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Tissue-specific accumulation of polybrominated diphenyl ethers (PBDEs) including Deca-BDE and hexabromocyclododecanes (HBCDs) in harbor seals from the northwest Atlantic

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ABSTRACT

Polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecanes (HBCDs) are widely used flame retardants that enter coastal waters from multiple sources and biomagnify in marine food webs. PBDEs have been detected at relatively high concentrations in harbor seals, apex predators in the northwest Atlantic. Whereas tri- to hexa-BDEs readily biomagnified from prey fishes to seal blubber, Deca-BDE (BDE-209) did not biomagnify in blubber. To explore tissue-specific differences in the accumulation/biomagnification of BFRs, we analyzed tri- to Deca-BDES in liver of 56 harbor seals (6 adult males, 50 pups), and compared hepatic concentrations and biomagnification potential with those in blubber. HBCDs were analyzed in seal liver and blubber to enable similar comparisons. Hepatic ΣPBDE (tri- to Octa-BDE) concentrations (range 35–19,547 ng/g lipid weight, lw) were similar to blubber concentrations, while α-HBCD levels in seal liver (range 2–279 ng/g lw) were significantly higher than levels in blubber. Tissue distribution of PBDEs and α-HBCD varied significantly by age and, surprisingly, by gender among the pups. Biomagnification of α-HBCD from fish to seal liver and blubber was negligible to low, implying that harbor seals can metabolize this persistent isomer. Similar to the patterns in blubber, tri- through hexa-BDEs were highly biomagnified from fish to seal liver. In contrast, BDE-209 concentrations in liver were up to five times higher than those in blubber, which is consistent with observations that BDE-209 migrates to perfused tissues such as the liver in biota. Although detection frequency was low, BDE-209 levels in seal liver were up to ten times higher than those in their prey fish, suggesting that the accumulation/biomagnification of Deca-BDE in marine food webs is tissue-specific. As BDE-209 is the dominant PBDE found in marine sediments, its biomagnification in marine ecosystems is of concern.

1. Introduction

Polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecanes (HBCDs) are brominated flame retardants (BFRs) that have been widely used as additives in household and commercial products to increase their flame ignition resistance and meet fire safety standards (Alaee et al., 2003; Covaci et al., 2006; Shaw et al., 2010). Yet, their propensity to leach from the products and to become ubiquitous global contaminants has raised concerns in recent years (DiGangi et al., 2010, Shaw and Kannan, 2009). BFRs enter coastal and marine waters from multiple sources, readily biomagnify in marine food webs, and are found at high concentrations in top predators such as marine mammals (Covaci et al., 2006; de Wit, 2002; Hites, 2004; Shaw and Kannan, 2009).

Because of health and environmental concerns, the Penta- and Octa-BDE commercial mixtures have been banned or phased out of production and use in the US and Europe, although they are still used in other parts of the world (Shaw and Kannan, 2009; Shaw et al., 2010). In May 2009, the Stockholm Convention included Penta- and Octa-BDE in the list of persistent organic pollutants (POPs) that by definition are environmentally persistent, bioaccumulative, and toxic to humans and the environment, and subject to long-range transport (Stockholm Convention, 2009). In contrast, Deca-BDE remains a high-production volume flame retardant, although partial restrictions have been imposed on its use in Europe and in some US states, and manufacturers have agreed to discontinue production of Deca-BDE in the US as of 2013 (US EPA, 2010). Comparatively few data exist regarding the environmental distribution of highly brominated PBDEs, either present in or derived from Octa- and Deca-BDE

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formulations, in part, due to analytical challenges in the accurate quantification of BDE-209, the main constituent of Deca-BDE (Alcock et al., 2011, Covaci et al., 2007). These hydrophobic congeners bind to particles and are concentrated in soils, sewage sludge, and indoor dusts, especially near urban areas (Hale et al. 2006). BDE-209 is the dominant PBDE found in marine sediments (de Boer et al. 2003), thus, the deep oceans are vast reservoirs for ongoing inputs of Deca-BDE and its debromination products to marine ecosystems (Shaw and Kannan, 2009).

