Dietary Exposure Assessment of Spanish Citizens to Hexabromocyclododecane through the Diet

ARTICLE *in* JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY · MARCH 2014

Impact Factor: 2.91 · DOI: 10.1021/jf405007x · Source: PubMed

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

pubs.acs.org/JAFC

Dietary Exposure Assessment of Spanish Citizens to Hexabromocyclododecane through the Diet

Ethel Eljarrat,*[,†] Marina Gorga,[†] Montserrat Gasser,[‡] Jordi Díaz-Ferrero,[‡] and Damià Barceló[†,§]

[†]Water and Soil Quality Research Group, Department of Environmental Chemistry, IDAEA, CSIC, Jordi Girona 18-26, 08034 Barcelona, Spain

[‡]IQS Environmental Laboratory, Universitat Ramon Llull, Via Augusta 390, 08017 Barcelona, Spain

[§]Catalan Institute for Water Research (ICRA), University of Girona, Scientific and Technological Park, H2O Building, Emili Grahit 101, 17003 Girona, Spain

ABSTRACT: A study was performed to assess exposure of the Spanish population to hexabromocyclododecane (HBCD). Based on consumption data statistics, food items from six food groups, i.e., fish and seafood, meat, animal fat, dairy products, eggs, and vegetable oils, were sampled and analyzed for HBCD followed by per capita intake calculations. The highest levels of HBCD were found in the fish and seafood samples (mean value of 11.6 ng/g lw), followed by meat samples (mean value of 2.68 ng/g lw), eggs (mean value of 1.75 ng/g lw), dairy products (mean value of 0.78 ng/g lw), animal fat (mean value of 0.74 ng/g lw), and vegetable oils (mean value of 0.45 ng/g lw). The daily ingestion rate of HBCD was estimated at 2.58 ng (kg of body weight)$^{-1}$ day^{-1}. HBCD mainly came from fish and seafood (56%), but also dairy products (14%) and meat (12%) contributed.

KEYWORDS: *brominated flame retardants, daily intake, enantiomeric fraction, food*

INTRODUCTION

Hexabromocyclododecane (HBCD) is a highly lipophilic brominated flame retardant (BFR), used in a variety of materials such as synthetic polymers, electronic equipment, plastics, textiles, and building materials, to prevent them from catching fire. Commercial HBCD is composed of a mixture of isomers, consisting mainly of γ-HBCD (70%), α-HBCD (16%), and β-HBCD (13%). The ban on the production and use of penta- and octabromodiphenyl ethers in Europe appears to be compensated by an increased use of the commercial HBCD.[1] That is because an ongoing increase of the HBCD levels in the environment has been reported by Law et al.[2] Moreover, different monitoring studies indicated its presence in wildlife and in humans, making it an ubiquitous contaminant.[3,4]

Some studies indicate a possible role of HBCD as an endocrine disruptor.[5,6] Moreover, reproduction-related effects and neurotoxic effects in mice were also found in other studies.[7] On October 2012, the Persistent Organic Pollutants Review Committee, a subsidiary body of the Stockholm Convention on Persistent Organic Pollutants (POPs), adopted a recommendation to include HBCD in the Convention's Annex A for elimination, with specific exemptions for expanded and extruded polystyrene needed to give countries time to phase in safer substitutes.

Human exposure to persistent organic pollutants (POPs) such as HBCD is mainly via the diet, and food from animal origin contained the highest levels. Dietary intake estimations are therefore important tools when estimating the exposure of POPs in a population. However, little information is available regarding HBCD levels in foods, and, therefore, not many dietary intake assessments were previously published. To our knowledge, there were no studies done yet on dietary exposure to this compound in Spain. The aim of this study was to assess the dietary intake of HBCD in the Spanish adult population by the analysis in different food groups, and compare the results with those obtained in other countries around the world. Moreover, the diastereoisomeric and enantiomeric determinations will aid to get insight into the patterns of that exposure.

MATERIALS AND METHODS

Sampling. The best of the convenient ways to estimate the human exposure to POPs via food is to analyze market basket food samples. Food samples were purchased during spring 2009 from the markets in Catalonia (NW of Spain). Since most products were obtained from brands spread across Spanish territory, food sampled for the study was representative for that consumed in the whole territory. Since HBCD is a lipohilic contaminant, only food products containing fat or animal origin were taken into account. Food items were divided into different food groups depending on their characteristics: fish and seafood, meat, animal fat, dairy products, eggs, and vegetable oils. A total of 47 food items, representatives of the variety of Spanish diet, were collected (Table 1).

Standards and Reagents. Hexane and acetone (Ultra-Resi quality), and acetonitrile, ethanol, methanol, and water (HPLC-analyzed quality) were supplied by J.T. Baker (Deventer, Holland). Dichloromethane and diethyl ether (Pestipur quality) were acquired from Carlo Erba (Val de Reuil, France), potassium oxalate (reagent grade) was provided by Fluka (Buchs, Switzerland), and concentrated hydrochloric acid (reagent grade) and concentrated sulfuric acid (reagent grade) were supplied by Scharlau (Barcelona, Spain). Alumina solid phase extraction cartridges were from IST (Uppsala, Sweden). d_{18}-labeled α-, β-, and γ-HBCDs standards were from Wellington Laboratories Inc. (Guelph, Canada) and were of minimum 98% purity. α-, β-, and γ-HBCDs standards were obtained from

Received: November 8, 2013
Revised: February 21, 2014
Accepted: March 4, 2014
Published: March 4, 2014

