DIETARY INTAKE OF HEXABROMOCYCLODODECANE IN JAPAN

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Introduction

Hexabromocyclododecane (HBCD) is a brominated flame retardant (BFR) that has been used for many years in plastics and textile coatings around the world. In Japan, the domestic use of HBCD has recently become more common, as it has increasingly replaced or supplemented other BFRs. A total of 2,600 t of HBCD was produced in or imported into Japan in 2011.

HBCD's toxicity and the environmental threat it poses are subjects of current discussion. HBCD has been identified in environmental samples from birds, mammals, fish and other aquatic organisms, as well as in soil and sediment. In May 2013, the Stockholm Convention on Persistent Organic Pollutants (POPs) added HBCD to the Convention's Annex A for elimination. In May 2014, Japan added HBCD to its list of class I specified substances of the Chemical Substances Control Law to enact a ban on the import and production of HBCD. However, HBCD is still diffusing into the environment from waste materials that contain HBCD.

It is important to estimate humans' dietary intakes of HBCD. A total diet (TD) study is a useful method of estimating the average dietary intake of contaminants. We reported that the Japanese populace is exposed to HBCD mostly via fish and shellfish among the TD samples studied[1,2]. The dietary intake of HBCD can thus be estimated in an analysis of TD samples consist of fish and shellfish. In the present study, we analyzed HBCD in the TD samples and estimated the daily intake of HBCD across Japan.

Materials and methods

Chemicals

Non-labeled and $^{13}C_{12}$-labeled α-, β-and γ-HBCD analytical standards were purchased from Cambridge Isotope Laboratories (Andover, MA, USA). Dichloromethane, n-hexane, acetone, cyclohexane, methanol and distilled water (washed with hexane) of dioxin or pesticide analysis grade were purchased from Kanto Chemical (Tokyo, Japan). The 44% sulfuric acid-impregnated silica gel was purchased from Wako Pure Chemical Industries (Osaka, Japan).

Samples

TD samples were prepared at 10 locations in seven regions across Japan in 2012. The constituents of the TD samples were designed based on the official food classification and consumption data obtained by the National Health and Nutrition Survey in Japan. The collected various kinds of fish and shellfish were cooked or prepared in typical ways for consumption. The samples were then blended for TD samples. The TD samples were maintained at temperatures below –20°C until analysis.

Sample Preparation

The protocol for the HBCD analysis is illustrated in Fig. 1. Each 5 g of TD sample was mixed with 5 g of diatomaceous earth powder. After mixing, the sample was spiked with $^{13}C_{12}$-labeled α-, β- and γ-HBCD, and was extracted using a Dionex ASE-350 Accelerated Solvent Extractor (Thermo Scientific, Sunnyvale, CA, USA) under conditions of 1,500 psi, with acetone/hexane (1:3) as an extraction solvent. The extracts were shaken with 5% sodium chloride solution, and the organic layer was dried over anhydrous sodium sulfate and concentrated to dryness in order to determine the lipid content gravimetrically. The residue was dissolved in 10 mL of acetone/cyclohexane (3:7), and 2 mL of this solution was subjected to gel permeation chromatography. HBCD was fractionated over 12.5–16.5 min after large molecules such as crude fatty acids eluted in 10–12 min. The fraction was concentrated and dissolved in 0.3 mL of hexane, re-purified with a 1 g of 44% sulfuric acid-impregnated silica gel-packed mini-column, and then reconstituted to 50 μL using methanol.

Analytical Methods and Instrumentation

The HBCD concentrations were determined using liquid chromatography/mass spectrometry (LC/MS) performed on a Quattro Ultima Pt mass spectrometer (Waters, Milford, MA, USA) connected to a 2695 liquid

chromatography system (Waters). The HPLC column was a 150 mm × 2.1 mm i.d. 5-μm Inertsil ODS-3 (GL-Science, Tokyo, Japan). The detection limit of both α- and γ-HBCD was 0.02 ng/g wet weight (ww); that for β-HBCD was 0.01 ng/g ww.

