BIOLOGICAL HALF-LIVES OF POLYBROMINATED DIPHENYL ETHERS AND TETRABROMOBISPHENOL A IN EXPOSED WORKERS

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Introduction

Tetrabromobisphenol A (TBBPA) and polybrominated diphenyl ethers (PBDEs) are major brominated flame retardants with a predicted annual world wide production of 60.000 and 40.000 tonnes, respectively[1,2]. PBDEs have been reported as contaminants in both wildlife and humans[3-5] while TBBPA has been identified in sediments[6]. Recently, low but increasing ppb levels of tetra-, penta- and hexaBDEs were found in human milk[5]. Another study reported human blood concentrations of tetra- to decaBDEs among hospital cleaners, clerks and personnel dismantling electronics for recycling[4]. Ambient air analyses at the electronics dismantling plant showed that a heptaBDE, 2,2',3,4,4',5',6-heptabromodiphenyl ether (BDE-183), decabromodiphenyl ether (BDE-209) and TBBPA were major pollutants[7]. Significantly increased levels of BDE-183 and BDE-209 were detected in serum from the dismantling personnel[4]. The clearance rate of PBDEs and TBBPA in humans is of a great interest considering evaluation of both health risks and hygienic intervention strategies in work places. The kinetics in humans for these compounds are, however, not known.

The general objectives of the present study were to assess whether TBBPA could be detected in serum from workers at an electronics dismantling plant and to model the kinetics of TBBPA and different PBDE congeners in the workers.

Material and methods

Four workers at an electronics dismantling plant donated blood samples just before and at multiple times during their summer vacation in 1998. Three of the subjects had also donated blood samples prior to and at the end of the summer vacation in 1997[4]. Serum was centrifuged and transferred to acetone-washed glass bottles and deep-frozen until chemical analysis.

All solvents and chemicals used in the analysis were of the highest commercial grade available. Isopropanol and methyl tert-butyl ether (MTBE) were glass-distilled prior to use. The method used for extraction of serum samples have been described in detail elsewhere[8]. Briefly, internal surrogate standards 2,2',3,4,4',5'-hexabromodiphenyl ether (BDE-138), 3,3',5'-tribromo-5-chlorobisphenol A (TrBCBPA) and 2,3,3',4,4',5,5'-heptachlorobiphenyl (CB-189) were added to the serum samples (5 g). The samples were extracted twice by hexane/MTBE (1:1) (6 ml and 3 ml), after denaturation of proteins employing hydrochloric acid (1 ml; 6 M) and isopropanol (6 ml), respectively. The extracts were washed and their lipid content were determined gravimetrically. A neutral and acidic fraction were obtained by partitioning with potassium hydroxide (0.5 M in 50% ethanol). Clean-up of the neutral fraction containing the

PBDEs and PCBs were made on a silica/sulfuric acid column (silica/H_2SO_4 2:1 by weight; 1 g) with hexane (8 ml) as the mobile phase. The acid fraction containing TBBPA, was treated with diazomethane to derivatize phenolic compounds into their corresponding methyl ethers. Clean-up was subsequently made on a silica/sulfuric acid column (silica/H_2SO_4 2:1 by weight; 0.5 g). Two fractions were collected from this latter column, the first in hexane/dichloromethane (DCM) (1:1 by volume; 5 ml) and the second eluted with DCM (15 ml). The first fraction contained hydroxylated PCBs and the second TBBPA dimethyl ether. PBDEs and TBBPA were subsequently analyzed by gas chromatography/mass spectrometry (GC/MS) in the electron capture negative ionization (ECNI) mode employing a Finnigan TSQ 700 instrument (TermoQuest, Bremen, Germany) connected to a Varian 3400 GC. Selective ion monitoring (SIM) was performed on the prevalent ion formed, representing the bromine isotopes (*m/z* 79 and 81). Split-less injection were performed for analysis of TBBPA and PBDE congeners with less than five bromine substituents and on-column injection employing an septum equipped programmable injector (SPI) for analysis of PBDE congeners with six or more bromine substituents. The chromatographic separations were performed on a DB-5 (30 m x 0.25 mm i.d., 25 μm film thickness, J&W Scientific, USA) and DB-5HT (15 m x 0.25 mm i.d., 0.10 μm film thickness) capillary columns, for the split-less and on-column analyses, respectively. A signal to noice (S/N) ratio greater than ten was used to define the limit of quantification (LOQ), when no interferences were present in the blank samples analyzed (n=6). If interferences were present in the blanks, the amount of the analyte in the sample had to be at least five times the average blank level to be accepted. Thus, when interferences were present, the LOQ is directly related to the blank sample level.

Identification of the analytes was made by comparison of the relative retention times (RRT) with that of authentic reference substances. The identification was further supported by standard addition to one of the samples to approximately twice the original concentration. This sample was then analyzed by GC/MS(ECNI) and the chromatographic peaks were studied for any deformities, which would indicate an inaccurate identification.