Table 1. Levels of HBCD in Food, Based on Samples Collected in Catalonia (NW of Spain)[a]

	no. of samples	fat (%)	α-HBCD	γ-HBCD	α-/γ-HBCD	∑HBCDs	daily ave consumption (g)[b]
pandora	1	3.73	4.75	9.64	0.5	14.4	
sole	1	0.88	11.3	6.29	1.8	17.6	
gilthead	3	9.10	3.08	2.38	2.2	5.46	
trout	3	5.32	3.15	1.29	2.4	4.44	
mackerel	3	3.71	9.49	3.03	3.4	12.5	
anchovy	2	3.75	9.45	2.17	4.5	11.6	
sardine	1	5.06	5.81	4.88	1.2	10.7	
mussels	2	2.30	7.27	9.68	1.2	17.0	
scallops	1	1.50	11.3	6.81	1.7	18.1	
cod liver	5	71.5	2.86	1.68	4.9	4.54	
∑fish and seafood	22		6.84	4.79	2.4	11.6	94.4
chicken	3	2.42	2.03	4.53	0.6	6.56	
pork	2	9.52	0.99	1.61	0.6	2.61	
turkey	2	4.66	0.35	0.45	0.8	0.81	
veal	1	7.22	0.36	0.37	1.0	0.73	
∑meat	8		0.93	1.74	0.8	2.68	167
sausage	2	32.4	0.20	0.27	0.8	0.47	
fat	2	90.1	0.60	0.40	2.0	1.00	
∑animal fat	4		0.40	0.34	1.4	0.74	20.3
milk	3	2.88	0.27	0.34	0.9	0.61	
yoghurt	2	6.52	0.27	0.32	0.8	0.59	
cheese	2	16.8	0.53	0.59	0.9	1.12	
∑dairy products	7		0.36	0.42	0.9	0.78	351
eggs	4	9.41	0.51	1.25	0.8	1.75	31.2
vegetable oils	2	100	0.17	0.28	0.9	0.45	33.6

[a]Values are given in ng/g lw and are presented as mean concentrations. [b]Data from ref 18.

Cambridge Isotope Laboratories Inc. (WI, USA) and were of minimum 97% purity.

Sample Preparation. For meat, egg, dairy products, vegetable oil, and fat samples, the equivalent to one gram of fat was weighed. For fish samples, 20 g was weighed. Each sample aliquot was spiked with d_{18}-labeled α- and γ-HBCD as internal standards. Meat and fish samples were extracted in a Soxhlet apparatus with a mixture of hexane:dichloromethane (1:1). Dairy products were extracted in a separatory funnel with a mixture of ethanol, diethyl ether, and hexane and previous addition of an aqueous potassium oxalate solution. Egg samples were hydrolyzed with concentrated hydrochloric acid, previously extracted with hexane. The treated egg samples were extracted in a separatory funnel with a mixture of diethyl ether:hexane (1:1). Oil and fat samples were dissolved in 10 mL of hexane. After the extraction, lipid content was determined gravimetrically and the resulting extracts were redissolved in hexane. Then, fat was removed by an acid treatment: extracts were cleaned up by the addition of 10 mL of concentrated sulfuric acid. Organic fraction (containing HBCD) was evaporated to 3 mL, and the whole cleanup procedure was repeated until the acid did not turn yellow. Final purification of the extract was carried out in 5 g alumina SPE cartridges. Cartridges were conditioned with 20 mL of hexane and eluted with 30 mL of hexane:dichloromethane (1:2). Purified extracts were concentrated with a gentle nitrogen flow and spiked with d_{18}-labeled β-HBCD as injection standard, and reconstituted to a final volume of 200 μL.[4]

To avoid the presence of interfering substances, glassware was rinsed with acetone and hexane just after its use. After solvent wash, it was also cleaned with water and detergent and purified water. Finally, it was rinsed again with acetone and heated at 300 °C in an oven overnight.

Diastereosiomer Analysis. A Symbiosis Pico (Spark Holland, Emmen, The Netherlands) with a Symmetry C18 column (2.1 mm × 150 mm, 5 μm) preceded by a C18 guard column (2.1 × 10 mm) supplied by Waters (Massachusetts, USA) was used for isomeric HBCD determinations. Experiments were carried out with H_2O:methanol (3:1 v/v) as eluent A and methanol as eluent B, at a flow rate of 0.25 mL/min. The elution program started at an initial composition of 100% A and was ramped to 0% A in the first eight minutes, then eluent A increased to 10% in 17 min, and initial conditions were reached again in 3 min and returned to the starting conditions in 15 min. The injection volume was set at 10 μL.

Mass spectrometric analysis was carried out with a hybrid triple quadrupole/linear ion trap MSD Sciex 4000QTRAPTM (Applied Biosystems, Foster City, CA, USA) instrument equipped with an electrospray (ESI) Turbospray interface, working in negative ionization mode. Data acquisition was performed in selected reaction monitoring (SRM). The $[M - H]^- \rightarrow Br^-$ transitions at m/z 638.7 → 78.9 and 638.7 → 80.9 were monitored for unlabeled HBCDs. The labeled HBCDs were monitored at the 655.8 → 78.9 and 655.8 → 80.9 transitions. The MS−MS detection conditions were optimized to obtain the highest relative intensity: curtain gas at 50 psi, collision gas at 4.5×10^{-5} Torr, temperature of the turbo gas in the TurboIonSpray source at 350 °C, ion source gas 1 at 50 psi, and ion source gas 2 at 10 psi.[8]

Quality Assurance/Quality Control. Method blank samples were performed to check for interferences or contamination from solvents and glassware. No presence of analytes of interest (α-, β-, and γ-HBCDs diastereoisomers) was observed. Spiked samples with native compounds (α-, β-, and γ-HBCD) were analyzed by the described methodology to check recoveries. Recovery values ranged from 79 to 119%, with a relative standard deviation (n = 5) values between 7% and 10%.

