Ecotoxicology and Environmental Safety ■ (■■■■) ■■■–■■■

Contents lists available at ScienceDirect

Ecotoxicology and Environmental Safety

journal homepage: www.elsevier.com/locate/ecoenv

Biotransformation of tetrabromobisphenol A (TBBPA) in anaerobic digester sludge, soils, and freshwater sediments

Drew C. McAvoy [a,*], Charles A. Pittinger [b], Alison M. Willis [c]

[a] University of Cincinnati, Department of Biomedical, Chemical, and Environmental Engineering, 40 Engineering Research Center, Cincinnati, OH 45221, USA
[b] LLC, 6547 Edwood Avenue, Cincinnati, OH 45224, USA
[c] Toxicology Excellence for Risk Assessment, 2300 Montana Avenue Suite 409, Cincinnati, OH 45211, USA

ARTICLE INFO

Article history:
Received 9 March 2015
Received in revised form
6 July 2015
Accepted 7 July 2015

Keywords:
Tetrabromobisphenol A (TBBPA)
Biotransformation
Disappearance time (DT_{50})
Aerobic
Anaerobic

ABSTRACT

The biotransformation of tetrabromobisphenol A (TBBPA) was evaluated in anaerobic digester sludge, soils, and freshwater sediments. In anaerobic digester sludge, TBBPA biotransformed rapidly with a 50% disappearance time (DT_{50}) of 19 days, though little mineralization (1.1%) was observed. In aerobic soils, mineralization of TBBPA ranged from 17.5% to 21.6% with 55.3–83.6% of the TBBPA incorporated into the soils as a non-extractable bound residue. The DT_{50} for TBBPA in aerobic soils ranged from 5.3 to 7.7 days. In anaerobic soils, 48.3–100% of the TBBPA was incorporated into the soils as non-extractable bound residue with $<4\%$ mineralized. The soil fate studies demonstrated extensive incorporation of TBBPA into the solid matrix and this association was related to the amount of organic carbon in the soils (i.e., greater association of TBBPA with soil at higher organic carbon content). In anaerobic sediments the DT_{50} for TBBPA ranged from 28 to 42 days, whereas in aerobic sediments the DT_{50} for TBBPA ranged from 48 to 84 days and depended on the initial dose concentration. Most of the TBBPA in the sediment studies was incorporated as a non-extractable bound residue with little mineralization observed. Sediment extracts revealed three unknown biotransformation products and bisphenol A (BPA). These results were consistent with previously published studies where TBBPA biotransformed in anaerobic environments (digester sludge and sediments) by debromination and slowly mineralized in the test environments (anaerobic digester sludge, soils, and freshwater sediments).

1. Introduction

Tetrabromobisphenol A (TBBPA) is a widely used brominated flame retardant that has been shown to undergo debromination in the environment by bacterial and photolytic pathways (Arbeli and Ronen, 2003; Eriksson et al., 2004). TBBPA is used during the production of plastic polymers and electronic circuit boards, as well as being incorporated into consumer electronics, office and communication equipment, automotive, aviation, and entertainment equipment (BSEF, 2012). TBBPA is a solid at room temperature with the following properties: low vapor pressure $<1.19\times10^{-5}$ Pa at 20 °C (ACCBFRIP, 2001 as cited in Environment Canada, 2013); moderately high octanol/water partitioning (log Kow=4.5–6.5) that is dependent on the ionization state and matrix pH (Kuramochi et al., 2008); and low to moderate solubility that increases with pH, e.g., 0.17-4.16 mg L^{-1} at neutral pH and 27.9 mg L^{-1} at pH 9.5 (Kuramochi et al., 2008).

TBBPA can enter the environment from municipal wastewater treatment plants via biosolids amendment to soil and effluent discharge to receiving waters. The fate and exposure of TBBPA when introduced into these environments will depend on its physicochemical properties and composition of the solids it encounters. In particular, the formation of bound residues in a soil or sediment environment can significantly affect the bioavailability and biodegradation of an organic compound (Hatzinger and Alexander, 1995). The process of forming bound residues is thought to involve either physical entrapment of an organic compound into small pores within the solid matrix (referred to as sequestration) thus inhibiting free movement of the compound, or the incorporation of the organic compound within the solid matrix organic matter (referred to as aging).

