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Brominated flame retardants and Dechloranes in European and American eels from glass to silver life stages

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Abstract

The populations of American (*Anguilla rostrata*) and European eels (*Anguilla anguilla*) have been declining rapidly in the last decades. Organic contaminants are suspected to be one of the possible causes for the decline; however, so far there have been few investigations of the uptake of specific compounds by different life cycle stages (e.g. freshwater or marine stage) and how the contamination patterns develop throughout the eel's life cycle. In the present study we measured concentrations of polybrominated diphenylethers (PBDEs), alternate brominated flame retardants (alternate BFRs) and Dechloranes (Decs) in different life stages of European and American eels to compare the contamination patterns and their development throughout the eel's life cycle.

In general, concentrations of flame retardants (FRs) were similar to or higher in American than in European eels, and a greater number of FRs were detected. PBDE congeners that are characteristic of the Penta-PBDE formulation were the most abundant FRs in all adult eels as well as American glass eels. In European glass eels the alternate BFR 2,3-dibromopropyl-2,4,6-tribromophenylether (DPTE) and Dechlorane Plus were the dominating FRs, with average concentrations of 1.1 ± 0.31 ng g^{-1} ww and up to 0.32 ng g^{-1} ww respectively. Of the PBDEs BDE-183 was the most abundant congener in European glass eels. Low concentrations (less than 10 % of the total contamination) of Tetra and Penta-PBDEs in juvenile European eels indicated that bans of technical Penta-PBDE in the European Union are effective. Enrichment of PBDEs was observed over the life stages of both European and American eels. However, a greater relative contribution of PBDEs to the sum FR contamination in American eels indicated an on-going exposure to these substances. High contributions of alternate BFRs in juvenile eels indicated an increased use of these substances in recent years. Concentrations seemed to be driven primarily by location, rather than life stage or age.

Keywords

European eel, American eel, Brominated Flame Retardants, PBDEs, alternate BFRs, Dechloranes, Bioaccumulation, Life Cycle

1. Introduction

European eel (*Anguilla anguilla*) and American eel (*Anguilla rostrata*) are facultatively catadromous, carnivorous, and, during their continental phase, benthic species with unusual life cycles (Dekker 2000, Ministry of Natural Resources 2007, van Ginneken et al. 2005). Both spawn in the Sargasso Sea, hatch, and are transported as larvae by oceanic currents to the North African, European and American coastal waters (Dekker 2000, Ministry of Natural Resources 2007). There they first metamorphose into glass eels and develop further to elvers and yellow eels. During their continental growth phase, eels build up large energy resources (Belpaire & Goemans 2007, Belpaire et al. 2009). Prior to maturation and migration back to their spawning grounds, eels undergo a silvering process accompanied by drastic changes in physiology including the degeneration of the alimentary tract (Durif et al. 2005). Stored fat is used to develop gonads and as energy reserves for their migration back to the Sargasso Sea to reproduce once and die (Dekker 2000).

The European eel is of high economic value. However, its population has been declining rapidly since the 1980s (ICES 2008, Fisheries Forum 2003) leading to its listing under Appendix II of CITES (the Convention on International Trade in Endangered Species of Wild Fauna and Flora) as well as on the Red List of species (IUCN), rating it as "critically endangered". A similar downward trend in American eel has led to the closure of commercial yellow eel fishery in Lake Ontario in 2004 and the rating “threatened” in Canada by COSEWIC (Committee on the Status of Endangered Wildlife in Canada) in 2012.

Chemical contaminants are postulated as one of the possible causes for the decline of freshwater eel populations because, due to their high lipid contents (Palstra et al. 2006, Belpaire & Goemans 2007, Belpaire et al. 2009), eels are predestined to accumulate potentially harmful lipophilic organic pollutants.

Halogenated flame retardants (HFRs) are a group of possibly harmful and accumulating organic contaminants. They are used in a variety of consumer products such as textiles, electronic equipment, plastics, and furniture (de Wit et al. 2002). The largest group among the currently used HFRs are brominated flame retardants (BFRs). For several decades polybrominated diphenyl ethers (PBDEs) were the most widely used additive BFRs (de Wit et al. 2002). However, due to their adverse effects on the environment and human health, PBDEs have been banned for production and usage in the European Union (EU) (European Court of Justice 2008), and are being voluntarily withdrawn or phased out in North America (US EPA 2009). Congeners used in the technical penta- and Octa-PBDE mixtures have been classified as

Persistent Organic Pollutants (POPs) under the Stockholm Convention (SCOP 2009).