The LC−MS−MS identification was based on three main criteria. First of all, simultaneous responses for the two monitored transitions (SRM1 and SRM2) must be obtained at the same retention time than those of available standards. Then, signal-to-noise ratios must be >3. And finally, relative peak intensity ratio must be within ±20% of the theoretical values obtained with standard solutions. Limits of detection, defined as the minimum amount of analyte which produces a peak with a signal-to-noise ratio equal to 3, were 0.03 ng/g for α-HBCD, 0.05 ng/g for β-HBCD, and 0.06 ng/g for γ-HBCD. Limits of quantification, defined as the minimum amount of analyte which produces a peak with a signal-to-noise ratio equal to 10, were 0.11, 0.18, and 0.20 ng/g for α-, γ-, and β-HBCD, respectively.

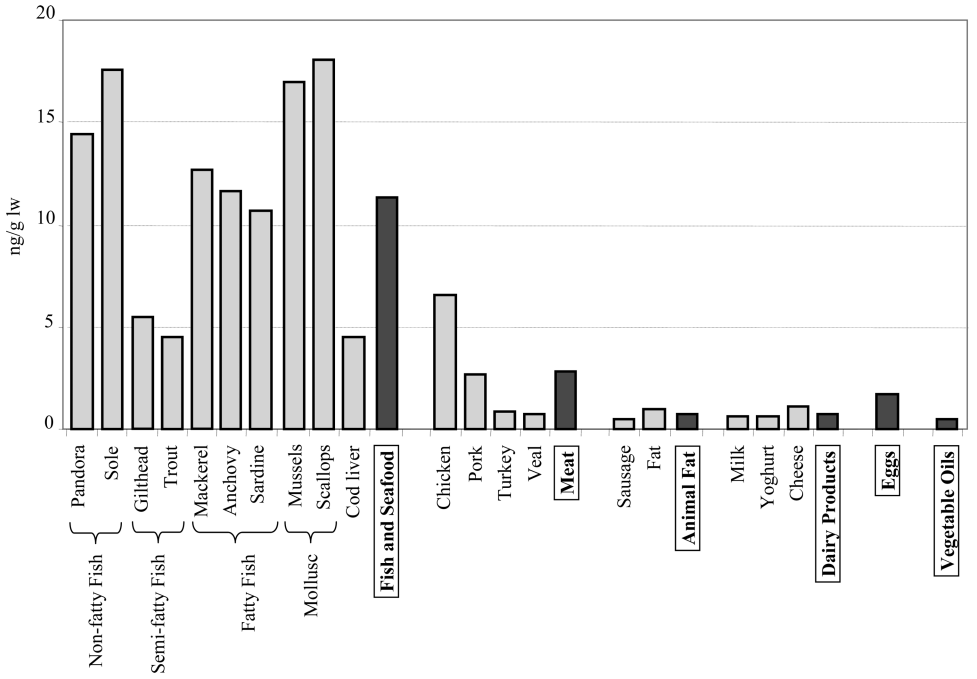

Figure 1. Occurrence of HBCD in Spanish food. Data are mean values (ng/g lw).

Enantiomeric Analysis. A Nucleodex β-PM (4.0 mm × 200 mm × 5 μm) column supplied by Macherey-Nagel (Düren, Germany) was used to perform the enantiomer-specific determination. The separation was achieved using methanol, acetonitrile, and water as mobile phase. Experiments were carried out using 70% water:30% methanol as eluent A and 70% AcN:30% methanol as eluent B, at a flow rate of 0.50 mL min^{-1}. The injection volume was set at 10 μL. The elution program started at an initial concentration of A at 50% decreased to 0% along the first 8 min, and was maintained for 17 min, and initial conditions were reached again in 5 min and maintained for additional 12 min.[9] Mass spectrometric analysis was performed using the same conditions as for diastereoisomer analysis.

Enantiomeric composition was expressed as enantiomeric fraction (EF), which is calculated by eq 1:

$$\mathrm{EF} = \frac{A_+}{(A_+ + A_-)} \quad (1)$$

where A_+ and A_- are the peak areas of eluting enantiomers. It is well-known that ESI is subjected to sample matrix effects that can cause enhancement or suppression of the target analytes' signal and can adversely affect their quantification. In order to avoid this effect that can affect EF calculations, these values are calculated according to eq 2,[10] which makes a correction based on the use of isotopic labeled standards (d_{18}-HBCDs) since d_{18}-labeled enantiomeric analogues behave in an identical manner to their native counterparts:

$$\mathrm{EF}_{\mathrm{corrected}} = \frac{([A_+]/[A_{+\mathrm{d18}}]) \times \mathrm{pg}\, A_{+\mathrm{d18}}}{([A_+]/[A_{+\mathrm{d18}}]) \times \mathrm{pg}\, A_{+\mathrm{d18}} + ([A_-]/[A_{-\mathrm{d18}}]) \times \mathrm{pg}\, A_{-\mathrm{d18}}} \quad (2)$$

where $[A_+]$ and $[A_-]$ are the peak areas of the respective (+) and (−) HBCD enantiomers, $[A_{+\mathrm{d18}}]$ and $[A_{-\mathrm{d18}}]$ are the peak areas of the respective (+) and (−) HBCD d_{18}-labeled standards, while pg $A_{+\mathrm{d18}}$ and $A_{-\mathrm{d18}}$ stand for the noncorrected determined HBCD concentrations.