Most definitions of bound residues depend on the extraction procedure being used. For example, labile or bioavailable fractions have been defined as those extractable by aqueous or weak organic solvent extraction methods, whereas non-extractable residues (NER) have been defined as those extractable under harsher

* Corresponding author.
E-mail addresses: capittinger@gmail.com (C.A. Pittinger), Willis@tera.org (A.M. Willis).

http://dx.doi.org/10.1016/j.ecoenv.2015.07.009
0147-6513/

conditions that include an organic solvent under pressure and/or increased temperature, e.g., using reflux, microwave, or accelerated solvent extraction (ECCTOC, 2010). The NER fraction is considered to be strongly associated with the solid matrix and thus unlikely to be available. ECETOC (2010) also defined bound residues as those tightly associated with the solid matrix, often forming covalent bonds. Such residues are indistinguishable from natural organic matter and not available to organisms. The ECETOC terminology for extractable, non-extractable, and bound residues will be use to describe the results of this study.

The fate of TBBPA in soil and sediment environments is also controlled by the ability of microbial populations to degrade and utilize it as a carbon and energy source. Several studies have shown that TBBPA can microbially degrade and mineralize in sequential anaerobic and aerobic environments (Ronen and Abeliovich, 2000; Voordeckers et al., 2002; Arbeli and Ronen, 2003; Ravit et al., 2005). These researchers have demonstrated that TBBPA is reductively debromonated to bisphenol A (BPA) during the anaerobic phase and then BPA is mineralized during the aerobic phase. Arbeli and Ronen (2003) also identified monobromobisphenol A, dibromobisphenol A, and tribromobisphenol A as biotransformation products in their anaerobic laboratory experiments. These studies illustrate that understanding the ability of microbial populations to metabolize TBBPA is an important aspect in evaluating its fate and exposure.

The purpose of this paper is to summarize results from six laboratory studies conducted by three TBBPA producers (Albemarle, Chemtura, and ICL) over a period from 1989 to 2006. The protocols used in the 1980s were state-of-the-art at that time, while the studies conducted in the 2000s were based on OECD guidelines (OECD, 2002a, 2002b). Included are biotransformation and mineralization results of TBBPA in anaerobic digester sludge, soils, and freshwater sediments. The original study reports can be obtained from the lead author by request.

2. Materials and methods

The studies described within were either conducted by Wildlife International or Springborn Life Sciences using Good Laboratory Practice (GLP) procedures.

2.1. Mineralization and transformation in anaerobic digester sludge (Wildlife International, 2006a)

A 120 day study was conducted to assess the rate and extent of primary biotransformation and mineralization of uniform ring-labeled ^{14}C-TBBPA in anaerobic digester sludge (Wildlife International, 2006a). The protocol was based on the OECD 308 Guideline: Aerobic and Anaerobic Transformation in Aquatic Sediment Systems. Anaerobic digester sludge was collected from a local wastewater treatment plant (Federalsburg, Maryland) and sieved through a 6.3 mm sieve prior to use. The test chambers consisted of 500 mL glass media bottles with rubber stoppers. Each test chamber contained 100 mL of either live or sterile digester sludge plus 100 mL of a mineral salt solution. The live inoculum contained 23,353 mg total-solids L^{-1} (2.3% TS) and the sterile inoculum contained 24,660 mg total-solids L^{-1} (2.5% TS). The pH of the anaerobic digester sludge was adjusted to 7.1 prior to use and all test systems were incubated under anaerobic conditions in the dark at approximately 35 °C.

A parallel set of dose systems was established to evaluate mineralization and biotransformation of TBBPA. The test chambers were dosed at a concentration of 50 μg ^{14}C-TBBPA (kg dry-sludge)$^{-1}$ with ethanol as the carrier solvent. The radiolabeled TBBPA had a specific activity of 56 mCi $mmol^{-1}$, radiochemical purity of 99.6%, and formula weight of 545.7 mg $mmol^{-1}$. The radiolabeled test substance was obtained from Amersham Biosciences, UK. The concentration of the ^{14}C-TBBPA dosing solution (0.445 μg μL^{-1}) was verified by liquid scintillation counting (LSC).