Government regulations require consumer products to meet certain standards for flame retardancy, which has encouraged the use of substitutes such as alternate (non-PBDE) flame retardants both brominated or chlorinated such as Dechloranes (Decs) (Covaci et al. 2011). There is little knowledge concerning production, usage, or the persistence potential of these substitutes for PBDEs, yet many are suspected to at least partially fulfil the criteria for POPs (Harju et al. 2009, Sverko et al. 2011, Covaci et al. 2011).

This paper presents a comparison of concentrations and contamination patterns of PBDEs, alternate BFRs, and Decs throughout the life cycle of European and American eels. The aim was to identify the decisive factors for spatial and life cycle dependent distribution of halogenated flame retardants.

2. Material and Methods

2.1. Samples

The life stages examined were glass eels, elvers, yellow and silver eels for European eels, and glass eels, young yellow eels, yellow eels, silver eels for American eels.

One hundred European glass eels, originally caught at the French Atlantic coast, were purchased from a glass eel distributer and combined into ten samples. Data for elvers and adult European eels from the Elbe and Rhine River in Germany were previously published in Sühring et al. (2013). Thirty-seven American glass eels from Baie des Sables, Matane, Quebec, Canada were pooled into three samples. Ten young American yellow eel samples were taken from the Saint Lawrence River, Canada at each of the Beauharnois Dam, Quebec and the Moses-Saunders Dam, Ontario. Fifteen muscle tissue samples were taken from older yellow eels sampled from Lake Ontario and the upper Saint Lawrence River; dorsal muscle tissue was excised posterior to the anus. Data for American silver eels from Lake Ontario were previously published in Byer et al. 2013a,b.

The primary sampling areas (Lake Ontario/ Saint Lawrence River, Canada and Elbe River, Germany) are both major waterways in industrialised areas with major urban areas such as Toronto and Hamilton (Lake Ontario), and Dresden and Hamburg (River Elbe). Including the estuary both the Elbe and the Saint Lawrence River are over 1000 km in length (Netzband et al. 2002, Canadian Geographic 2008). However, the Saint Lawrence River is downstream the Laurentian Great Lakes, and therefore, potentially receives contaminants from a large geographic area, while the river Elbe originates from a spring in the Riesengebirge. Another major difference is

the average discharge of the rivers with over 16 000 m^3 s^{-1} for the Saint Lawrence River and ~860 m^3 s^{-1} for the Elbe River (Netzband et al. 2002, Environment Canada 2009).

A detailed list of the analysed samples can be found in Table S1.

2.2. Extraction and clean-up

The frozen yellow eel samples were homogenised with anhydrous Na_2SO_4 (Merck) (2:1; w/w) for approximately 20 min. using a stainless steel/glass 1 L laboratory blender (neoLab Rotorblender). For glass eel samples, 28 x 60 mm glass-fibre extraction thimbles for Soxhlet extraction were filled with Na_2SO_4- eel -mixture (equal to 3 g eel tissue). All samples were spiked with mass labelled surrogate standards ^{13}C-HBB, ^{13}C-BDE-77, ^{13}C-BDE-138, and ^{13}C-synDP.

Glass eel samples were Soxhlet-extracted using DCM at 55°C for 24h. Adult eels were extracted with DCM by accelerated solvent extraction, using the method described in Sühring et al. 2013. The lipid content of samples was determined gravimetrically from separate sample aliquots. Extracts were purified as described by Sühring et al. 2013. Briefly, a gel permeation chromatography (GPC) was used as first clean-up step, using 30 g Bio Beads SX-3 and DCM:hexane (1:1; v:v) as eluent. The first fraction (75 mL) was used to determine the lipid content of the sample; the second fraction (110 mL) contained the target substances and was reduced in volume to about 2 mL. 2.5 g 10 % H_2O deactivated silica gel was used as a second clean-up step. Analytes were eluted with 20 mL hexane and the volume reduced to 150 µL under a gentle stream of nitrogen. Finally, 500 pg (absolute) ^{13}C PCB-208 was added as an injection standard to each sample.

