TEMPORAL TREND STUDIES ON HEXABROMOCYCLODODECANE (HBCD) IN MARINE MAMMALS FROM ASIA-PACIFIC

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Introduction

Hexabromocyclododecane (HBCD, $C_{12}H_{18}Br_6$) is the principal brominated flame retardant (BFR) in polystyrene foams used mainly as thermal insulation in the building industry, but is also used in upholstery textiles including furniture, draperies and wall coverings.[1] Technical mixtures contain the isomers α-, β-, and γ-HBCDs, which are diastereoisomers, but the γ-isomer always forms the majority (>70%) with a small amount of the others. The isomeric compositions of the residues in biota, however, show an increase of α-HBCD substantially different from the technical mixtures.[2] The factors influencing the isomeric pattern of HBCD have not been fully understood. Biomagnification of HBCD is demonstrated in the food webs in Lake Ontario[3] and in the North Sea[2], facilitated by their high lipophilicity and resistance to degradation in the environment. Since widening pollution and potential environmental impacts of HBCD are of particular concern, Japanese government classifies this chemical as a Type I Monitoring Chemical Substance, whereas in Europe and the U.S. it is not subject to regulatory restriction.

The global demand for HBCD has increased as an alternative to certain formulations of polybrominated diphenyl ethers (PBDEs) which are regulated by RoHS (the restriction of the use of certain hazardous substances in electrical and electronic equipment) Directive in EU which will come into force on 1 July 2006. Temporal studies in Europe and North America have shown that the levels of HBCDs are increasing in environmental matrices,[4,5] presumably due to the increased use of this BFR in consumer products. In Japan, HBCD consumption tends to increase slightly, and its annual consumption (2600 tons) in 2004 eventually exceeded that of the DecaBDE (2000 tons). However, there is a lack of information on HBCD contamination in Asian region despite its relatively heavy usage, ca. 25% of global demand in 2001.

In the present study, levels and temporal trends of HBCD contamination in Asia-Pacific waters were investigated by analyzing its diastereoisomers (α, β, and γ) in the fat tissue of marine mammals archived during 1972-2001.

Materials and Methods

Samples: The archived fat tissue of three marine mammal species including female northern fur seals (*Callorhinus ursinus*) collected off Sanriku, Japan during 1972-1998 (*n*=35), male melon-headed whales (*Peponocephala electra*) mass stranded along Japanese coast in 1982 and 2001 (*n*=10), and male finless porpoises (*Neophocaena phocaenoides*)

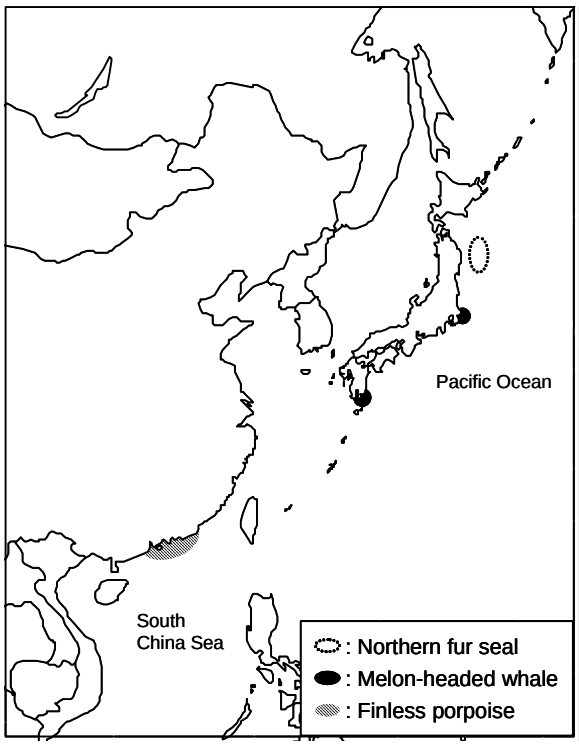

Figure 1. Map showing the sampling locations of marine mammals from Asia-Pacific.

incidentally caught at coastline of South China Sea in 1990 and 2000/01 (*n*=12) were employed for chemical analysis. Sampling locations and biological data of the animals analyzed in the present study are given in Figure 1 and Table 1, respectively. Fat tissue were excised from the animals and kept in a deep freezer at -20°C until chemical analysis.

