IMPORTANCE OF DUST AND DIET FOR THE HUMAN EXPOSURE TO PBDEs AND HBCDs

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Abstract
Food, dust and serum of 19 Belgian volunteers were analysed for the presence of PBDEs and HBCDs. Duplicate diet samples contained tri-hepta BDEs and BDE 209 between < 1 - 128 and < 20 – 7750 pg/g wet weight (ww), respectively. ΣHBCDs were present in diet at lower concentrations < 1 – 35 pg/g ww. PBDE concentrations in dust ranged between 5 – 70 and 19 – 588 ng/g dry weight (dw) for tri-hepta congeners and BDE 209, respectively. ΣHBCDs in dust ranged between 33 – 758 ng/g dw. Serum samples contained tri-hepta BDEs between < 1 – 7 ng/g lipid weight (lw) and ΣHBCDs between < 0.5 – 11 ng/g lw. Overall levels are consistent with those of a background exposed European population. For PBDEs, no correlations were found between concentrations in dust and/or diet and those in the corresponding serum samples. In contrast, dust intake and serum concentrations for ΣHBCDs were significantly correlated ($r = 0.86$, $p < 0.01$), but no correlation was evident between ΣHBCDs in serum and dietary intake. Despite the absence of correlation between dietary intake and serum concentrations, estimation of exposure in this study was consistent with the diet making an important contribution to overall intake of both PBDEs and HBCDs. The lack of correlation is hypothesised to reflect the fact that our estimates of dietary exposure covered an insufficiently long period (1 week) to represent accurately long-term exposure via this pathway.

Introduction
Polybrominated diphenyl ethers (PBDEs) have been used extensively in Europe and the US as brominated flame retardants (BFRs) in a variety of products, such as textiles, furniture upholstery and electronic equipment. Restrictions on their manufacture and use in both Europe and the US have led to the rise of various substitutes, including hexabromocyclododecanes (HBCDs). Both compounds have already been detected in dust, air and food[1-3], the principal sources of human exposure to BFRs. As a direct consequence, these BFRs have been measured in human tissues such as liver, fat and serum[4,5]. Although toxic effects have been proven for both compounds[6,7], no study has investigated to date the combined contribution of food and dust ingestion and their importance for the human serum levels. The aim of this study was to investigate the contribution of food and dust to the total PBDE and HBCD intake in a Belgian group of students and to relate this exposure to concentrations in corresponding serum samples.

Materials and methods
Nineteen student volunteers aged between 20-25 years participated in the study. Duplicate diet samples were collected on a daily basis during one week, dust samples were taken according to a standardised procedure [1] and serum samples were collected at the end of the study period. Food and dust samples were Soxhlet extracted (2 h) with hexane:acetone (3:1) and cleaned-up on acidified silica[3]. Serum samples underwent solid phase extraction (Oasis HLB) and clean-up with acid silica. Extracts were analysed at the University of Antwerp for PBDEs on GC-ECNI/MS using a 15 m x 0.25 mm x 0.10 µm DB-5 column. PBDEs were confirmed in dust on GC-EI/MS using a 25 m x 0.22 mm x 0.25 µm HT-8 column. Food samples where a peak corresponding to the retention time of HBCD was observed in the GC-ECNI/MS chromatograms and all dust samples were further analysed at the University of Birmingham, Here, food and dust were extracted using accelerated solvent extraction (ASE)

and cleaned up with acidified silica. Food, dust and serum extracts were analysed for HBCDs by LC/MS-MS using RP-C18 (for HBCD diastereomers) and Nucleodex β-PM (for HBCD enantiomers) columns[8].

Quality control was achieved by regular analysis of procedural blanks, certified materials and through regular participation in interlaboratory comparison exercises for matrices, such as serum, dust and biological samples. For each analyte, the mean procedural blank value was used for subtraction. After blank subtraction, the limit of quantification (LOQ) was set at 3 times the standard deviation of the procedural blank, which ensures > 99% certainty that the reported value originated from the sample. When concentrations were below LOQ, they were reported as *f**LOQ with *f* being the fraction of samples above LOQ.

Results and discussion

Food. PBDE concentrations in food were lower compared to European market basket studies, ranging between < 1 - 128 and < 20 – 7750 pg/g wet weight (ww) for tri-hepta PBDE congeners and BDE 209, respectively. Reduction of PBDE levels due to cooking and overall healthy dietary habits were most likely to cause this background exposure. HBCD levels in food were lower compared to PBDE concentrations, ranging between < 1 – 35 pg/g ww. Low detection frequencies and peak-concentrations in fish-containing meals characterised HBCD-profiles in food. The γ-HBCD isomer was the dominant HBCD isomer in most dietary samples, except those of animal origin, which were mainly composed of α-HBCD.