2.3. Instrumental Analysis

Extracts were analysed by gas chromatography/mass spectrometry (GC/MS; 6890 GC/5973 MSD) in negative chemical ionisation mode (NCI) with a method developed by Möller et al. (2010). Eels were analysed for nine PBDEs (BDE–28, –47, -66, -85, –99, –100, –153, –154, –183), 10 alternate BFRs (PBBz, PBT, DPTE, HBB, PBEB, TBB, BTBPE, TBPH, OBIND, HCDBCO), DP, aCl11DP, aCl10DP, 1,5-DPMA and Dechlorane 602, 603 and 604. A detailed list of standards can be found in Table S 2.

Peak areas of the obtained chromatograms were integrated using Agilent Technologies MassHunter Workstation Software Quantitative Analysis B.05.02 for GCMS. Further data analysis was performed with Microsoft Office Excel 2010 and Origin Lab 9.0 SR1.

2.4. QA/QC

Extraction and clean-up of juvenile European and American eels (glass eels and young yellow eels) were conducted in a clean lab (class 10000). Adult American eels (yellow eels) were extracted in a regular laboratory. Materials containing FR were avoided during sample preparation and analysis.

Surrogate recoveries were determined for every sample. Mean recoveries were 58 ± 18% for ^{13}C-HBB, 130 ± 20% for ^{13}C-BDE-77, 117 ± 22% for ^{13}C-BDE-138, and 78 ± 23% for ^{13}C-DP. All concentrations were recovery corrected.

A blank test, using Na_2SO_4 treated similar to real samples, was conducted with every extraction batch (five samples). Concentrations of FR in blanks processed in the clean lab were in general low; PBT, BDE-99 and BDE-183 were measured in one blank samples each at concentrations of 0.002 ng g^{-1} wet weight (ww), 0.0016 ng g^{-1} ww and 0.078 ng g^{-1} ww respectively. BDE-47 was detected in two blank samples at 0.088 ng g^{-1} ww and 0.24 ng g^{-1} ww. DPTE was detected in the majority of blank samples with average concentrations of 0.19 ± 0.036 ng g^{-1} ww. Samples processed at the regular laboratory showed greater contamination by technical Penta-PBDE and Octa-PBDE, with average concentrations between 0.12 ± 0.011 ng g^{-1} ww for BDE-66 and 1.75 ± 0.76 ng g^{-1} ww for BDE-47. Of the alternate BFRs, PBT, PBEB, and HBB were detected at average concentrations of 0.45 ± 0.12 ng g^{-1} ww, 0.075 ± 0.014 ng g^{-1} ww and 0.12 ± 0.0069 ng g^{-1} ww, respectively. DPTE was found in one blank sample at 0.12 ng g^{-1} ww. SynDP and antiDP were found in two and three blank samples with concentrations up to 0.14 ng g^{-1} ww and 0.21 ng g^{-1} ww, respectively. Blank concentrations were considered in the calculation of the sample concentrations and limit of detection (LOD) of the appropriate batch. In case of high blank values and detection frequencies, as e.g. in the case of DPTE, only samples with concentrations at least one order of magnitude higher than the average blank were considered in order to ascertain that concentrations found in the samples were environmental concentrations and not caused by contamination in the lab. The average blank value was then subtracted from the concentration found in the samples (see supplement information Tables S 3 and 4 for a detailed list of blank values, LOD and LOQ).

The limit of detection (LOD) was calculated from a signal to noise ratio of three or by using the blank standard deviation method (where applicable). The limit of quantification (LOQ) was calculated from a signal-to-noise ratio of ten or using the blank standard deviation method (where applicable). For juvenile eels, LODs ranged from 0.0022 ng g^{-1} ww for BDE-66 to 0.45 ng g^{-1} ww for BDE-47. For adult American eels, LODs ranged from 0.005 ng^{-1} g ww for BDE-153 to 4.03 ng g^{-1} ww for BDE-47

due to the higher average blank levels. The LOQ for juvenile European and American eels (glass eels and young yellow eels) ranged from 0.0073 ng g^{-1} ww for BDE-66 to 1.51 ng g^{-1} ww for BDE-47. The LOQ for large American yellow eels ranged from 0.017 ng^{-1} g ww for BDE-153 to 13.45 ng g^{-1} ww for BDE-47. Due to the high blank levels, BDE-47 results for American yellow eels were considered semi-quantitative.