The estimates of the biological half-lives were mainly based on the data presented in this study. However, for the PBDEs data from a control population of 20 non-occupationally exposed female cleaners were also utilized[4]. For the kinetic calculations NONMEM version V, level 1.1 was employed[9]. Approximate 95% confidence intervals (CI) were calculated.

Results and Discussion

TBBPA was detected in serum from all four workers sampled before summer vacation (Table 1). The serum concentrations of TBBPA (2-7 pmol/g lipid) were about half of those of BDE-183, but very similar to those of BDE-209. The serum concentrations of TBBPA before, and at multiple times during the summer vacation in 1998 are given in Table 1. The estimated half-live of TBBPA was 2.2 days (95% CI 1.4-2.9). Serum concentrations of BDE-183 and BDE-209 before and during the summer vacations in 1997 and 1998 are given in Table 1. The estimated half-live of BDE-183 was 86 days (95% CI 43-128) and of BDE-209 6.8 days (95% CI 3-16).

The air concentration of TBBPA was high at the dismantling plant, 13-110 pmol/m^3 as compared with 0.018-0.13 pmol/m^3 in ordinary offices[10]. It is thus not surprising that TBBPA is present in serum from the electronics dismantling workers. The rapid decrease in concentration of this phenolic substance during the exposure free period further supports that the exposure of

TBBPA to these workers is mainly work-related.

The observed half-life of approximately 2 days of TBBPA in humans is in agreement with experimental data obtained from rats[11,12]. The fast turnover of TBBPA is most likely due to that TBBPA, as a phenolic substance, may form glucuronide and sulfate ester conjugates without any prior biotransformation. Consequently, it is unlikely that TBBPA forms any reactive intermediates prior to conjugation.

Vacation day (Year)	Serum concentration (pmol/g lipid weight)		
	BDE-183	BDE-209	TBBPA
Subject A			
0 (1997) [a]	12	7.2	[b]
0 (1998)	8.7	2.9	7.4
3 (1998)	7.1	2.3	5.0
10 (1998)	7.8	1.7	<2 [c]
14 (1998)	7.9	1.8	<2 [c]
21 (1998)	6.7	2.1	<2 [c]
28 (1998)	7.7	1.3	<2 [c]
Subject B			
0 (1997) [a]	20	6.3	[b]
28 (1997) [a]	14	<0.7	[b]
0 (1998)	18	7.2	5.6
4 (1998)	13	4.3	3.2
10 (1998)	16	2.6	<2 [c]
17 (1998)	14	3.2	<2 [c]
25 (1998)	14	2.3	3.0
Subject C			
0 (1997) [a]	16	5.0	[b]
35 (1997)	12	11 [d]	[b]
0 (1998)	12	6.6	2.0
4 (1998)	11	3.1	3.1
10 (1998)	8.9	2.2	<2 [c]
17 (1998)	9.2	1.6	<2 [c]
24 (1998)	10	2.1	<2 [c]
28 (1998)	8.4	1.7	<2 [c]
35 (1998)	8.8	1.3	<2 [c]
Subject D			
0 (1997) [e]			
3 (1998)	6.6	1.2	2.1
6 (1998)	6.6	1.5	<2 [c]
17 (1998)	6.8	0.74	<2 [c]
26 (1998)	6.1	1.1	<2 [c]
31 (1998)	7.4	0.93	<2 [c]

Table 1. *Serum concentrations of 2,2',3,4,4',5',6-heptaBDE (BDE-183), decabromo-diphenyl ether (BDE-209) and tetrabromobisphenol A (TBBPA) in four electronics dismantling workers, before and during summer vacations in 1997 and 1998.*

[a] Data from[4]; [b] Not determined; [c] Concentration below LOQ (limit of quantification), defined as five times average blank sample (n=6) level; [d] Excluded in the half-life calculations; [e] No serum sample available for this subject.

Estimated half-lives of BDE-183 and BDE-209 in humans are also reported for the first time. In a previous study we observed that PBDE congeners with a high bromine content, in particular BDE-209, seem to have shorter half-lives in humans than lower brominated diphenyl ethers[4]. These observations are in line with experimental data, indicating BDE-209 to be rapidly cleared by rats[13,14]. In contrast tetra- and pentaBDEs (i.e., BDE-47 and BDE-99) were found to be slowly excreted[15,16]. The presently observed half-lives of approximately one week for BDE-209 is significantly shorter than that of BDE-183, with a half-life of 86 days. Considering the lipophilic characteristics of BDE-209 [log K_{ow} = 9.97][17] and the observed short half-life being more than ten times shorter than that of BDE-183, it seems unlikely that BDE-209 should be excreted in an unmetabolized form. Metabolites of BDE-209 were in fact indicated in the bile from rats dosed with ^{14}C-labeled BDE-209[14] but the identities of any BDE-209 metabolites have not yet been reported. The short half-life of BDE-209 must be followed up by a detailed metabolism study to determine the transformation products formed and to assess the toxicity of this compound in relation to its reactivity. More data are also requested for TBBPA since it is still of interest to determine if this compound may act as an endocrine disrupter through interactions with protein transport proteins or receptors.

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