Despite regulations, large amounts of PBDEs are present in long-lived, in-service and discarded polymer products and electronics, creating outdoor reservoirs for future dispersal to the ocean environment for decades (Harrad and Diamond, 2006; Shaw and Kannan, 2009). In North America, where consumption of Penta-BDE dominated the global market for three decades (BSEF, 2003), PBDE levels in marine biota and humans are the highest in the world and are increasing (Costa et al., 2008; Hites, 2004; Shaw and Kannan, 2009; Shaw et al., 2010). HBCDs are still in worldwide use, but have been cited as priority action chemicals in Europe and Japan and are under evaluation as a POP by the Stockholm Convention (UNEP, 2010). In 2010, the European Commission banned HBCD use in polystyrene building insulation under its Registration, Evaluation, Authorization & Restriction of Chemicals (REACH) program (European Commission, http://ec.europa.eu/environment/chemicals/reach/reach_intro.htm). Nevertheless, the global demand for HBCDs is growing, and levels of the most persistent stereoisomer, α-HBCD, are increasing in marine food webs (Covaci et al., 2006; Stapleton et al., 2006a; Tanabe et al., 2008).

Exposure of wildlife and humans to PBDEs and HBCDs has been associated with endocrine disruption, immunotoxicity, and reproductive/developmental effects including developmental neurotoxicity (Birnbaum and Staskal, 2004; Costa and Giordano, 2007, 2011; Covaci et al., 2006; Shaw and Kannan, 2009). In young gray seals, PBDE burdens are predictive of reduced probability of first year survival (Hall et al., 2009), whereas thyroid hormone alterations have been reported in gray seals and harbor seals co-exposed to PBDEs and PCBs (Hall and Thomas, 2007; Hall et al., 2003). Recently, Frouin et al. (2010) demonstrated that environmentally relevant levels of PBDEs disrupt in vitro immune responses of harbor seal granulocytes and suggested that PBDEs may compromise innate immune responses of highly exposed wild seals.

PBDEs have been detected at relatively high concentrations in harbor seals (*Phoca vitulina concolor*), apex predators in the northwest Atlantic marine ecosystem (Shaw et al., 2008). Congeners found in harbor seals and their fish prey (tri- through Deca-BDEs) showed a "Penta-BDE" signature, but were suggestive of exposure to all three PBDE commercial mixtures (Shaw et al., 2008, 2009). The low BDE-209 levels (1–8 ng/g lw) detected in seal blubber and whole fish samples (Shaw et al., 2009) were similar to those retained in blubber of captive gray seals (*Halichoerus grypus*) exposed to Deca-BDE spiked food, suggesting that seals may accumulate this congener through feeding (Thomas et al., 2005). Whereas tetra- through hexa-BDE congeners were highly biomagnified from fish to seal blubber, BDE-209 did not biomagnify in blubber (Shaw et al., 2009). BDE-209 partitioning in marine mammal tissues is not understood, but studies in rats and fish have suggested that BDE-209 preferentially binds to blood proteins and migrates to perfused tissues, such as the liver (Huwe et al., 2008; Mörck et al., 2003; Stapleton et al., 2006b).

The aim of this study was to investigate the accumulation of PBDEs, including BDE-209 and other highly brominated congeners, in harbor seal liver samples (n = 56), and to compare concentrations and congener patterns in liver with those previously analyzed in blubber (Shaw et al., 2008). HBCDs were measured in all liver samples and also in blubber samples from a subset of seals (n = 11) to enable similar comparisons. Most of the animals investigated were pups, providing an opportunity to examine tissue distribution of PBDEs and HBCDs resulting from maternal transfer. To assess tissue differences in biomagnification of PBDEs and HBCD, biomagnification factors (BMFs) were calculated from marine fishes to adult male seal liver and compared with BMFs previously calculated from fish to seal blubber (Shaw et al., 2009). This is the first study to characterize tissue-specific biomagnification of BFRs in the northwest Atlantic marine food web.

2. Materials and methods

2.1. Samples

PBDEs and HBCDs were analyzed in liver samples collected from 56 harbor seals (6 adult males, 28 female pups, 22 male pups) that stranded along the northwest Atlantic coast between 2001 and 2006 (Figure SI-1). HBCDs were also analyzed in blubber samples collected from a subset of 11 seals (4 adult males, 6 female pups, 1 male pup). Seals were weighed, and standard length and axillary girth were measured. Age class was estimated based on body size. Samples were stored in a freezer at −20 °C until analysis.