3. Results and Discussion

Results for European yellow eels and silver eels were previously published in Sühring et al. 2013. Results for American silver eels were published by Byer et al. 2013a,b. The average results for PBDEs, alternate BFRs, and Dechloranes from this study are compared to recent studies in Table 1. A detailed list of all results is provided in supplement information Tables S 4 and 5.

		Glass eels (Estuary, FR)	Elvers (Vidå, GER)	Yellow Eels (Elbe, GER)	Silver Eels (Elbe, Rhine, GER)	Glass eels (Estuary, CA)	Young Yellow Eels (LO, SLR, CA)	Yellow eels (LO, CA)	Yellow eels (SLR, CA)	Silver Eels (LO, CA)	Silver Eels (LO, CA)
∑PBDEs	ng g^{-1} ww	1.8 ± 0.89	0.22 ± 0.08	8.9 ± 3.4	14.9 ± 11.9	1.7 ± 0.85	4.4 ± 2.7	16 *	5 *	26.7 ± 21.4	n.a.
	ng g^{-1} lw	176.0 ± 98.1	10.2 ± 1.3	33.5 ± 13.0	59.7 ± 47.7	168.8 ± 85	44 ± 27	77 *	23*	n.a.	
BDE-47	ng g^{-1} ww	<LOD	<LOD - 0.088	6.0 ± 2.2	10.06 ± 7.8	1.1 ± 0.55	2.1 ± 1.8	11 *	4 *	15.3 ± 14.3	n.a.
	ng g^{-1} lw		<LOD - 6.5	22.5 ± 8.3	40.2 ± 31.3	114.2 ± 55	21 ± 18	53 *	18 *	n.a.	
ATE	ng g^{-1} ww	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
	ng g^{-1} lw										226 ± 223 pg g^{-1} lw
BEHTBP	ng g^{-1} ww	<LOD	0.10 ± 0.032	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	n.a.	n.a.
	ng g^{-1} lw		7.4 ± 2.4								25.7 ± 26.3 pg g^{-1} lw
DPTE	ng g^{-1} ww	2.0 ± 0.31	0.22 ± 0.08	0.19 ± 0.18	0.74 ± 0.68	<LOD - 0.76	<LOD - 0.76	2.0 ± 0.78	1.4 ± 0.54	n.a.	n.a.
	ng g^{-1} lw	199 ± 31	16.06 ± 5.7	0.67 ± 0.30	3.0 ± 2.7	<LOD - 76	<LOD - 7.6	9.5 ± 3.7	6.7 ± 2.6		
HBB	ng g^{-1} ww	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	< LOD	n.a.	n.a.
	ng g^{-1} lw										3.72 ± 4.06 pg g^{-1} lw
PBEB	ng g^{-1} ww	< LOD	< LOD	0.020 ± 0.010	0.022 ± 0.014	<LOD - 0.027	<LOD - 0.020	< LOD	< LOD	n.a.	n.a.
	ng g^{-1} lw			0.28 ± 0.19	0.086 ± 0.057	<LOD - 2.7	<LOD - 0.20				
PBT	ng g^{-1} ww	0.012 ± 0.0013	< LOD	< LOD	< LOD	0.023 - 0.19	0.027 ± 0.014	< LOD	<LOD - 0.12	n.a.	n.a.
	ng g^{-1} lw	1.2 ± 0.13				2.3 - 19	0.27 ± 0.14		<LOD - 0.57		0.91 ± 1.09 pg g^{-1} lw
∑DP	ng g^{-1} ww	<LOD - 0.32	<LOD - 0.46	0.041 ± 0.027	0.043 ± 0.048	< LOD	0.17 ± 0.092	0.19 ± 0.086	0.29 ± 0.20	n.a.	n.a.
	ng g^{-1} lw	<LOD - 31.6	<LOD - 33.8	0.14 ± 0.065	0.17 ± 0.19		1.7 ± 0.92	0.90 ± 0.41	1.4 ± 0.95		66.9 ± 48.1 pg g^{-1} lw
DPMA	ng g^{-1} ww	< LOD	< LOD	< LOD	< LOD	< LOD	<LOD - 0.037	0.070 ± 0.019	0.10 ± 0.016	n.a.	n.a.
	ng g^{-1} lw						<LOD - 0.37	0.33 ± 0.090	0.48 ± 0.076		0.37 ± 0.57 pg g^{-1} lw
Dec-602	ng g^{-1} ww	< LOD	<LOD - 0.66	<LOD - 0.25	0.044 ± 0.048	< LOD	0.0070 - 0.29	2.7 - 1.2	0.26 - 2.4	n.a.	n.a.
	ng g^{-1} lw		<LOD - 48.8	<LOD - 0.73	0.18 ± 0.19		0.070 - 2.9	12.9 - 5.7	1.2 - 11.4		882 ± 515 pg g^{-1} lw
Dec-603	ng g^{-1} ww	< LOD	< LOD	< LOD	<LOD - 0.076	< LOD	<LOD - 0.020	0.14 ± 0.015	0.12 ± 0.067	n.a.	n.a.
	ng g^{-1} lw				<LOD - 0.37		<LOD - 0.20	0.67 ± 0.071	0.57 ± 0.32		12.4 ± 5.08 pg g^{-1} lw
f(syn)		0.94 ± 0.08	0.80 ± 0.14	0.97 ± 0.11	0.40 ± 0.09	n.a.	0.71 ± 0.34	0.64 ± 0.21	0.89 ± 0.17	n.a.	0.44
Average lipid content	%	1	1.4	27	25	1	10	21	23	20	20
Ref.		this study	Sühring et al 2013	Sühring et al 2013	Sühring et al. 2013	this study	this study	this study	this study	Byer et al. 2013b	Byer et al. 2013a

