vapour pressure to be extractable. Benzothiazoles are much more soluble in water than many contaminants (e.g., polychlorinated biphenyls). Consequently, their extraction from water is somewhat more difficult, and relatively large volumes of polar solvents are required when using solvent extraction. The octanol-water partition coefficients of BT and MMBT are about 100 and 1000, respectively (Table 1).

Samples were analyzed by gas chromatography and gas chromatography-mass spectrometry. In most cases, the benzothiazoles were just one (or more) of many compounds analyzed in the samples. In Table 2, the existing data on the occurrence of benzothiazoles in water are summarized. They have been found in wastewater (20-60 ppb for BT), river water (1-2 ppb for BT), and drinking water (no concentrations given). The BT from one tire plant wastewater persisted several miles downstream from the plant in the receiving water (Jungclaus *et al.*, 1976). In the one case in which sediments were also analyzed, no benzothiazoles were detected.

Table 2. Reported Occurrences of Benzothiazoles in the Aquatic Environment

Source	Benzothiazole identified
Drinking water Cincinnati, OH*	BT, MMBT, 2-HOBT
River water†	BT (2 ppb)
Tire plant wastewater ‡	BT (20 ppb)
Tire plant wastewater ‡	BT (60 ppb), MBT (30 ppb)
River Waal, Netherlands §	BT (1 ppb)
Delaware River, Philadelphia, PA**	BT (major contaminant), MMBT, MeBT

* Coleman *et al.* (1980); 2-HOBT = 2-hydroxybenzothiazole.
† Jungclaus *et al.* (1978); location not specified.
‡ Jungclaus *et al.* (1976); plants not identified.
§ Meijers and Van der Leer (1976).
** Burnham *et al.* (1973); MeBT = methylbenzothiazole, isomer not determined.

Two more references should be mentioned. Since they are in the Japanese literature, the information is from the abstracts only. One group analyzed for MBT in fish (carp) and aquarium water by extracting with methyl isobutyl ketone and analysis by high-speed liquid chromatography (Ishiwata *et al.*, 1978). Detection limits were 1 and 0.01 ppm in fish and water, respectively. The second group analyzed for MBT in river water and sediment by extraction with methylene chloride and analysis by gas chromatography using a flame photometric detector (Shinohara *et al.*, 1978). Detection limits were 0.04 and 2 ppb in water and sediment, respectively.

BIOLOGICAL PROPERTIES

Benzothiazoles display a wide spectrum of biological activity. They are used as fungicides, with MBT being effective against some fungi at 5-10 ppm (Owens, 1969). Many derivatives of benzothiazole, including the simple ones such as MBT and 2-(ethylthio)benzothiazole, have moderate antiviral activity against some viruses *in vitro*, e.g., vaccinia, Western equine encephalitis, and Newcastle disease virus (Rada *et al.*, 1979). The 2-aminobenzothiazole was effective against influenza virus *in vivo* (Akerfeldt, 1970). Some chloroplatinic complexes of substituted benzothiazoles possess moderate anti-tumor activity (Doadrio *et al.*, 1978). MBT induces synthesis of four proteins in cultured chick embryo cells (Levinson *et al.*, 1979). A biological property of potential importance to sewage treatment facilities and receiving waters is the ability of benzothiazoles to inhibit nitrification (Tomlinson *et al.*, 1966). MBT and DBBT gave a 75% inhibition at 3 and 38 ppm, respectively. No toxicity data were found for aquatic organisms such as zooplankton or fish.

Very little is known about the biodegradability of benzothiazoles. Testing under static conditions showed that 2-hydroxybenzothiazole is readily degradable; MBT and MMBT are degraded more slowly (Chudoba *et al.*, 1977). Activated sludge acclimatized to benzothiazole-2-sulfonic acid degraded 2-hydroxybenzothiazole, BT and MBT (Mainprize *et al.*, 1976). Previous exposure of the microorganisms to benzothiazole compounds appears to be required for them to degrade these compounds. Therefore, the degrading enzymes are inducible.

The metabolism of MBT in carp has been studied using ^{14}C-labelled material (Hashimoto *et al.*, 1978). The MBT that is assimilated from the intestine appears in the bile as

MBT-glucuronide. Ultimately, nearly 100% of the administered dose was found in the water due to excretion, with 0.3% in the bile and tissue.

MAMMALIAN TOXICITY AND HUMAN HEALTH

Few data are available on the toxicity of these compounds to animals, but their toxicity appears to be low. Toxicity of MBT to cell cultures is low, and single dose LD_{50} to mice is 440 mg/kg (intraperitoneal) and 2000 mg/kg (oral) (Guess and O'Leary, 1969). Sub-chronic exposure produced liver damage and prolonged sleeping time in mice (Guess and O'Leary, 1969). Single dose LD_{50} for DBBT was 12 000 mg/kg for rats and mice (Vernot *et al.*, 1977). Uptake and excretion of ^{14}C-MBT have been reported for the guinea pig (Nagamatsu *et al.*, 1979). The main excretion route was the urine with 92% excretion 6 h after dosing. The urinary metabolites were assumed to be the glucuronide and sulphate conjugates.

The Russian literature contains many references to occupational exposure of workers to MBT and DBBT. From the abstracts it is clear only that these compounds are of some concern; the exact nature of the problem is not clear.

MBT is a contact allergen (Baer *et al.*, 1973) and is one of the compounds responsible for the allergy to rubber products exhibited by some people.

SUMMARY

Although little is known about the environmental aspects of industrially important benzothiazoles, they are found in the aquatic environment, are moderately persistent, possess biological activity, and may have other effects such as metal chelation. On balance, they do not appear to pose an imminent threat to the aquatic environment. Where found, their levels are below that required for strong biological activity. They may cause aesthetic problems by imparting taste and/or odour to water (Verschueren, 1977). In Canagagigue Creek, their presence provides an excellent scientific opportunity to study the behaviour of volatile, slightly persistent, moderately water soluble organic compounds in a natural aquatic ecosystem.

To broaden our knowledge of this class of compounds and provide a basis for modelling their behaviour in aquatic ecosystems, the following research is needed:

1. Some refinement of analytic methods is required for water samples due to the relatively high solubility of these compounds;
2. Cleanup procedures are needed so the analysis can be extended to plant and animal tissue for bioaccumulation measurements;
3. Physical data are required to calculate rates of volatilization;
4. Biodegradation studies can be done in the laboratory to determine potential degradation products;
5. Biodegradation studies in the field are needed to provide estimates of the actual rates of degradation and persistence of degradation products; and
6. Some simple bioassay procedures should be considered to investigate potential biological effects in the ecosystem.

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