RESULTS AND DISCUSSION

HBCD Occurrence in Spanish Food Samples. This study is the first to report the concentrations of α-, β-, and γ-HBCDs in different foods in Spain. HBCD levels were analyzed in fish and seafood, meat, animal fat, dairy products, eggs, and vegetable oils since HBCD is a POP and is most likely found in food with fat from animal origin. The results for the 47 products analyzed are shown in Table 1. HBCDs were detected in all analyzed foods. The highest concentrations of HBCD were found in fish and seafood (1.91−23.4 ng/g lw), followed by meat (0.73−12.9 ng/g lw), eggs (0.28−3.42 ng/g lw), dairy products (0.32−1.35 ng/g lw), animal fat (0.27−1.13 ng/g lw), and olive oils (0.18−0.72 ng/g lw). This fact would indicate a higher accumulation of HBCD in fish, similar to other POPs such as PCBs, PCDD/Fs, or PBDEs.

In samples from the meat group, HBCD levels decreased from chicken > pork > turkey > veal. As regards samples from the fish group, molluscs showed the highest levels of contamination (10.5−23.4 ng/g lw), followed by fatty fishes (10.7−14.2 ng/g lw), and then semifat fishes (3.31−7.60 ng/g lw). However, and surprisingly, HBCD levels in sole and pandora, two nonfatty fishes, are among the highest (14.4−17.6 ng/g lw) (Figure 1).

If the concentrations are expressed on fresh weight basis, the highest concentrations were those from products with high fat content: cod liver (1.37−8.44 ng/g ww) and animal fat (0.11−1.11 ng/g ww). Even in this case, fish, and seafood samples showed higher concentrations (mean value of 1.05 ng/g ww) than those detected in animal fat (mean value of 0.53 ng/g ww), vegetable oils (mean value of 0.45 ng/g ww), eggs (mean value of 0.17 ng/g ww), meat (mean value of 0.13 ng/g ww), or dairy products (mean value of 0.07 ng/g ww).

Determined HBCD concentrations were compared to those found in the literature. Most authors reported higher concentrations of HBCD in fish than in the other food categories.[11−15] Our levels in fish and seafood (mean value of 11.6 ng/g lw, or 0.63 ng/g ww) are lower than those detected by Goscinny et al.[11] in Belgium (42 ng/g lw), but higher than those reported by Törnkvist et al.[12] in Sweden (0.15 ng/g ww), Schecter et al.[15] in the US (<0.03−0.35 ng/g ww), and Shi et