2.2. Sample preparation and analysis

Seal liver samples (2–2.5 g) and blubber (0.2–0.3 g) were ground with sodium sulfate and spiked with internal standards (BDE 77, BDE 128, ^{13}C-BDE 209 and ^{13}C-HBCDs). Samples were extracted for 2 h by hot Soxhlet (Buchi) with a mixture of acetone/hexane (1/3, *v*/*v*). The extract was evaporated and cleaned by passing through 8 g of acid silica (H_2SO_4, 44%), from which pollutants were eluted with 20 ml hexane and 15 ml DCM (Covaci et al. 2008; Voorspoels et al., 2003). Minor adaptations were required as PBDEs were analyzed by GC-ECNI/MS or GC-EI/MS and HBCDs by LC–MS/MS. The cleaned extract was evaporated to dryness, redissolved in 0.5 ml hexane and eluted from pre-packed silica cartridges (500 mg, 3 ml, Varian) with 6 ml hexane (for GC analysis) and 6 ml DCM (for LC analysis). Both fractions were evaporated to dryness and redissolved in 100 μl *iso*-octane or methanol, respectively.

The analysis of PBDEs was performed by gas chromatography–mass spectrometry (GC–MS in electron capture negative ionization (ECNI) mode) (Agilent GC6890–MS5973) using a 15 m × 0.25 mm × 0.10 μm DB-5 capillary column (J&W Scientific). For confirmation of lower PBDEs, the extracts were injected into a GC/MS operated in electron ionization (EI) mode (Agilent GC6890–MS5973) and equipped with a 25 m × 0.22 mm × 0.25 μm HT-8 capillary column (SGE). The separation of α-, β-, and γ-HBCD isomers was achieved by liquid chromatography–tandem mass spectrometry (LC–MS/MS) in the electrospray negative ion mode (Agilent 6410 triple quadrupole) using a Zorbax Extend-C18 reversed phase analytical column (50 mm × 2.1 mm i.d., 3.5 μm particle size) (Agilent). Further details on the analysis of PBDEs and HBCDs are given in the Supplementary material section.

2.3. Quality assurance and quality control

QA/QC was performed through the analysis of procedural blanks, a replicate sample and a standard reference material (SRM 1945, PBDEs in whale blubber, which has also indicative values for HBCDs, NIST). For the replicate and SRM 1945, the relative standard deviations (RSD) were <10% for most analytes. Additionally, the method performance was assessed through successful participation to interlaboratory studies organized by NIST (PBDEs in marine mammals). Procedural blanks of PBDEs and HBCDs were consistent (RSD<20%) and therefore the mean value of each analyte in the procedural blanks was used for subtraction. Method quantification limits (LOQs) for individual PBDE congeners were based on procedural blanks (3× SD) and the amount of sample taken for analysis. LOQs for tri-hepta

PBDEs ranged between 1 and 2 ng/g lw. LOQs for Octa-BDEs (BDE-196, -197, -203) were 2 ng/g lw.

Due to higher instrumental LOQ for BDE-209, to its presence in the procedural blanks, to variable sample amounts taken into analysis and to different lipid contents of the samples, the method LOQ for BDE-209 was variable and ranged between 5 and 35 ng/g lw (mean 18.6 ng/g lw). Similar to other PBDE congeners, method LOQ was based on procedural blanks (3× SD) after mean blank subtraction. Special precaution was taken to ensure reliable results, e.g., avoiding presence of dust in the lab, avoiding UV degradation of BDE-209 through use of amber glass, use of ^{13}C-BDE-209 as internal standard. Even in these conditions, procedural blanks of BDE-209 had a higher SD than for other congeners, resulting in higher LOQ. LOQs were 1.5 ng/g lw for α-HBCD and 2 ng/g lw for β- and γ-HBCD.

An interlaboratory comparison was conducted to determine the comparability of PBDE concentrations measured by our laboratory in seal liver (n = 56) with those measured earlier by a different laboratory in blubber of 42 harbor seals (7 adult males, 8 adult females, 6 male yearlings, 8 female yearlings, 6 male pups, 7 female pups) (Shaw et al., 2008). Eleven (11) of the 42 blubber samples were re-analyzed for PBDEs (tri- to Deca-BDE congeners) and concentrations were compared with those previously measured in blubber from the same 11 individuals. Interlaboratory differences in blubber concentrations of ΣPBDEs (sum of 8 dominant congeners) were below 10% and thus considered negligible: mean ± standard deviation: 2047 ± 3470 and 1890 ± 3049 ng/g lw, and concentrations of the five most abundant PBDE congeners (BDE-47, -99, -100, -153, -154) and % lipids were strongly correlated ($R^2 > 0.80$) in the paired samples. Thus, the larger blubber dataset (n = 42) was used for comparisons with the liver dataset (n = 56) in this study.