Chemical Analysis: Fat samples were extracted in a Soxhlet apparatus with a mixture of diethyl ether and hexane. An aliquot of the extract was added to a gel permeation chromatography (GPC) column for lipid removal. The GPC fraction containing organohalogens was concentrated and passed through an activated silicagel (Wakogel S-1) column for further clean-up. Identification and quantification of HBCD isomers was performed using liquid chromatography combined with tandem mass spectrometry (LC-MS-MS) in a multiple reaction monitoring (MRM) mode.

Results and Discussion

The residual concentrations of northern fur seals, melon-headed whales, and finless porpoises are summarized in Table 1. HBCDs were detected in almost all the samples analyzed except northern fur seals collected in 1972, indicating widespread contamination by HBCDs in Asia-Pacific. The HBCD residue levels measured in this study are apparently lower than levels in seals and cetaceans from Europe[2,6] and comparable with those in California sea lion (*Zalophus californianus*).[5] This trend may reflect the greater demand for HBCD in Europe relative to Americas and Japan. To our knowledge, this is the first comprehensive study revealing the accumulation of HBCDs in marine mammals from the Asia-Pacific waters.

Northern fur seal

HBCDs in northern fur seals from off Sanriku ranged in concentration from a low of <0.1 ng/g lipid weight in 1972 to a high of 67 ng/g lipid weight in 1997 (Table 1). The α-HBCD was the predominant isomer contributing more than 95% of the total HBCDs in all the samples analyzed. No β-isomer was found at the quantification limit of the analysis (0.1 ng/g lipid weight).

Retrospective analysis of fat tissue of fur seals demonstrated that HBCD residues increased from 1972 to mid-1990s, and then slightly decreased in 1998 to about 70 % of its peak values (Figure 2). Although γ-HBCD was detected in the samples collected after 1990, isomeric composition did not change temporally, implying that the commercial mixtures of the same composition have been used around the Pacific during 1972-1998. Fur seals collected prior to 1991 accumulated HBCDs levels apparently lower than PBDEs,[7] whereas comparable levels of these BFRs were detected in the late 1990s (Table 1). Annual consumption of HBCD in Japan is about 2000 tons from the year 2000, while the use of TetraBDE and OctaBDE were voluntarily withdrawn from the Japanese market in 1991 and 2000, respectively (Figure 2). The appearance of the peak contamination of HBCDs in fur seal tissues later than PBDEs may be related to the relative production and use of HBCD compared to PBDEs in Japan.

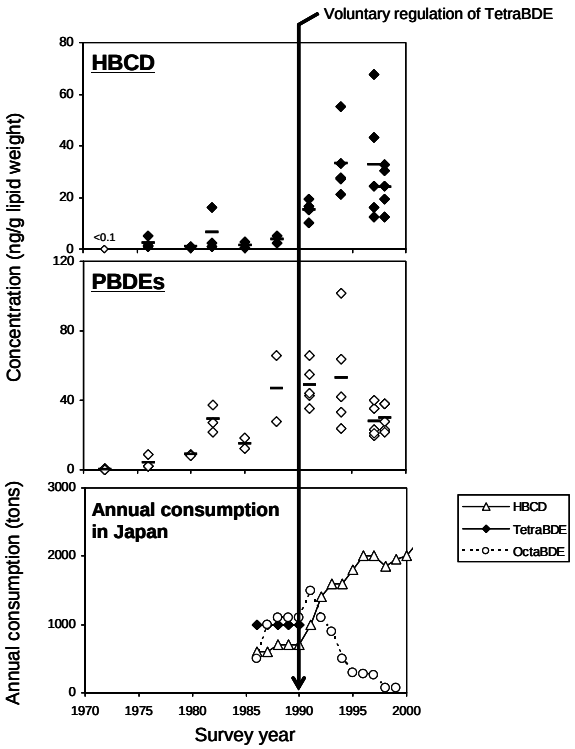

Figure 2. Comparison between temporal trend in levels of HBCD and PBDEs[7] in northern fur seals from the Pacific and the annual consumption of commercial HBCD and PBDE products in Japan. Horizontal bars indicate mean values.