dx.doi.org/10.1021/jf405007x | J. Agric. Food Chem. 2014, 62, 2462−2468

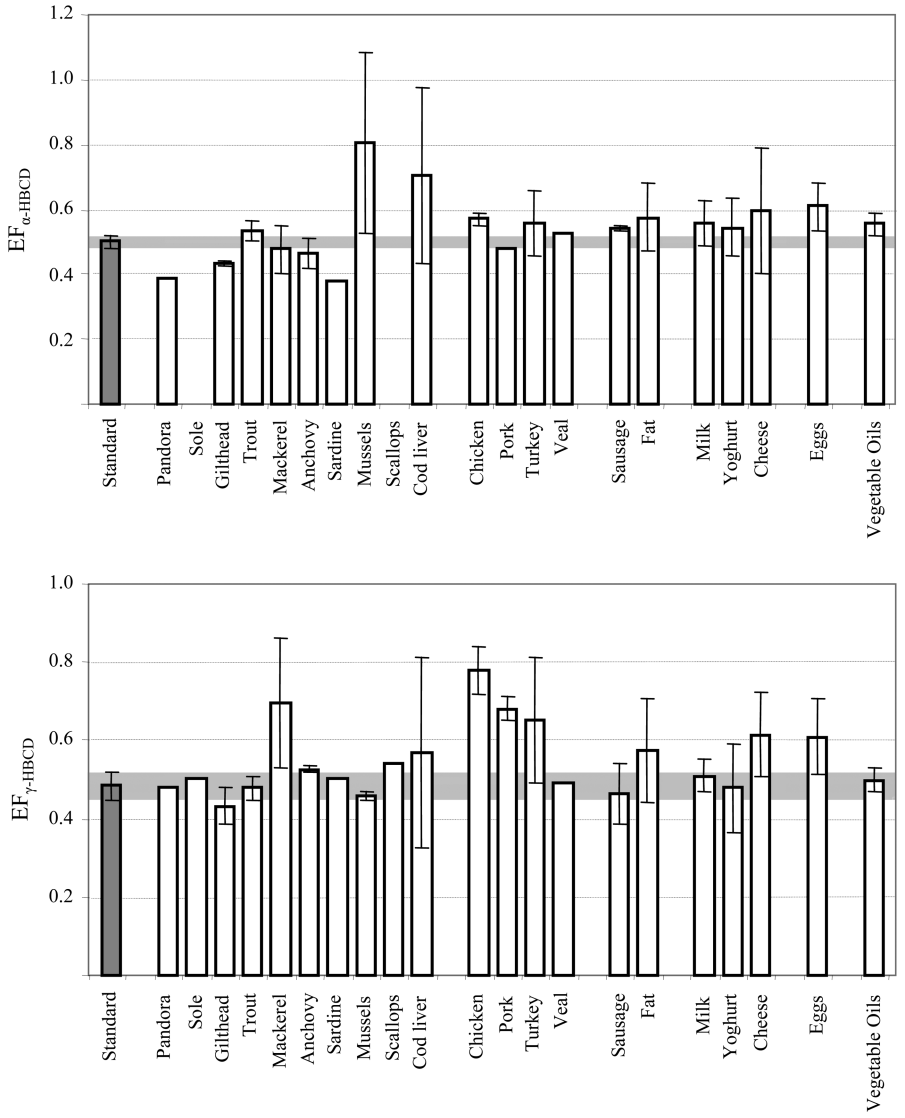

Figure 2. Enantiomeric fraction values (mean ± standard deviation) for α-HBCD and γ-HBCD in Spanish food compared to standards.

al.[13] in China (nd−2.22 ng/g lw). For some specific categories of fish and seafood (mackerel, sole, mussels), HBCD concentrations detected in our study are in the same range as those reported by Van Leeuwen and de Boer[16] in The Netherlands.

Isomeric Patterns. In all the samples, α- and γ-HBCDs were observed, whereas β-HBCD was not detected in any sample. The α- and γ-HBCD contribution was studied for the different groups of food. For fish and seafood samples, the contribution of α-HBCD was greater. In 14 out of 17 fish samples analyzed, α-HBCD concentration was higher than γ-HBCD, with α/γ ratios between 1.2 and 8.7. Similar results were found in the Belgian study, in which α-HBCD was the predominant isomer in fish samples.[11] For most of the other food samples, the trend is the inverse: γ-HBCD concentration was higher than that of α-HBCD. For meat samples, α/γ ratios ranged between 0.34 and 0.99, while for dairy products it ranged between 0.44 and 1.26. Three out of the four egg samples analyzed showed also α/γ ratios lower than 1.

Enantiomeric Fractions. EF values were calculated from the chiral column LC data, and the values were corrected according to eq 2 (Figure 2). In the case of γ-HBCD, it was observed that EF values were between 0.39 and 0.87, whereas EF for α-HBCD ranged between 0.38 and 1. If we compare these EF values with those obtained with standard solutions, in general, no significant differences were detected for γ-HBCD. The EF mean value obtained for standard solutions was 0.49 (with 7.6% of relative standard deviation (RSD)). In general, samples were between the EF value ± RSD range established with standard solutions. However, samples of mackerel (fish) (mean EF value ± SD of 0.70 ± 0.17) and chicken and pork (meat) (mean EF value ± SD of 0.78 ± 0.06 and 0.68 ± 0.03 respectively) presented an increase of EF value compared to standard value, indicating an enrichment of the (+)-γ-HBCD enantiomer in these samples.

In the case of α-HBCD, and for some fish samples (gilthead, pandora, and sardine), results showed a decrease of EF values (EF value of 0.43, 0.39, and 0.38 respectively) with respect to the standards (mean value of 0.50 with 3.6% of RSD). Thus, we can assume that an enrichment of the (−)-α-HBCD enantiomer was observed for these fish samples. Similar behavior was also detected in samples of herring muscle and falcon eggs,[17] and also significant enrichments for (−)-α-

dx.doi.org/10.1021/jf405007x | J. Agric. Food Chem. 2014, 62, 2462−2468

HBCD enantiomer was observed in human breast milk samples.[4]

Daily Intake Estimation. The estimated dietary intake (EDI) of HBCD is based on HBCD data from the present study combined with the consumption data from a Spanish diet model for determining consumer exposure to chemicals.[18] It is also important to note that we analyzed fresh foods, whereas preparation and different cooking methods can influence the levels of contaminants.[19] The Spanish dietary exposure to HBCD was 177 ng day^{-1}. Assuming the average adult body weight as 68 kg, the EDI of HBCD was 2.58 ng $(kg\ bw)^{-1}$ day^{-1}.

Our daily intake through the diet estimation was compared with other assessments in different countries around the world (Table 2). However, this comparison is not easy because there are some differences in sampling strategies, there is a lack of a standardized method for HBCD detection, and there is also a great variety in the number of analyses or different calculation methods for non detects or dietary intake. Once these parameters are taken into account, a comparison is anyway interesting.

Table 2. HBCD Daily Intake Estimation Worldwide

country	food	HBCD levels (ng/g)	daily intake (ng $(kg\ bw)^{-1}$ day^{-1})	ref
		Europe		
Sweden	fish, meat, dairy, eggs, oils	not reported	0.58−21.5	20
The Netherlands	fish, meat, dairy, eggs, oils	0.02−7.9 (ww[a])	1.5	21
The Netherlands	fish	nd[b]−230 (ww)	0.06−0.17	16
Norway	fish, meat, dairy, eggs, oils	nd−3.61 (ww)	0.3	22
Scotland, U.K.	seafood	0.03−12.1 (ww)	5.9−7.9	23
Belgium	fish, meat, dairy, egg, oils	nd−14.7 (lw[c])	0.99	11
Sweden	fish, meat, dairy, eggs, fats	nd−0.63 (ww)	0.14	12
Spain	fish, meat, dairy, eggs, oils	0.18−23.4 (lw)	2.58	this work
		Asia		
China	fish, meat, dairy, eggs	nd−9.21 (lw)	0.09−1.72	13
Japan	all market basket food	not reported	1.4−2.2	14
Japan	fish	nd−36.9 (ww)	1.3−3.7	24
Japan	seafood	12−5200 (lw)	0.45−34	25
		North America		
Canada	not reported	not reported	14	26
USA	fish, meat, dairy, eggs, vegetables	nd−0.59 (ww)	0.23	15

[a]Wet weight. [b]Not detected. [c]Lipid weight.