2.4. Statistics

Values below the detection limit were replaced with 1/2 LOQ for calculation of means and totals. Statistical analyses were conducted with SPSS 15.0. Non-parametric statistics were used because most variables were not normally distributed and group sample sizes varied. Age-gender, tissue, and spatial differences in concentrations were explored with Kruskal–Wallis H tests and Mann–Whitney U tests. Temporal trends were examined with Spearman correlations.

3. Results and discussion

Of 19 PBDEs targeted in harbor seal liver samples (n = 56), 16 congeners were detected: BDEs-28, -47, -49, -85, -99, -100, -153, -154, -155, -181, -183, -184, an unidentified hepta-, -191, -197, and -209. BDEs-66, -196, and -203 were not detected. Of three HBCD isomers targeted, the α-HBCD isomer was predominant in liver and blubber. Isomers β- and γ-HBCD were detected only in 12 and 2 liver samples, respectively, and were not detected in blubber. Therefore, all statistical analyses focused on α-HBCD.

3.1. Concentrations of BFRs in harbor seal tissues

ΣPBDE (tri- to Octa-BDE) concentrations in seal liver samples ranged from 35 to 19,500 ng/g lipid weight, with an overall mean ± standard deviation of 2670 ± 3570 ng/g lw; n = 56. Although the samples were not paired, the levels in liver were similar to concentrations previously reported in blubber (Shaw et al., 2008). In contrast, α-HBCD levels were significantly higher in seal liver than in blubber (p = 0.04). Overall α-HBCD levels in liver and blubber were 38 ± 48 ng/g lw (range 2–279 ng/g lw, n = 55), and 12 ± 9.5 ng/g lw (range 2–29 ng/g lw, n = 10), respectively. Two outliers were detected: a male pup with a hepatic α-HBCD level of 11,600 ng/g lw and a female pup with a blubber level of 182 ng/g lw. These were excluded from the statistical analysis.

3.2. Influence of age and gender on tissue distribution

Mean ∑ PBDE concentrations in harbor seal liver by age/gender were (in descending order): 4400 ± 4870, 1680 ± 1810, and 969 ± 775 ng/g lw in male pups, female pups, and adult males, respectively, and differences among the three groups were significant (Kruskal–Wallis test: $X^2 = 9.3$, p = 0.01) (Fig. 1). Among the pups, males (n = 22) had two-fold higher hepatic ΣPBDE concentrations than females (n = 28, p = 0.01). The highest concentration in liver (19,500 ng/g lw) was observed in a male pup from southern Maine. In addition, male pups had significantly higher levels of BDE congeners -28, -47, -99, -100, -153, and -155 than female pups (p < 0.02). In contrast, mean blubber ΣPBDE concentrations in the female pups (6220 ± 9600 ng/g lw; n = 7) were an order of magnitude higher than those in male pups (639 ± 559 ng/g lw; n = 6) (Shaw et al., 2008). The difference was not significant, likely owing to the small number of pups and large variability in the earlier study. Compared with the adult male seals, male pups had significantly higher hepatic ΣPBDE concentrations (p = 0.02); levels of BDE congeners -47, -99, and -100 were also higher in male pups (p = 0.02) (Fig. 1).

Mean α-HBCD concentrations in liver were (in descending order): 48 ± 46, 31 ± 53, and 21 ± 14 ng/g lw in male pups, female pups, and adult males, respectively. Age-gender differences were marginally significant among the three groups ($X^2 = 5.3$, p = 0.07). Similar to the PBDE pattern, male pups had higher α-HBCD levels in liver than female pups (p = 0.03). In liver, mean α-HBCD levels in the male pups were two-fold higher than those in the adult males, although the difference was not significant. Because of small sample sizes, age-gender differences in blubber levels of α-HBCD could not be compared.