***data on BDE-47 in American yellow eels is semi-quantitative**

Table 1: Comparison of the mean (± SD) flame retardant concentrations [ng g^{-1} ww], [ng g^{-1} lw] and contribution of synDP to Σ DP (fsyn) found in European glass eels from France (FR), American glass eels from Canada (CA), young American yellow eels and yellow eels from Lake Ontario (LO) and the Saint Lawrence River (SLR) in Canada from this study with concentrations [ng g^{-1} ww], [ng g^{-1} lw], [pg g^{-1} lw] reported in recent studies on European elvers from the river Vidå at the German- Danish border (GER), yellow eels from the river Elbe in Germany, as well as European and American silver eels from the river Rhine, Elbe (Germany) and from Lake Ontario and the Saint Lawrence River, respectively

3.1. PBDEs

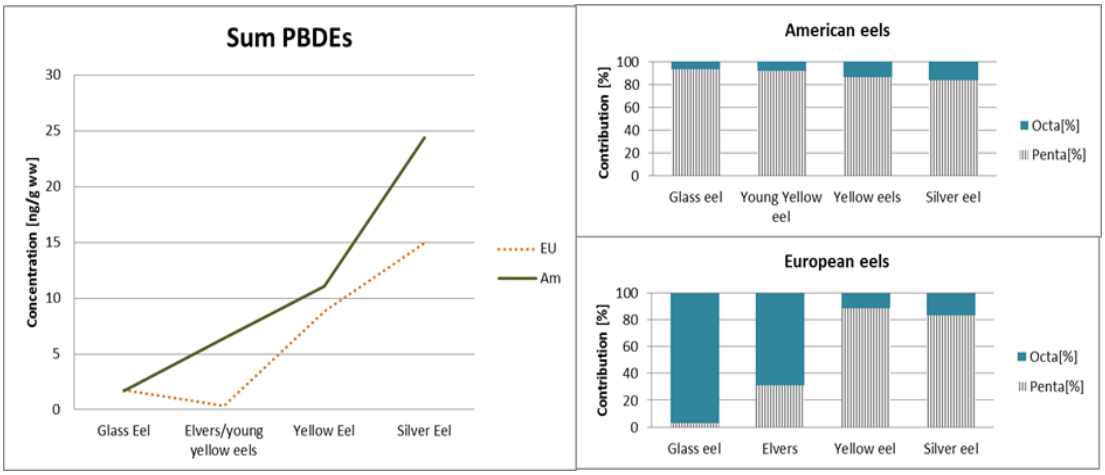

Figure 1: Concentration [ng g^{-1} ww] of Sum PBDEs in American and European eels throughout their life cycle stages (left) and contribution [%] of technical Penta- and OctaBDE(right)

The sum concentrations of PBDEs were similar in European and American glass eels (1.8 ± 0.89 ng g^{-1} ww and 1.7 ± 0.84 ng g^{-1} ww, respectively) yet more congeners were detected in American eels (Table 1).

