FOOD RISK ASSESSMENT OF BROMINATED FLAME RETARDANTS IN THE FRENCH POPULATION

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Introduction

Brominated flame retardants (BFRs) are chemical substances incorporated in the plastic parts of electronic devices and electronic circuits to give them fire-retardant properties. They are also present in foams and padding materials (domestic and industrial), car and aircraft interiors and some textiles. This family encompasses numerous structurally different compounds, including hexabromocyclododecane (HBCD), polybrominated biphenyls (PBBs), and polybrominated diphenyl ethers (PBDEs) (209 congeners, chemically related to PCBs).
Due to their wide use, BFR became widespread environmental pollutants. Consequently, the general population is exposed to them by numerous routes (food, dust, inhalation…) however, food was shown to be the main route of exposure for some BFRs such as PBDEs (1, 2).
In laboratory animals BFRs were shown to have toxic effects particularly on hepatic, hormonal, reproductive, nervous and immunological functions. Some of these compounds accumulate in the body. Whereas carcinogenicity data are still limited, PBDEs, PBBs and HBCDs were not shown to be genotoxic.
The characterization of the chronic human toxicity of BFRs is difficult since they have often been experimentally studied as mixtures. In a recent opinion on PBBs, EFSA considered difficult to define a health-based guidance value, but proposed to compare data on exposure to PBBs with a NOAEL of 0.15 mg/kg bw/day observed in rats (induction of hepatic carcinomas) (3). Regarding PBDEs, 7 to 8 congeners are generally chosen for experimental studies. In 2006, the French food agency concluded that it was not possible to define a health-based guidance value (4) while JECFA considered that no harmful effects can occur in rodents after oral exposure to PBDE-47 and PBDE-99 (known to be the most toxic) at levels lower than 100 µg/kg bw/day (5). Given that the chemical structure of PBDEs is similar to that of NDL-PCBs, their modes of action should be similar (6). Pending the defining of a health-based guidance value for PBDEs and as a precautionary measure, Anses' panel on food contaminants proposed to compare exposure to the eight PBDEs with the threshold of 10 ng/kg bw/day defined by Afssa in 2007 for the six NDL-PCBs that are most frequently found in food (7). Regarding HBCDs, EFSA identified neurodevelopmental effects on behaviour as the critical endpoint, and derived a benchmark dose lower confidence limit for a benchmark response of 10 % ($BMDL_{10}$) of 0.79 mg/kg body weight/day. Due to the limitations and uncertainties in the current data base, EFSA concluded that it was inappropriate to use this BMDL to establish a health based guidance value, and instead used a margin of exposure (MOE) approach for the health risk assessment of HBCDs. Since elimination characteristics of HBCDs in animals and humans differ, the body burden as the starting point for the MOE approach was finally used (8).
To assess the risk led by food exposure to brominated flame retardants, 8 PBDE (PBDE-28, 47, 99, 100, 153, 154, 183 and 209), 3 PBB (PBB-52, 101 and 153) and 3 HBCD (α, β and γ stereoisomers) were measured in the food samples collected for the second total diet study performed in France (9).

Materials and methods

Food sampling

Core foods were selected to be representative of the French population diet. The selection was based on the results of the second individual and national study on food consumption survey (10, 11).

The most consumed foods by adults and/or children were selected (consumer rate of at least 5%). In addition, the main known or assumed food contributors of the substances included in this study were also selected (if not already selected by the first criterion). The core foods (n=212) covered about 90% of the whole diet of adults and children, and were divided into 41 food groups.
The sampling was performed between June 2007 and January 2009 in eight great metropolitan regions (33 cities), and each food collected in a region was sampled during two different seasons, when possible.
To be as representative as possible of the French food consumption habits, each food sample was composed of up to 15 subsamples of equal weight of the same food, taking into account the market share, origin, species, processing and packaging, flavoring, etc to be representative of the French dietary habits. Altogether, 19 830 products or subsamples were purchased, then prepared "as consumed" according to the cooking practices, e.g. vegetables and fruits were mainly washed and peeled, meat and seafood were cooked (braised, pan-fried, grilled, baked, deep-fried...). Finally, 15 subsamples were frozen and pooled by a single cryomilling phase into 1 319 composite samples for analysis. More details about the methodology can be found in Sirot et al. (5). Concentrations of the 14 BFR compounds were determined in 576 food composite samples.