Dust. PBDE concentrations in dust are at the low end of those reported in other European studies, especially UK studies which are characterised by high BDE 209 concentrations, and much lower compared to US and Canadian studies that are characterised by high levels of congeners predominant in the Penta-BDE formulation. Dust concentrations of sum tri-hepta PBDEs and BDE 209 ranged between 5 – 70 ng/g dry weight (dw) and 19 – 588 ng/g dw, respectively. HBCD levels in dust were between 33 – 758 ng/g dw, also situated at the low end of reported European concentrations. Dust samples were composed mainly of α-HBCD (60%), followed by γ-HBCD (30%), in accordance with recent publications[9].

Serum. Tri-hepta PBDE congeners in serum ranged between 1 – 7 ng/g lw, which corresponds to a background exposed population, while BDE 209 could not be detected in any of the serum samples. HBCDs in serum ranged between < 0.5 – 11 ng/g lw, composed solely of α-HBCD.

Intake of PBDEs. Intake of PBDEs through food is characterised by low variation between individuals and low intake levels compared to other dietary market basket studies. Intake through food ranged between 6 – 22 and 50 – 238 ng/day for tri-hepta PBDE congeners and BDE 209, respectively. Dust ingestion was calculated based on an average dust exposure of 20 mg/day and a high dust exposure of 50 mg/day[10]. This led to dust ingestion exposure estimates of tri-hepta congeners (average: 0.1 – 1.4 and high: 0.3 – 3.5 ng/day) and BDE 209 (average: 0.4 – 11 and high: 1 – 29 ng/day) Table 1). Comparing food and dust intake revealed dietary intake as the most important exposure route for PBDEs for this small group of subjects. No correlation between either exposure route and serum concentrations could be found despite the fact that correlations between food intake and concentrations in human tissues such as serum have been detected in exposed populations[11]. This supports our hypothesis that serum concentrations of background exposed populations are more likely to be driven by occasional spikes in exposure.

Intake of HBCDs. The HBCD intake through food ranged between 1.2 – 20 ng/day which is at the low end of the concentration range reported for other European studies. Dust intake was estimated in a similar fashion to PBDEs with average intake between 1.1 – 15 ng/day and high intake between 2.8 – 38 ng/day. Comparing both exposure routes, there is a far more equal contribution from both diet and dust for HBCDs (Table 2). The importance of dust ingestion is further underlined by the existence of a correlation between dust intake and serum concentrations ($r = 0.86$; $p=0.01$), which decreased in significance when food intake was added ($r = 0.34$; $p>0.1$) (Figure 1). The greater contribution of dust exposure to body burdens might contribute to the isomeric shift from mainly γ-HBCD in food and mainly α-HBCD in dust to serum levels solely composed of α-HBCD.

Table 1. Intake (ng/day) of Σtri-hepta BDEs and BDE 209 from food and dust ingestion in adults in the present and related studies.

Intake	Country	Compounds	Median	Range	Reference
food (ng/day)	**Belgium**	**Σtri-hepta[a]**	**10.0**	**5.9 – 22.0**	*this study*
		BDE 209	**95**	**50 - 238**	
	UK	Σtri-hepta[b]	91	37 - 235	12
	USA	Σtri-hepta[c]	-	54 - 90	13
	Belgium	Σtri-hepta[c]	-	23 - 48	3
dust (ng/day)	**Belgium**	**Σtri-hepta[a]**	**0.24[d]**	**0.11 – 1.4**	*this study*
			0.59[e]	**0.27 – 3.5**	
	Belgium	**BDE 209**	**1.8[d]**	**0.4 – 11**	*this study*
			4.6[e]	**1.0 – 29**	
	UK	Σtri-hepta[a]	1.2[d]	0.2 – 7.8[f]	1
			2.9[e]	0.5 – 19[f]	
		BDE 209	56[d]	4.7 – 4 100[f]	1
			140[e]	12 – 10 000[f]	
	USA	Σtri-hepta[a]	33[d]	11 – 200[f]	1
			82[e]	27 – 510[f]	
		BDE 209	26[d]	12 – 60[f]	1
			65e	29 – 150[f]	
food + dust (ng/day)	**Belgium**	**Σtri-hepta[a]**	**10.4[d]**	**6.0 – 22.4**	*this study*
			10.8[e]	**6.3 – 23.0**	
	Belgium	**BDE 209**	**95[d]**	**61 – 240**	*this study*
			96[e]	**62 – 244**	

a- sum of BDE 28, 47, 99, 100, 153, 154 and 183; b – BDE 47, 99, 100, 153 and 154; c – BDE 28, 47, 99, 100, 153, 154 and 183; d,e - intake through dust was calculated based on an average and high dust intake of 20 (d) and 50 (e) mg/day, respectively, as suggested by Jones-Otazo et al. (*10*) and Harrad et al. (*1*); f – 5-95 percentile.