The concentrations of congeners attributed to the technical Penta-PBDE mixture (BDE-47, BDE-99, BDE-100 and low amounts of BDE-153 and-154) were noticeably lower in European compared to American glass eels. More than 90% of PBDE in American glass eels was comprised of a technical Penta-PBDE mixture (Figure 1). In contrast 97% of the PBDE contamination in European glass eels consisted of BDE-183 and BDE-153, which are congeners of the technical Octa-PBDE mixture. The presence of technical Octa-PBDE in European glass eels has two possible explanations: The detected concentrations could indicate an on-going exposure to technical Octa-PBDE despite the restrictions. It could, however, also indicate an exposure to technical Deca-PBDE and subsequent debromination to lower brominated PBDE congeners as described by Eljarrat et al. 2011.

The difference in the congener pattern between American and European glass eels exhibits a fundamental difference between the contamination glass eels are exposed to in the European and American coastal environments. The low Penta-PBDE concentrations in European glass eels might indicate that restrictions on importation and use of technical Penta-PBDE in the European Union are having an effect on environmental inputs. The continued application of technical Deca-PBDE, on the other hand, could be the reason for the high contribution of its debromination product BDE-183 The high contribution of technical Penta-PBDE in American glass eels

reflects its historically higher use in North America compared to the EU (7100 T/a in North American vs 150 T/a in the EU in 2001 (www.bsef.com)), but can also be an indication for continued emissions. This would be congruent with the findings of Csiszar et al. (2013), who estimated Penta- and Octa-PBDE (BDE-28, -47, -100, -154, -183) emissions into the air of Toronto in 2008 to be 18 kg y^{-1}. They concluded, that, despite the restrictions, many buildings, homes and vehicles were still equipped with Penta- and Octa-PBDE containing materials, making them possible contamination sources (Csiszar et al. 2013). Higher current as well as historical emissions along with the persistence of PBDEs lead to generally higher concentrations in the aquatic environment in North America (US EPA 2010). However, the up to 16 x lower PBDE concentrations in both European and American glass eels compared to the other life stages indicate that the primary uptake of PBDEs occurs in the later life stages. The uptake of PBDEs is therefore probably driven by ingestion or dermal uptake due to contact with sediments, because eels become more predatory with size (before they stop feeding in their silver stage) and become benthic during their yellow eel stage (Tesch 2003, p.152). The primarily pelagic glass eels (Tesch 2003, p.122) are therefore mostly exposed to contamination through water, plankton, suspended matter or maternal transfer.

Technical Penta-PBDE was the predominant analysed flame retardant in European and American yellow and silver eels, contributing 89-92% and 86-91% of PBDEs, respectively (Figure 1), reflecting its persistence in the environment and biota. In general, the congener profile followed distributions reported in previous studies (Belpaire 2008) with an order of abundance of BDE-47 > BDE-100 > BDE-153 > BDE-99 > BDE-154 > BDE-183. High concentrations of BDE-47 were expected due to its high uptake rate and biomagnification within the aquatic food web (Domínguez et al., 2011), as well as its formation via enzymatic debromination of higher PBDEs during metabolism in fish (Eljarrat et al. 2011). However, in young American yellow eels, BDE-100 and BDE-47 were found in similar concentrations (2.9 ± 0.93 ng g^{-1} ww and 2.8 ± 1.8 ng g^{-1} ww respectively) indicating a continued exposure of juvenile eels to congeners from the technical Penta- and Octa-PBDE mixtures. PBDE concentrations increased significantly (significant trend at 99 % confidence level according to Neumann trend test) over the life cycle, consistent with the bioaccumulation of PBDEs (Figure 1).