This accumulation pattern shows that the highest lifetime exposure to lipophilic BFRs results from placental and lactational transfer, possibly placing pups at risk for adverse effects during development. The first weeks of life represent a period of rapid

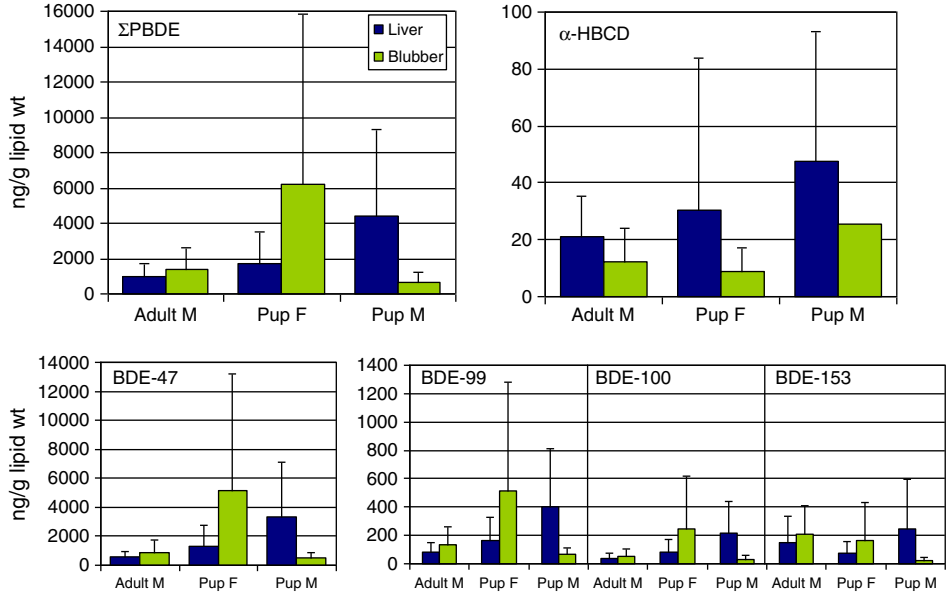

Fig. 1. Mean concentrations (ng/g lw) and standard deviations of ΣPBDE, individual PBDE congeners, and α-HBCD in harbor seal liver and blubber by age and gender. ΣPBDE in liver includes tri–octa-BDE congeners and ΣPBDE in blubber includes tri–hexa-BDE congeners.

growth, during which harbor seal pups almost triple their birth weight and lay down layers of blubber prior to weaning. Since exact ages of the pups were unknown, we used biometric data to examine the possible influence of growth on hepatic BFR concentrations. No significant correlations between concentrations of PBDEs or α-HBCD and body weight, body length, or lipid content of the samples were found for male or female pups (Figure SI-4). Nevertheless, these findings were interesting, and suggestive of possible gender differences in metabolism and elimination/retention of BFRs among young seals.

3.3. Congener and isomer composition of BFRs

Eight PBDE congeners (BDEs-28, -47, -49, -99, -100, -153, -154, and -155) contributed 81–99.9% of the total PBDE content in seal liver and 91–99.6% in blubber (Fig. 2). BDE-47 was the dominant congener, contributing 62–75% of the total in tissues. PBDE profiles in pup liver and blubber followed the order: BDE-47 > 99 > 100 > 153 > 154 > 155 > 28 > 49 and tissue distributions were similar with the exception that blubber of male pups contained a higher proportion of BDE-28 than liver ($p = 0.04$), and male pup liver contained a higher proportion of BDE-153 than blubber ($p = 0.007$). Between pups and adult males, congener profiles in liver differed significantly with respect to BDE-28 ($X^2 = 8.0$, $p = 0.018$), -153 ($X^2 = 8.8$, $p = 0.012$), and -155 ($X^2 = 13$, $p = 0.002$). BDE-28 contributed more to the total PBDE content in pup liver, whereas the hexa-BDE congeners -153 and -155 contributed proportionally more to the total in the adult males. Congener profiles in blubber were similar. These differences likely reflect the different exposure pathways between adults and pups, as well as age-related differences in the ability to metabolize and eliminate PBDE congeners. Whereas the profiles in the adult males reflect uptake and accumulation through feeding, the pattern in the pups suggests efficient placental and lactational transfer of BDE-28, BDE-47 and to a lesser degree, BDEs-99, and -100, but very limited transfer of the hexa-BDE congeners. In gray seals, maternal transfer efficiency was shown to decline with increasing degree of bromination, as a function of increasing K_{ow} values (Ikonomou and Addison, 2008). This may be a consequence of molecular size of higher PBDE congeners, which may limit diffusion and lipid/water partitioning in females during lactation.