Some studies were carried out in different European countries. A median intake of 141 ng of HBCD per day (2.15 ng kg bw^{-1} day^{-1}) was estimated for the Swedish population, with a maximum of 1076 ng per day (21.5 ng kg bw^{-1} day^{-1}).[20] In another study,[21] the EDI was estimated at 1.5 ng kg bw^{-1} day^{-1} for the Dutch population. Later, van Leeuwen and de Boer[16] estimated the fish-related dietary exposure of HBCD for the average Dutch population. The median bound intake in that study was estimated at 8.3 ng day^{-1} for a 70 kg person (0.12 ng kg bw^{-1} day^{-1}). Dietary exposure to HBCD in the Norwegian population was established at 0.3 ng kg bw^{-1} day^{-1}.[22] The estimated adult dietary intake of HBCD arising from the United Kingdom diet resulted on higher levels than those mentioned so far, being in the range of 5.9−7.9 ng kg bw^{-1} day^{-1}.[23] This latter study only considered the intake due to seafood consumption. More recently, the intake of HBCD in Sweden was estimated by Törnkvist et al.[12] to be 0.14 ng kg bw^{-1} day^{-1} (10.2 ng day^{-1}), and Goscinny et al.[11] estimated an EDI of 0.99 ng kg bw^{-1} day^{-1} for the Belgian population. Our results were slightly higher than those reported in these studies for the European population. However, it must be taken into account that most of these studies were carried out a few years ago. It is expected that HBCD consumption has been increasing over the years due to the prohibition of PBDE use.

Few studies refer to EDI values in other continents. In Asia, Shi et al.[13] reported an EDI value in the range of 0.09−1.72 ng kg bw^{-1} day^{-1} for the Chinese population. Murata et al.[14] estimated a dietary HBCD intake of 2.2 and 1.4 ng kg bw^{-1} day^{-1} in 2002 and 2005, respectively. They analyzed 14 different food groups. However, HBCD was only detected in the fish group. In a later study, and based only on fish consumption, Nakagawa et al.[24] estimated an EDI between 1.3 and 3.7 ng kg bw^{-1} day^{-1} for the Japanese population. Higher estimation resulted from the study of Ueno et al.[25] They reported an EDI value between 0.45 and 34 ng kg bw^{-1} day^{-1}, based only on seafood consumption. These EDI values seem to be slightly higher than those reported for European citizens. Given that fish is the food group that provides the highest levels of HBCD contamination, a possible explanation could be the high consumption of fish in Asian diets.

As regards the American population, only two studies have been published dealing with HBCD EDI values. Schecter et al.[15] estimated, for the USA, an EDI value of 0.23 ng kg bw^{-1} day^{-1}, which matched reasonably well with European values. However, Environment Canada[26] established an EDI of 14 ng kg bw^{-1} day^{-1} for the Canadian population.

The calculated contributions of the different food groups to the total EDI of HBCD showed that fish and seafood was a major contributor (56%) for Spanish consumers (Figure 3). Meat (12%) and dairy products (14%) were also important

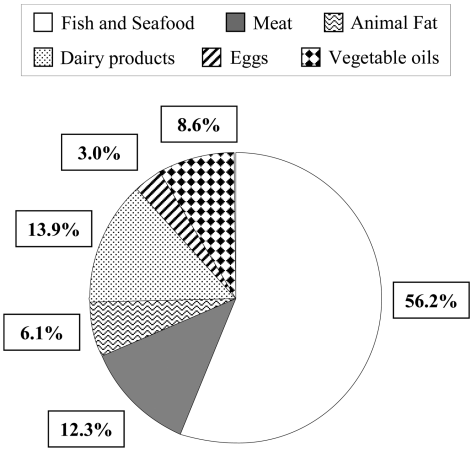

Figure 3. Relative contribution of the different food categories to the dietary intake of HBCD for Spanish consumers.

dx.doi.org/10.1021/jf405007x | J. Agric. Food Chem. 2014, 62, 2462−2468

food groups contributing to the Spanish intake of HBCD. Other studies have also reported fish as the major contributor to the HBCD intake. In the Swedish estimation[12] HBCD mainly came from fish (65%), but also dairy products (24%) and meat (10%) contributed. However, in the Belgian estimation[11] fish accounts for only 7.1% of the EDI, and the meat group was the highest contributor with 43%. This is due to the high consumption of meat and low consumption of fish in Belgium.

Previous studies have been carried out to estimate the human exposure to PBDEs through the diet in Spain. Bocio et al.[27] estimated the dietary intake of PBDEs for the general population living in Catalonia (Spain), and they found a value of 97 ng day^{-1} (1.4 ng kg bw^{-1} day^{-1}). A later study in the same area[28] showed an EDI decrease of 23%, with value of 1.1 ng kg bw^{-1} day^{-1}. An even lower value was reported by Gómara et al.[29] in a study including different locations in Spain, with an EDI value of 39 ng day^{-1} (0.6 ng kg bw^{-1} day^{-1}). However, these differences could be attributed in part to the different consumption data applied in each study, but also to other factors such as the sampling strategy, the way concentrations were given (upper, medium, and lower bound) or the number of PBDE congeners analyzed. In any case, the three values (0.6−1.4 ng kg bw^{-1} day^{-1}) obtained for the dietary intake of PBDEs in Spain were lower than the value obtained in our study for the dietary intake of HBCD (2.6 ng kg bw^{-1} day^{-1}).