Table 1. Biological information and concentrations of BFR (ng/g lipid wt.) in the fat of marine mammals from Aisa-Pacific[a]

Species	Location	Year	*n*	Age	BL (cm)	Lipid (%)	HBCDs	PBDEs[b]
Northern fur seal	off Sanriku, Japan	1972	3	20 (20-21)	128 (125-130)	67 (64-69)	<0.1 -	0.34 (0.33-0.34)
		1976	3	21 (20-21)	134 (132-136)	22 (2.7-60)	2.4 (0.69-5.1)	4.3 (2.0-8.7)
		1980	2	21 (20, 21)	135 (131-138)	75 (71-79)	0.77 (0.56-0.99)	8.5 (8.1-8.9)
		1982	3	21 (20-22)	127 (120-136)	33 (23-38)	6.5 (0.84-16)	29 (22-37)
		1985	2	23 (22, 23)	140 (132-148)	54 (48-60)	1.6 (0.46-2.7)	15 (12-18)
		1988	2	20 (20, 20)	135 (131-138)	76 (70-82)	3.6 (2.3-5.0)	47 (22-66)
		1991	5	18 (17-20)	128 (123-134)	74 (64-80)	15 (10-19)	49 (35-66)
		1994	5	15 (12-21)	130 (126-134)	75 (72-80)	33 (21-55)	53 (24-100)
		1997	5	16 (11-22)	130 (125-135)	82 (77-100)	33 (13-67)	28 (21-40)
		1998	5	14 (12-17)	133 (121-140)	78 (72-88)	24 (12-32)	30 (22-38)
Melon-headed whale	the Pacific coast, Japan	1982	5	na	248 (236-258)	62 (46-80)	7.0 (2.7-9.7)	26 (23-32)
		2001	5	na	256 (245-270)	68 (63-74)	390 (330-460)	320 (300-340)
Finless porpoise	South China	1990	7	na	149 (121-167)	82 (71-92)	18 (4.7-37)	110 (84-170)
		2000/01	5	na	149 (121-163)	47 (32-65)	35 (21-55)	710 (230-980)

[a] Figures in parentheses indicate the range
[b] Data were cited from refs 7-9.
n : number of samples
BL: body length
na: no data available

Melon-headed whale

Melon-headed whales used in this study were mass stranded at the Pacific coast of Japan in 1982 and 2001, which provided an opportunity to examine the changes in the accumulation of BFRs in the past 20 years around Japan. HBCD levels in 2001 were significantly higher than 1982, showing about fifty times increase (Table 1). Among the three isomers identified, α-HBCD was the only isomer detected in the animals in 1982, but small quantities of β- and γ-isomers were also accumulated in whales collected in 2001. Occurrence of β-HBCD in whale blubber confirms at least some release into the environment and uptake and bioaccumulation of β-isomer which is present in commercial mixture only in minute amount. HBCD levels were one order of magnitude lower than PBDEs[8] in the specimens collected in 1982, but in 2001, HBCD levels exceeded PBDEs, showing a similar pattern observed in northern fur seals.

Finless porpoise

HBCDs were also detected in all the finless porpoises incidentally caught at South China Sea coast (Table 1), indicating that the contamination by HBCD has already become evident even in developing countries. In general, the diastereomeric profiles in finless porpoises were also strongly dominated by α-HBCD. Comparing the BFR residue levels in porpoises collected in 1990 and 2000/01, HBCD levels doubled during the past decade, whereas PBDE levels increased about five times[9]. Unlike in the case of fur seals and whales collected around Japan, HBCD concentrations in finless porpoises in 2000/01 were still much lower than PBDEs, approximately twenty times lower. These results may indicate that HBCD demand as an alternative for PBDE formulations in South China is not increasing rapidly.

Few studies report the temporal trends of HBCDs with PBDEs in biological samples. Sellström et al.[4] showed an increase in HBCDs in guillemot (*Uria algae*) eggs from the Baltic Sea from the early 1980s, but concentrations have remained relatively stable from 1991 to 2001, whereas PBDE levels decreased rapidly from late 1980s. Another study conducted in U.S. demonstrated that HBCD appeared to be increasing in the blubber of California sea lions between 1993 and 2003 whereas PBDE concentrations appear to be stable.[5] On the other hand, peregrine falcon (*Falco peregrinus*) eggs from South Greenland showed a significantly increasing trend of PBDE concentrations with no clear trend for HBCD.[10] Considering the apparent rate of increase in HBCD concentrations in marine mammals from Asia-Pacific in this study, and the potential toxicity concerns, more studies on the loadings and concentrations of HBCD in other environmental media may be warranted.

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