3.2. Alternate BFRs

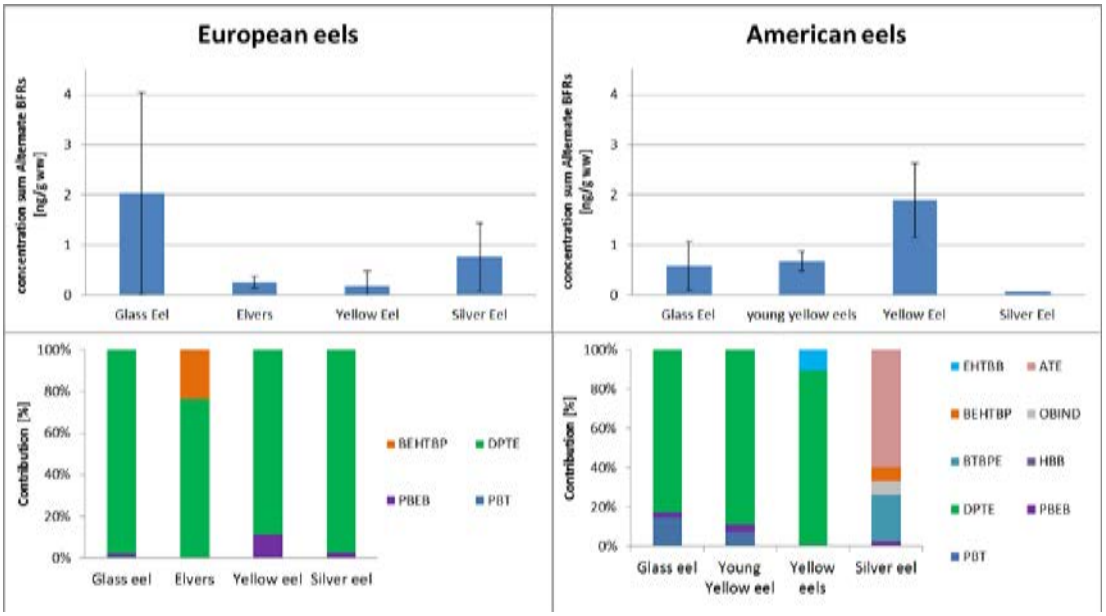

Figure 2: Concentration [ng g^{-1} ww] of sum Alternate BFRs (top) and contribution [%] of individual substances (bottom) to the different groups throughout the life cycle of European (left) and American (right) eels

DPTE was detected in European eels of all life cycle stages analysed with average concentrations of 1.1 ± 0.31 ng g^{-1} ww in glass eels, n.d. - 1.7 ng g^{-1} ww in yellow eels and 0.12 – 2.4 ng g^{-1} ww in silver eels. In American eels, DPTE was detected in the majority of the glass eel samples with up to 0.76 ng g^{-1} ww, and all yellow eel samples (Saint Lawrence River and Lake Ontario) with a mean of 1.68 ± 0.73 ng g^{-1} ww. However, DPTE was only detected in two of the young American yellow eel samples indicating that the contamination was not driven by life stage or age of the eel, but rather by local contamination sources such as e.g. contaminated sediments. European silver eels showed similar concentrations of DPTE (0.12 - 2.4 ng g^{-1} ww) to yellow eels indicating that this substance does not accumulate strongly throughout the life cycle, has been reintroduced recently, or is metabolised and excreted as soon as the eels stop feeding in their silver eel stage. The high concentration and abundance in American and European glass eels supports the hypothesis enunciated in our previous study that the uptake of DPTE happens in estuaries as well as rivers and is mostly driven by local contamination sources and not by age or life stage of individual eels (Sühring et al. 2013). It could, however, also be an indication for maternal transfer of DPTE. There are no data on current DPTE production (Vetter et al. 2010). However, it is thought to be persistent in sediments, a possible source of DPTE contamination for aquatic species (Fisk et al. 2003). The

higher concentrations and abundance in European eels can be explained by its former production and application in Germany (Vetter et al. 2010).

PBEB was also detected in European eels with similar average concentrations in different life stages, yet the frequency of detection increased with life stage. A variety of alternate BFRs were detected in American eels of all life stages, in lower concentrations than DPTE (Figure 2). In American glass and young yellow eels, the pattern of alternate BFRs concentrations was similar, with DPTE > PBT > PBEB. Byer et al. (2013a) reported a different distribution and more substances by high-resolution mass spectrometry in electron ionisation mode, but lower concentrations in American silver eels; the order of concentrations was ATE > BTBPE > OBIND > TBPH > PBEB > HBB > PBT. The difference in patterns might be due to differences in the analytical process especially because most alternate BFRs were detected in concentrations close to the limit of detection. In yellow eels from the upper Saint Lawrence River, TBB was detected in the majority of the samples, suggesting proximity to a point source (Table 1).