Human exposure to HBCD occurs through multiple routes. For non occupationally exposed persons, the major intake of HBCD is probably from food and indoor air or dust. However, the relevance of human HBCD exposure originating from house dust versus food-based HBCD exposure is still unknown. Nondietary exposure, mainly through dust in homes, offices, schools, cars, and public environment can substantially contribute, and in some cases even dominate, the total human exposure to HBCD, especially for toddlers and other children.[30] Our study has focused on the intake of HBCD through the diet. Estimated daily intake of this contaminant for the Spanish population resulted in 2.58 ng (kg bw)$^{-1}$ day^{-1}. The main contributor to this intake was fish group, followed by dairy products and meat. Therefore, the exposure of frequent fish consumers could be higher than for the average consumers estimated in our study.

AUTHOR INFORMATION

Corresponding Author

*Tel: +34 934006100. E-mail: eeeqam@cid.csic.es.

Funding

This work was funded by the Spanish Ministry of Economy and Competitiveness through the project SCARCE (Consolider Ingenio 2010 CSD2009-00065). This work has also been partly funded by the Generalitat de Catalunya (Consolidated Research Group Water and Soil Quality Unit 2009-SGR-965).

Notes

The authors declare no competing financial interest.

REFERENCES

(1) EFSA. The European Food Safety Authority, 2006. Advice of the Scientific Panel on contaminants in the food chain on a request from the Commission related to relevant chemical compounds in the group of brominated flame retardants for monitoring in feed and food. EFSA J 328, 1−4. http://www.efsa.eu.int.

(2) Law, R. J.; Bersuder, P.; Allchin, C. R.; Barry, J. Levels of the flame retardants hexabromocyclododecane and tetrabromobisphenol A in the blubber of harbour porpoises (Phocoena phocoena) stranded or bycaught in the UK, with evidence for an increase in HBCD concentrations in recent years. *Environ. Sci. Technol.* **2006**, *40*, 2177−2183.

(3) Guerra, P.; de la Cal, A.; Marsh, G.; Eljarrat, E.; Barceló, D. Transfer of hexabromocyclododecane from industrial effluents to sediments and biota: Case study in Cinca river (Spain). *J. Hydrol.* **2009**, *369*, 360−367.

(4) Eljarrat, E.; Guerra, P.; Martínez, E.; Farré, M.; Alvarez, J. G.; López-Teijón, M.; Barceló, D. Hexabromocyclododecane in human breast milk: levels and enantiomeric patterns. *Environ. Sci. Technol.* **2009**, *43*, 1940−1946.

(5) Palace, V. P.; Pleskach, K.; Halldorson, T.; Danell, R.; Wautier, K.; Evans, B. Biotransformation enzymes and thyroid axis disruption in juvenile rainbow trout (*Oncorhynchus mykiss*) exposed to hexabromocyclododecane diastereoisomers. *Environ. Sci. Technol.* **2008**, *42*, 1967−72.

(6) van der Ven, L. T. M.; Slob, W.; Piersma, A. H.; Opperhuizen, A. The benchmark approach is the preferred method to describe the toxicology of tetrabromobisphenol A (TBBPA) - response to Banasik et al. (2009). *Toxicology* **2009**, *260*, 153−154.

(7) Eriksson, P.; Fischer, C.; Wallin, M.; Jakobsson, E.; Fredriksson, A. Impaired behaviour, learning and memory, in adult mice neonatally exposed to hexabromocyclododecane (HBCDD). *Environ. Toxicol. Pharmacol.* **2006**, *21*, 317−322.

(8) Guerra, P.; de la Torre, A.; Martínez, M. A.; Eljarrat, E.; Barceló, D. Identification and trace level determination of brominated flame retardants by LC-quadrupole linear ion trap (Qq-LIT) mass spectrometry. *Rapid Commun. Mass Spectrom.* **2008**, *22*, 916−924.

(9) Guerra, P.; Eljarrat, E.; Barceló, D. Enantiomeric specific determination of hexabromocyclododecane by LC-quadrupole linear ion trap (Qq-LIT) mass spectrometry in sediment samples. *J. Chromatogr.* **2008**, *1023*, 81−87.

(10) Marvin, C.; MacInnis, G.; Alahee, M.; Arsenault, G.; Tomy, G. Factors influencing enantiomeric fractions of hexabromocyclododecane measured using liquid chromatography/tandem mass spectrometry. *Rapid Commun. Mass Spectrom.* **2007**, *21*, 1925−1930.

(11) Goscinny, S.; Vandevijvere, S.; Maleki, M.; Van Overmeire, I.; Windal, I.; Hanot, V.; Blaude, M. N.; Vleminckx, C.; Van Loco, J. Dietary intake of hexabromocyclododecane diastereoisomers (α-, β-, and γ-HBCD) in the Belgian adult population. *Chemosphere* **2011**, *84*, 279−288.

(12) Törnkvist, A.; Glynn, A.; Aune, M.; Darnerud, P. O.; Ankarberg, E. H. PCDD/F, PCB, PBDE, HBCD and chlorinated pesticides in a Swedish market basket from 2005 − Levels and dietary intake estimations. *Chemosphere* **2011**, *83*, 193−199.