Sample analysis

The extraction of fat was adapted to the physical characteristics of the samples (12, 13). The solid samples were freeze-dried and then ground. The liquid samples underwent protein precipitation through the addition of potassium oxalate. Internal standards were added before extraction (eight $^{13}C_{12}$-labelled PBDE congeners, one $^{13}C_{12}$-labelled PBB congener, three $^{13}C_{12}$-labelled HBCD congeners). After grinding, the lipid fraction was extracted using a mixture of toluene/acetone at high pressure and temperature. The solvents were then evaporated to determine the quantity of fat extracted. After reconstitution with 25 mL hexane, the extracts underwent two successive fat extractions using a solvent mixture of ethanol, ether and hexane.
Purification involved three stages involving silica, florisil and charcoal-celite columns. A quantification standard was added to each vial for each family of compounds ($^{13}C_{12}$-PBDE 138 for PBDEs and PBBs and fluorometholone for HBCDs) just before injection.
PBDEs and PBBs concentrations were determined by high-resolution gas chromatography-mass spectrometry. The detection thresholds depended on the matrices and congeners. The limits of detection were substantially lower than 0.001 ng/g fresh weight in most of the samples, allowing a significant number of congeners to be detected in most cases. The quantification of α-, β- and γ-HBCD stereoisomers was carried out by LC-MS/MS.

Dietary exposure

Values below the limits of detection or quantification are referred to as censored data. Censored data were processed according to the World Health Organization (WHO) recommendations (14). For items with a censoring rate of at least 60%, two assumptions were made about concentrations: the lowerbound (LB) and the upperbound assumption (UB). The LB assumption corresponds to a scenario in which non-detected values are estimated to be 0 and the values detected, but not quantified, are estimated to be equal to the LOD. The UB assumption corresponds to a scenario in which non-detected values are estimated to be equal to the LOD and the values detected but not quantified are estimated to be equal to the LOQ. The LB scenario represents the minimum possible value, and the UB scenario represents the maximum possible value. To estimate population dietary exposure, the mean levels of the two seasons sampled were considered for each food, both regionally and nationally, as applicable.

Dietary exposure to each contaminant was calculated individually, using the following formula:

$$E_{i,j} = \frac{\sum_{k=1}^{n} C_{i,k} \times L_{k,j}}{BW_i}$$

Where $E_{i,j}$ is dietary exposure to contaminant j of individual i, n is the number of foods in this diet, $C_{i,k}$ is the consumption of food k by individual i, $L_{k,j}$ is the level of contaminant j of food k, BW_i is the body weight of individual i.

Risk characterization

The use of mean concentrations (in composite samples) in the calculations enables a realistic and appropriate estimate of dietary exposure over the long term to the extent that these estimates are compared to the health-based guidance values listed above. Main foods contributing to the overall dietary exposure to RFB were considered (on average, over 10% according to *codex alimentarius* guidelines).

Results and discussion

Estimation of concentrations in foods

The percentage of non-detected congeners for BFRs was highly variable: it ranged from 7.1% for BDE-99 to 96.9% for PBB-101. Due to their prohibition, PBBs were generally less detected than PBDEs.

Highest mean concentrations for the sum of the 3 HBCD congeners were measured in fish, delicatessen meats, crustaceans and mollusks and meat. The other groups all had mean concentrations lower than 0.1 ng/g fw.

For the sum of the three PBB congeners, the highest concentrations were found in oils and margarine. Due to the high percentage of non-detected congeners, the lowerbound (LB) estimates were zero for several food groups.

For the sum of the seven PBDE congeners (excluding BDE-209), the food groups with the highest concentrations were fish, crustaceans and mollusks and butter. All food groups had concentrations around 4 to 12-fold lower than those reported for Europe (5). These differences may be related to the 2002 prohibition of certain formulations that started on 1 July 2006. Moreover, the data used by JECFA in 2006 were not solely European, but also American, while the PBDE profiles used in the United States can be extremely different from those used in Europe before their prohibition. When congener BDE-209 was added to the sum of the seven PBDE congeners, i.e. for the sum of the eight PBDEs, the most contaminated groups also included dairy-based desserts, sandwiches and snacks and margarine.