Although commercial HBCD mixtures consist mainly of γ-HBCD (75–89%), α-HBCD (10–13%), and β-HBCD (1–12%) (Heeb et al., 2005), stereoisomeric profiles of HBCDs in marine biota are generally dominated by α-HBCD (Covaci et al., 2006). In this study, the α-HBCD isomer contributed 95% and 100% of the total HBCD content in liver and blubber, respectively. β- and γ-HBCDs were detected in 21% and 3% of the liver samples, respectively, and were not detected in blubber.

3.4. Global comparisons

PBDE concentrations in liver of the harbor seals were relatively high on a global scale, reflecting the extensive use of Penta-BDE in the US (Shaw and Kannan, 2009) (Figure SI-2). Hepatic PBDE concentrations in the male pups (4400 ng/g lw) were higher than those reported in liver of seals from Europe (Boon et al., 2002; Routti et al., 2009) and Canada (Wolkers et al., 2006), and sea otters from California (Kannan et al., 2007, 2008), and orders of magnitude higher than levels reported in cetacean species from Europe (Pettersson et al., 2004; Weijs et al., 2010), Asia (Moon et al., 2010), and Brazil (Dorneles et al., 2010).

Extremely high HBCD levels (600 to 4700 ng/g lw) have been reported in cetaceans from Europe and the Asia-Pacific region (Isobe et al., 2009; Law et al., 2008; Morris et al., 2004; Zegers et al., 2005), reflecting the relatively high consumption of HBCD in Europe and Asia compared with North America (Covaci et al., 2006). In comparison, HBCD concentrations in northwest Atlantic harbor seals were orders of magnitude lower (mean range 9 to 48 ng/g lw) (Figure SI-3a,b). These differences may also reflect species-specific differences in the ability to metabolize HBCD between pinnipeds and cetaceans, as was observed for PBDEs (Weijs et al., 2009a,b). Hepatic HBCD concentrations in our male harbor seal pups (48 ng/g lw) were an order of magnitude lower than liver concentrations reported in finless porpoises and striped dolphins from the Japanese coast (Isobe et al., 2009, 2011) and slightly higher than those reported in liver of Atlantic white-sided dolphins (Peck et al., 2008). Blubber concentrations in the male pups (25 ng/g lw) and adult males (12 ng/g lw) were comparable to those

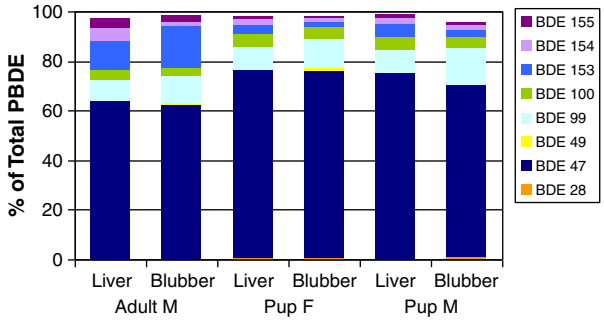

Fig. 2. Profiles of the eight predominant PBDE congeners in harbor seal liver and blubber by age class.

reported in blubber of harbor seals and polar bears from Norway (Jenssen et al., 2007), northern fur seals from Japan (Kajiwara et al., 2006), and ringed seals from Greenland (Letcher et al., 2009). Within the US, HBCD levels in male pup blubber were higher than those reported in California sea lions (Stapleton et al., 2006a) and bottlenose dolphins from Florida (Johnson-Restrepo et al., 2008), but five times lower than the levels in Atlantic white-sided dolphins (Peck et al., 2008) (Figure SI-3a).

3.5. Spatial and temporal trends

Considered relatively non-migratory, northwest Atlantic harbor seals nevertheless make seasonal movements along the coast from New Jersey to Maine (NMFS, 2007), following an urban/industrial–rural–remote gradient from south to north. However, no significant differences were found for hepatic ΣPBDE or α-HBCD concentrations in adult males or male/female pups that stranded in the industrialized southern area compared with those in the north (Mann–Whitney U tests, all $p > 0.10$). A similar lack of a spatial trend was previously reported for PBDEs in seal blubber samples (Shaw et al., 2008), and probably reflects the integrated exposure of adult seals to prey fish contaminated by diffuse local sources (e.g., wastewater treatment plants, sewage sludge applications, landfill leachate) across the harbor seal range.