(13) Shi, Z. X.; Wu, Y. N.; Li, J. G.; Zhao, Y. F.; Feng, J. F. Dietary exposure assessment of Chinese adults and nursing infants to tetrabromobisphenol-A and hexabromocyclododecanes: occurrence measurements in foods and human milk. *Environ. Sci. Technol.* **2009**, *43*, 4314−4319.

(14) Murata, S.; Nakagawa, R.; Ashizuka, Y.; Hori, T.; Yasutake, D.; Tobiishi, K.; Sasaki, K. Brominated flame retardants (HBCD, TBBPA and ΣPBDEs) in market basket food samples of Northern Kyushu district in Japan. *Organohalogen Compd.* **2007**, *69*, 1985−1988.

(15) Schecter, A.; Haffner, D.; Colacino, J.; Patel, K.; Paepke, O.; Opel, M.; Birnbaum, L. S. Polybrominated Diphenyl Ethers (PBDEs) and Hexabromocyclodecane (HBCD) in Composite U.S. Food Samples. *Environ. Health Perspect.* **2010**, *118*, 357−362.

(16) van Leeuwen, S. P.; de Boer, J. Brominated flame retardants in fish and shellfish - levels and contribution of fish consumption to dietary exposure of Dutch citizens to HBCD. *Mol. Nutr. Food Res.* **2008**, *52*, 194−203.

(17) Janák, K.; Sellström, U.; Johansson, A. K.; Becher, G.; de Wit, C.; Lindberg, P.; Helander, B. Enantiomer-specific accumulation pattern of hexabromocyclododecane in eggs of predatory birds breeding in Sweden. *Organohalogen Compd.* **2005**, *67*, 204−207.

(18) AESAN, 2006. Modelo de dieta española para la determinación de la exposición del consumidor a sustancias químicas. Rev. 1. Available at http://www.aesan.msc.es/AESAN/docs/docs/notas_prensa/modelo_dieta_espanola.pdf.
(19) Perelló, G.; Martí-Cid, R.; Castell, V.; Llobet, J. M.; Domingo, J. L. Concentrations of polybromianted diphenyl ethers, hexachlorobenzene and polycyclic aromatic hydrocarbons in various foodstuffs before and after cooking. *Food Chem. Toxicol.* **2009**, *47*, 709−715.
(20) Lind, Y.; Aune, M.; Atuma, S.; Becker, W.; Bjerselius, R.; Glynn, A.; Darnerud, P. O. Food intake of the polybrominated flame retardants PBDEs and HBCD in Sweden. *Organohalogen Compd.* **2002**, *58*, 181−184.
(21) de Winter-Sorkina, R.; Bakker, M. I.; van Donkersgoed, G.; van Klaveren, M. D. Dietary intake of brominated flame retardants by the Dutch population. RIVM report n° 310305001/**2003**.
(22) Knutsen, H. K.; Kvalem, H. E.; Thomsen, C.; Froshaug, M.; Haugen, M.; Becher, G.; Alexander, J.; Meltzer, H. M. Dietary exposure to brominated flame retardants correlates with male blood levels in a selected group of Norwegians with a wide range of seafood consumption. *Mol. Nutr. Food Res.* **2008**, *52*, 217−227.
(23) Fernandes, A.; Dicks, P.; Mortimer, D.; Gem, M.; Smith, F.; Driffield, M.; White, S.; Rose, M. Brominated and chlorinated dioxins, PCBs and brominated flame retardants in Scottish shellfish: methodology, occurrence and human dietary exposure. *Mol. Nutr. Food Res.* **2008**, *52*, 238−249.
(24) Nakagawa, R.; Murata, S.; Ashizuka, Y.; Shintani, Y.; Hori, T.; Tsutsumi, T. Hexabromocyclododecane determination in seafood samples collected from Japanese coastal areas. *Chemosphere* **2010**, *81*, 445−452.
(25) Ueno, D.; Isobe, T.; Ramu, K.; Tanabe, S.; Alaee, M.; Marvin, C.; Inoue, K.; Someya, T.; Miyajima, T.; Kodama, H.; Nakata, H. Spatial distribution of hexabromocyclododecanes (HBCDs), polybrominated diphenyl ethers (PBDEs) and organochlorines in bivalves from Japanese coastal waters. *Chemosphere* **2010**, *78*, 1213−1219.
(26) Environment Canada, 2010. Draft Screening Assessment: Cyclododecane, 1,2,5,6,9,10-hexabromo. Available at http://www.ec.gc.ca/lcpe-cepa/default.asp?lang=En&n=A27E7A60-1.
(27) Bocio, A.; Llobet, J. M.; Domingo, J. L.; Corbella, J.; Teixidó, A.; Casa, C. Polybrominated diphenyl ethers (PBDEs) in foodstuffs: human exposure through the diet. *J. Agric. Food Chem.* **2003**, *51*, 3191−3195.
(28) Domingo, J. L.; Martí-Cid, R.; Castell, V.; Llobet, J. M. Human exposure to PBDEs through the diet in Catalonia, Spain: temporal trend. A review of recent literature on dietary PBDE intake. *Toxicology* **2008**, *248*, 25−32.
(29) Gómara, B.; Herrero, L.; González, M. J. Survey of polybrominated diphenyl ether levels in Spanish commercial foodstuffs. *Environ. Sci. Technol.* **2006**, *40*, 7541−7547.
(30) EFSA. Scientific opinion on hexabromocyclododecanes (HBCDDs) in food. *EFSA J.* **2011**, *9* (7), 2296.