Results and discussion

Table 1 presents the results of the HBCD analysis of the TD samples (aggregated sample of cooked fish and shellfish following those consumption data). HBCD was detected in all samples prepared at all 10 locations. The concentrations of detected total HBCD were 0.17–1.24 ng/g ww when ND was assumed to be zero. The mean concentration of total HBCD was 0.68 ng/g ww. The concentrations of the isomers of all samples were in the order: α-> γ- > β-HBCD. We observed no correlation between the HBCD concentration and the fat content.

The estimates of HBCD daily intake basing on the concentration and consumption data are summarized in Table 2. As mentioned earlier, the Japanese populace is exposed to HBCD mostly via fish and shellfish, so that the daily intakes estimated basing on the concentrations of the TD sample consist of various kinds of fish and shellfish can be considered the entire dietary intake. The daily intake of HBCD was estimated to be 13.1–86.9 ng/day. In the case of 50 kg of body weight (bw), the daily intake was calculated as 0.26–1.74 ng/kg bw/day. In Japan, it was reported that the no observable adverse effect level (NOAEL) for HBCD was 10.2 mg/kg bw/day. Using an uncertain (safety) factor of 100, this suggests that the total daily intake of HBCD should be less than 0.102 mg/kg bw/day. Compared with this value, the levels of the HBCD obtained in this study are not considered a serious problem.

Acknowledgements

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References

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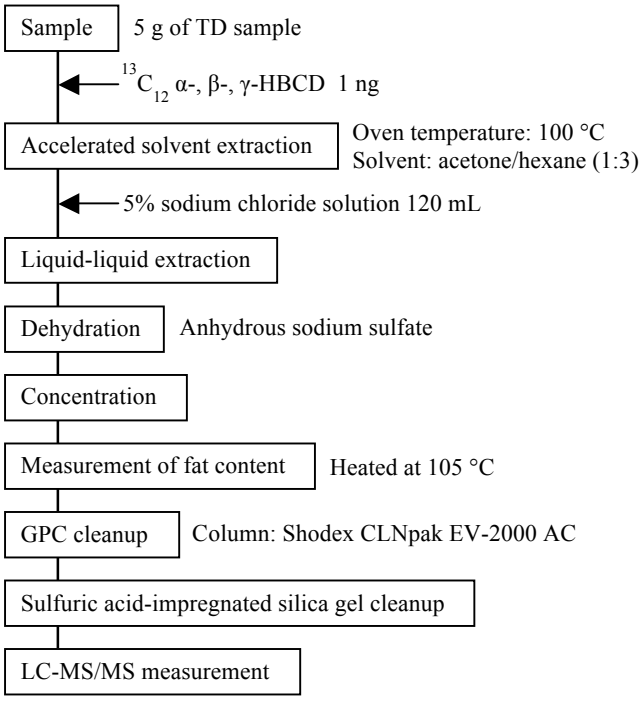

Fig.1 Protocol for HBCD analysis

Table 1 Concentrations of HBCD in the TD samples

No.	Location	Fat content (%)	HBCD concentration (ng/g ww)			
			α-HBCD	β-HBCD	γ-HBCD	Total HBCD
1	A	7.15	0.71	ND	0.08	0.79
2	B	6.27	0.66	ND	0.09	0.75
3	C	5.13	1.14	ND	0.10	1.24
4	D	5.98	0.70	ND	0.12	0.82
5	E	5.38	0.62	ND	0.10	0.72
6	F	4.76	0.43	ND	0.08	0.51
7	G	5.04	0.17	ND	ND	0.17
8	H	3.38	0.30	ND	0.04	0.34
9	I	3.90	1.14	ND	0.08	1.22
10	J	5.31	0.22	ND	0.03	0.25
Mean						0.68

Table 2 Daily intake of HBCD in 10 locations in Japan

No.	Location	Daily intake (ng/day)
1	A	65.9
2	B	49.5
3	C	81.8
4	D	67.1
5	E	58.9
6	F	32.8
7	G	13.1
8	H	22.5
9	I	86.9
10	J	17.8
Mean		49.6