To assess possible temporal trends of contamination by PBDEs and HBCD, concentrations in harbor seal pup liver samples collected between 2001 and 2006 were plotted against the year of sampling (Figure SI-5). Results show a lack of a temporal trend for ΣPBDE concentrations in male or female pups from 2001 to 2006, which is consistent with our earlier findings in blubber (Shaw et al., 2008) and with the lack of a trend observed in other studies over the same time period (Stapleton et al., 2006a; Tuerk et al., 2005). Collectively, the data suggest that PBDE levels were increasing in marine mammals between the 1970s and the mid-1990s (Shaw and Kannan; 2009; Tanabe et al., 2008), but may have stabilized or reached equilibrium over the past decade.

Similarly, our results show a lack of a temporal trend for α-HBCD levels in harbor seal pup liver samples between 2001 and 2006. HBCDs steadily increased in northern fur seals from Japan between 1980 and 2000 (Kajiwara et al., 2004, 2006) and in striped dolphins from Japan between 1992 and 2000 (Isobe et al., 2009). An exponential increase in HBCDs was reported in male California sea lions between 1993 and 2003 (Stapleton et al., 2006a), suggesting that consumption may be growing in the US, as HBCDs are being marketed as replacement flame retardants for PBDEs that have been banned or phased out (Shaw and Kannan, 2009; Shaw et al., 2010).

3.6. Highly brominated PBDE congeners

Several hepta- and Octa-BDE congeners were detected at low levels in seal liver including BDE-183, -197, and an unidentified hepta-BDE (Table 1). BDEs- 181, -184, and -191 were detected at trace levels in liver. Hepta- and Octa-BDE congeners were previously found in seal blubber samples at slightly higher concentrations (Shaw et al., 2008). While the presence of tetra- to hexa-BDE congeners in seal tissues indicates exposure to components of the Penta-BDE mixture, the occurrence of hepta- and Octa-BDE congeners suggests recent exposure to the Octa- and Deca-BDE commercial mixtures and/or BDE-209 debromination processes (Stapleton et al., 2006b).

Because of its large size (MW 959), BDE-209 appears to have limited absorption and to penetrate cells with difficulty. For example, BDE-209 cannot be absorbed through the intestinal tract by passive diffusion (Mörck et al., 2003); rather, its absorption into tissue may be facilitated by carrier proteins such as P-glycoprotein (Charman, 2000). The data suggest that oral absorption is 10–25% at most, and it is rapidly excreted through the feces, with a short half-life in serum (Thomas et al., 2005; Thuresson et al., 2006). In captive gray seals fed Deca-BDE spiked food, the half-life of BDE-209 in serum was 8.5–13 days, but once this congener migrated to blubber, it was retained in the body for months (Thomas et al., 2005). In the present study, BDE-209 concentrations ranging from 14 to 40 ng/g lw were detected in harbor seal liver samples. It should be noted that detection frequency for BDE-209 in harbor seal liver was very low (<10%), and the LOQ varied from 5 to 35 ng/g lw, thus the data herein are only suggestive. However, the BDE-209 concentrations in seal liver samples were up to five times higher than those previously found in blubber (Shaw et al., 2008), implying that liver may be a preferential tissue for BDE-209 retention in this species. Tlhis observation is consistent with results of laboratory studies showing preferential accumulation of BDE-209 in liver of rats and fish (Huwe et al., 2008; Mörck et al., 2003; Stapleton et al., 2006b), and with field studies showing selective hepatic retention of this compound in terrestrial wildlife, such as red foxes (Voorspoels et al., 2006) and Japanese raccoon dogs (Kunisue et al., 2008).

3.7. Biomagnification of PBDEs and HBCD from fish to seals

PBDEs and HBCDs are often elevated in species at the top of food webs, which clearly points to biomagnification. However, only a few studies have specifically examined the transfer of BFRs through aquatic/marine food chains. In this study, calculation of BMFs from fish to liver of the adult male harbor seals revealed a high biomagnification potential for Σtri- to hexa-BDE congeners (average BMFs 14–54), which was similar to that previously reported from fish to harbor seal blubber (BMFs 17 to 76) (Shaw et al., 2009) (Figure SI-6). Comparable BMFs for ∑PBDEs were reported between fishes and harbor seals in the North Sea (27 to 38) (Boon et al., 2002), between polar cod and harbor seals (12.4) and ringed seals (36.9) in Svalbard, Norway (Jenssen et al., 2007; Sørmo et al., 2006), and between teleost fishes and bottlenose dolphins in a

Table 1
Concentrations (range, ng/g lw) of hepta- to deca-BDEs in whole fish tissue (Shaw et al., 2009) and harbor seal blubber (Shaw et al., 2008) and liver (this study).