In the mineralization experiment, both live (biotic) and sterile (abiotic) anaerobic sludge test chambers were continuously purged with nitrogen gas to maintain anaerobic conditions. The resulting off-gas was monitored for the production of $^{14}CO_2$, $^{14}CH_4$, and other radiolabeled volatile compounds. The mineralization apparatus employed an alkali solution (1.5 N KOH) to trap $^{14}CO_2$ and a combustion furnace to oxidize $^{14}CH_4$ and the radiolabeled volatile compounds to $^{14}CO_2$ prior to being trapped in an alkali solution. The mineralization traps were sampled on days 7, 14, 28, 42, 56, 70, 84, 98, 112, and 120. The ^{14}C in the alkali traps was analyzed for radioactivity by LSC.

A secondary set of chambers was used to evaluate the biotransformation of ^{14}C-TBBPA. In this experiment, both live (biotic) and sterile (abiotic) anaerobic sludge test chambers were continuously purged with nitrogen gas and the off-gas was connected to a gas trap (i.e., water) to prevent air from entering the chambers. The transformation test chambers were sampled on days 0, 7, 14, 28, 42, 56, and 120. On each sampling day the content of the test vessels were transferred to 200 mL centrifuge bottles and centrifuged at 3000 rpm for 10 min. The water layers were decanted and analyzed for total radioactivity by LSC. The original test vessels were rinsed with 75 mL of acetonitrile (ACN) and the rinses added to the solids in the centrifuge bottles. The samples were sonicated for 5 minutes using a Branson Digital Sonifier and then centrifuged at 3000 rpm for 5 min. The ACN extracts were decanted into 1 L round bottom flasks and another 75 mL of ACN was added to the remaining solids. The extraction procedure was repeated two more times and the extracts analyzed for total radioactivity by LSC. A portion of the ACN extracts were further characterized using an Agilent 1100 Series High Performance Liquid Chromatograph (HPLC) equipped with a Variable Wavelength Detector (VWD) and IN/US β-RAM radioactivity detection (RAD). In addition, the water layer samples collected on day 120 were analyzed using Thin Layer Chromatography (TLC) with RAD.

2.2. Aerobic transformation in soil (Wildlife International, 2005)

This 6 month study was conducted to assess the mineralization and biotransformation of uniform ring-labeled ^{14}C-TBBPA in aerobic soil systems (Wildlife International, 2005). The test system consisted of 500 mL glass media bottles with rubber stoppers. Each test chamber was filled with 100 g dry weight of soil. The test chambers were dosed at a concentration of 50 μg ^{14}C-TBBPA (kg dry-soil)$^{-1}$ with ethanol as the carrier solvent. The ^{14}C-TBBPA had a specific activity of 56 mCi $mmol^{-1}$, radiochemical purity of 99.6%, and formula weight of 545.7 mg $mmol^{-1}$. The radiolabeled test substance was received from Amersham Biosciences, UK. The ^{14}C-TBBPA dosing solution (0.097 μg μL^{-1}) was verified using LSC. The off-gas from the test chambers passed through a charcoal sorbent trap for collecting volatile compounds and an alkali solution (1.5 N KOH) trap for trapping $^{14}CO_2$.

Four soil types (loamy sand, sandy clay loam, silt loam, and silty clay loam) were utilized in this investigation. The organic carbon content of the soils was 1.2% for the loamy sand, 2.1% for the sandy clay loam, 2.2% for the silty clay loam, and 5.5% for the silt loam. The pH of the soils was 7.0 for the loamy sand, 6.8 for the sandy clay loam, 5.7 for the silty clay loam, and 7.5 for the silt loam. Microbial biomass was measured using the fumigation-extraction method and it was 110 μg g^{-1} for the loamy sand, 254 μg g^{-1} for the sandy clay loam, 131 μg g^{-1} for the silty clay loam, and 696 μg g^{-1} for the silt loam.

The test systems were initially acclimated under aerobic

Please cite this article as: McAvoy, D.C., et al., Biotransformation of tetrabromobisphenol A (TBBPA) in anaerobic digester sludge, soils, and freshwater sediments. Ecotoxicol. Environ. Saf. (2015), http://dx.doi.org/10.1016/j.ecoenv.2015.07.009