Table 1 : Highest mean HBCD and PBB concentrations (ng/g fresh weight) in food

		HBCD			PBB	
	N	LB	UB		LB	UB
Fish	45	0.133	0.141	Margarine	0	0.015
Delicatessen meats	80	0.132	0.140	Oils	0	0.019
Crutaceans & mollusks	37	0.131	0.135			
Meat	64	0.120	0.126			

Table 2: Highest mean PBDE concentrations (ng/g fresh weight) in food

		7-PBDE		8-PBDE	
	N	LB	UB	LB	UB
Fish	45	0.495	0.496	0.538	0.539
Crutaceans & mollusks	37	0.101	0.103	0.130	0.132
Sandwiches & snacks	18	0.045	0.047	0.152	0.154
Margarine	4	0.043	0.047	0.153	0.157
Butter	6	0.076	0.080	0.121	0.125
Dairy-based desserts	22	0.013	0.014	0.290	0.292

Estimation of the dietary exposure in the French population

Hexabromocyclododecane

Mean dietary exposure to the sum of the three HBCD congeners was up to 0.211 ng/kg bw/day for adults and up to 0.320 ng/kg bw/day for children. The main contributors for adults and children were delicatessen meats (27-29%), meat (15-21%), fish for adults (14%) and mixed dishes for children (14%).These exposures were in the same order of magnitude as those reported by EFSA in 2011 (8). When compared to the $BMDL_{10}$ of 0.79 mg/kg bw/day established on the basis of neurodevelopmental effects, these are judged as of no public health concern.

Polybrominated biphenyls

In adults, mean dietary exposure to the sum of the 3 PBB congeners was up to 0.017 ng/kg bw/day and up to 0.030 ng/kg bw/day for children. In both populations, the main contributors to exposure were fish (around 80%). In light of the NOAEL of 0.15 mg/kg bw/day that was recently established by EFSA for PBBs (3), the margin of exposure in children, at the 95th exposure percentile, was 2.5 million for the upperbound. Risk related to PBB exposure therefore does not appear to be a public health problem.

Polybrominated diphenyl ethers

Mean dietary exposure to the sum of the seven PBDE congeners was up to 0.212 ng/kg bw/day for adults and up to 0.331 ng/kg bw/day for children. Main contributors were fish for both adults and children (>33%). These exposure levels were 12 to 15-fold lower than these estimated by AFSSA in 2006 (4).

When congener PBDE 209 was added to the previous sum, exposure levels increased by a factor of 2 to 3. In adults, mean exposure was up to 0.550 ng/kg bw/day and up to 1.026 ng/kg bw/day for children. The highest contributors to exposure for adults and children were dairy-based desserts (15-23%), fish (12-17%), and ultra-

fresh dairy products (11-15%). When adopting a conservative approach, the upper end of the 95th exposure percentile of exposure in children to the eight PBDEs was over 40,000 times lower than the value set by JECFA below which no toxic effects appear (100 µg/kg bw/day).

This exposure level was also lower than the value of 10 ng/kg bw/day proposed by Anses' panel on food contaminants to characterize risk related to PBDEs. PBDEs therefore do not pose a health risk to the French population in the current state of knowledge. This conclusion is in agreement with EFSA's conclusion using an individual congener approach that considered each "of-importance" PBDE congener separately (especially, BDE-47, -99, -153 and -209) since these compounds could show different toxicological potencies (15). Individual congeners exposures are shown below, corresponding margin of exposures are in the same order of magnitude than those reported by EFSA for the general European population.

Table 3 Mean and 95th percentile (P95) of estimated exposures in the French population (ng/kg bw/day)

Mixture approach					
		Adults		Children	
		LB	UB	LB	UB
HBCDs	Mean	0.165	0.211	0.237	0.320
	P95	0.391	0.448	0.616	0.734
PBBs	Mean	0.001	0.017	0.001	0.030
	P95	0.006	0.028	0.008	0.059
7 PBDEs	Mean	0.202	0.212	0.313	0.331
	P95	0.636	0.643	0.868	0.894
8 PBDEs	Mean	0.540	0.550	1.008	1.026
	P95	1.164	1.176	1.176	2.368

Individual congener approach					
		Adults		Children	
		LB	UB	LB	UB
BDE-153	Mean	0.012	0.015	0.020	0.024
	P95	0.029	0.031	0.050	0.055
BDE-209	Mean	0.327	0.349	0.675	0.714
	P95	0.672	0.722	1.704	1.804
BDE-47	Mean	0.092	0.092	0.139	0.139
	P95	0.346	0.347	0.472	0.472
BDE-99	Mean	0.042	0.042	0.066	0.067
	P95	0.091	0.092	0.145	0.146

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