	Plaice	Mackerel	White hake	Seal liver	Seal blubber
	Min–max (detection frequency) ng/g lipid wt				
BDE-181	ND	ND–1.7 (25%)	ND	ND–6.8 (9%)	
BDE-183	0.13	0.08–31 (100%)	ND	ND–13 (71%)	1.7–45 (100%)
BDE-184				ND–3.9 (9%)	
Unknown hepta-				ND–17 (43%)	ND–15 (50%)
BDE-191				ND–1.7 (9%)	
BDE-197	ND	0.06–22 (100%)	ND	ND–4.7 (14%)	ND–57 (58%)
Unknown octa-					ND–84 (25%)
BDE-203	ND	0.01–5.7 (100%)	ND	ND	
BDE-207	ND	0.11–39 (100%)	ND		
BDE-209	1.8	0.21–3.9 (100%)	ND–1.5 (50%)	ND–40 (7%)	ND–8 (25%)

ND = not detected.

Florida marine food web (range 3–85) (Johnson-Restrepo et al., 2005). Slightly lower BMFs (range 0.6 to 15) were reported for tri- through hexa-BDE congeners between prey fishes and harbor seals in the southern North Sea (Weijs et al., 2009b).

BDE-209 levels detected in harbor seal livers were up to 10-fold higher than those in their prey fish, which contrasts with the pattern from fish to blubber (Shaw et al., 2009) and suggests that the biomagnification of BDE-209 may be tissue-specific. However, further studies are needed to confirm this observation given the low detection frequency and relatively high LOQ for BDE-209 in these samples. BDE-209 is the dominant PBDE found in marine sediments (de Boer et al., 2003) and the discharge of Deca-BDE to aquatic and marine ecosystems has increased in recent years (Shaw and Kannan, 2009), thus the potential for BDE-209 to biomagnify in marine food webs is of concern. Moreover, most of the seals in this study were pups, implying that BDE-209 is subject to placental and/or lactational transfer to some extent.

Only a few studies have investigated the transfer of HBCD through the marine food chain (Covaci et al., 2006; Jenssen et al.; 2007; Sørmo et al., 2006; Tomy et al., 2009). In the present study, BMFs calculated for the transfer of α-HBCD from three prey fish species (alewife, Atlantic herring, and Atlantic mackerel) to tissues of adult male harbor seals were 3.0, 1.0, and 1.6 respectively for liver, and 1.6, 0.54, and 0.89 respectively for blubber. A similar low biomagnification of HBCD (BMFs 1.2 to 2) was reported from cod to blubber of Norwegian harbor seals (Jenssen et al., 2007), whereas a higher BMF of 11 was reported for HBCD from polar cod to ringed seal blubber (Sørmo et al., 2006), suggesting that harbor seals may possess a species-specific ability to metabolize α-HBCD.

4. Conclusions

This is the first study to characterize tissue-specific biomagnification of BFRs in top marine predators inhabiting the northwest Atlantic region. Levels of the less brominated PBDEs (tri- through hexa-BDE congeners) in harbor seal liver samples were similar to those previously found in blubber and were highly biomagnified from fish to both tissues in the seals. In contrast, α-HBCD levels were significantly higher in liver than in blubber and did not biomagnify in either tissue, confirming reports that harbor seals may possess the ability to metabolize this isomer. The large differences observed in the retention of PBDEs and α-HBCD in tissues of male and female pups was a surprising finding that warrants further investigation.

Although detection frequency for BDE-209 was low, our finding up to five-fold higher levels of this congener in harbor seal liver than in blubber is consistent with the preferential hepatic retention of BDE-209 observed in other species. In contrast to the lack of BDE-209 biomagnification observed in blubber, BDE-209 levels in seal livers were up to ten-fold higher than those in their prey fish, suggesting that the accumulation/biomagnification of BDE-209 in marine food webs is tissue-specific. The toxicological implications of low-level maternal transfer of BDE-209 to young seals are unclear, but the possible contribution of this congener, via metabolic debromination, to the burden of less brominated, more toxic PBDE congeners is of concern.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at doi:10.1016/j.envint.2012.01.001.

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