Table 2. Intake (ng/day) of ΣHBCDs from food and dust ingestion in adults in this and related studies.

Intake	Country	Median	Range	Reference
food (ng/day)	**Belgium**	**5.5**	**1.2 – 20**	*this study*
	Netherlands	-	174	14
	UK	-	354 - 474	15
	Norway	16	4 – 81	16
dust (ng/day)	**Belgium**	**2.3b**	**1.1 – 15**	*this study*
		5.7a	**2.8 – 38**	
	UK	33b	6 – 469	8
		81a	14 – 1 172	
	UK	15b	3 – 440	9
		37a	8 – 1 100	
	Canada	13b	3 – 24	
		32a	8 – 59	
	US	8b	2 – 60	
		19a	6 – 150	
food + dust (ng/day)	**Belgium**	**8**	**3.6 – 20**	*This study*
		13	**5.2 – 42**	

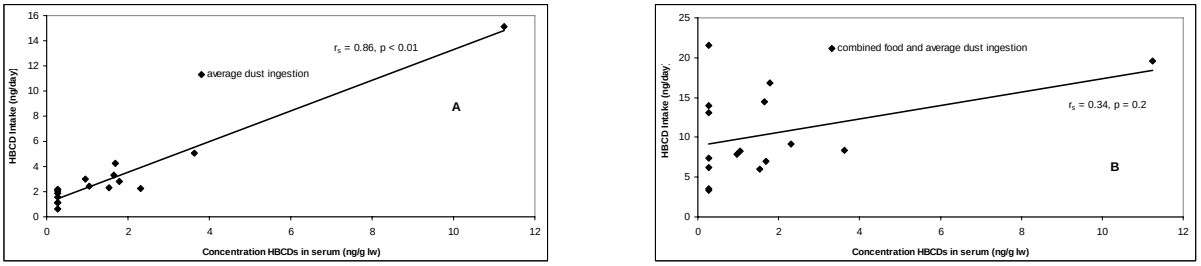
HBCD Enantiomeric patterns. The chiral signature (i.e. the relative abundance of the two enantiomers of a given isomer) of all detected isomers in food was racemic (EF = 0.5) or close to racemic in all samples above LOQ (Table 3). Since this study is the first to suggest a racemic chiral signature of HBCDs in duplicate diets, comparison with other studies is not possible. In dust samples, racemic or near-racemic chiral signatures were

also observed for all isomers (Table 3), consistent with recent observations[17]. Combined, these findings suggest that human exposure to HBCDs consists of racemic mixtures of HBCD isomers. The present study reports (-) α-HBCD as the dominating enantiomer in human serum, with an average EF of 0.28 ± 0.02 (Table 3). Similar selective enantiomeric enrichment of (-) α-HBCD was reported in human serum[18]. The combination reported here of racemic signatures in dust and diet suggests that the directionally-consistent and non-racemic signatures for α-HBCD in serum are attributable to enantioselective metabolism and/or excretion as opposed to external exposure to non-racemic matrices.

Table 3. Average ± SD enantiomeric fractions (EFs) of α-, β- and γ-HBCD in food, dust and serum. Racemic EF are 0.50.

	Food	**Dust**	**Serum**
N	12	9	9
α-HBCD	0.49 ± 0.04	0.52 ± 0.02	0.28 ± 0.02
β-HBCD	0.52 ± 0.02	0.48 ± 0.03	n.d.
γ-HBCD	0.51 ± 0.03	0.50 ± 0.02	n.d.

Figure 1. Relationship between serum concentrations of ΣHBCDs in serum and exposure to ΣHBCDs via dust ingestion (A) or combined food and average dust ingestion (B).



Conclusions. Food is an important pathway of human exposure to PBDEs and HBCDs for our small group of Belgian subjects. While dust ingestion is a comparatively minor contributor to intakes of PBDEs for our subjects, its contribution to HBCDs exposure is of a similar magnitude to that of diet. The absence of any correlation between dietary intake and concentrations in serum is hypothesised as arising from the fact that our study captured only a short (1 week) "snapshot" of dietary exposure that did not reflect accurately longer-term dietary exposure. We hypothesise further that body burdens of non-occupationally exposed populations are most likely driven by occasional spikes in dietary exposure through consumption of highly contaminated but relatively infrequently-consumed fish (e.g. eels) or via dust exposure arising from time spent in highly contaminated, but rarely frequented, environments.

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