In general, it was concluded that the contamination patterns of alternate BFRs were induced by local contamination sources. The high frequencies of specific compounds at specific locations indicated that American eels were exposed to point sources. A possible source close to the American eel sampling sites is the OxyChem manufacturing facility at Niagara Falls, NY, which is known to produce flame retardants such as Dechlorane Plus (Sverko et al. 2011). Other sources at Lake Ontario could be wastewater treatment plants of the major urban centres Toronto and Hamilton. In European eels there was no characteristic contamination pattern at specific sampling sites, indicating an exposure to diffuse sources (Sühring et al. 2013). Possible sources could be e.g. diffuse emissions from waste incineration plants or leaching from consumer products. The high contributions of alternate BFRs to the sum contamination in both American and European glass eels compared to the older life stages emphasise the increasing relevance of these compounds since the phase-out and restriction of PBDEs.

3.3. Dechloranes

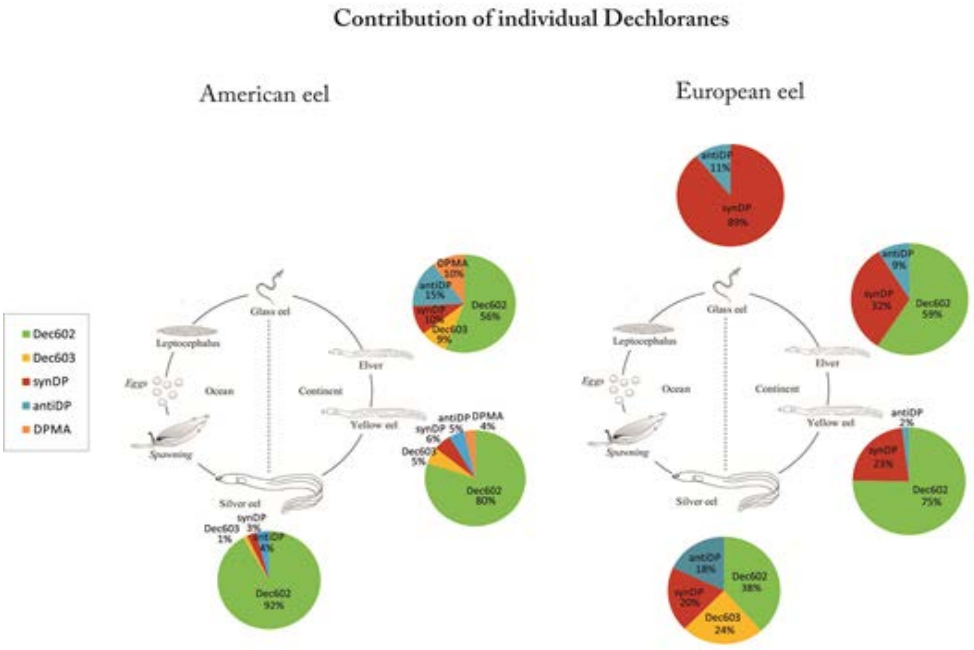

Figure 3: Contribution [%] of individual Dechloranes to the Sum Dechlorane contamination in American (left) and European (right) eels throughout their life cycle stages (picture life cycle: Dekker 2000)

In general, Dechlorane concentrations were highest in American yellow eels from Lake Ontario (1.7 – 5.0 ng g^{-1} ww), mostly driven by Dec-602 concentrations. Along the Saint Lawrence River, Dec-602 concentrations decreased towards the Atlantic Ocean, suggesting a source close to or at Lake Ontario (possibly OxyChem in Niagara Falls, NY, who are a known producer of DP (Sverko et al. 2011)).

DP concentrations were highest in yellow eels from the upper Saint Lawrence River (0.10 - 0.69 ng g^{-1} ww). In European eels, Dechlorane concentrations were similar in yellow and silver eels (0.013 - 0.50 ng g^{-1} ww in yellow eels and 0.017 - 0.38 ng g^{-1} ww in silver eels), suggesting that these eels were exposed to diffuse sources rather than to a specific point source. The overall contamination pattern was similar in European and American yellow and silver eels, with Dec-602 > DP > Dec-603 > DPMA (DPMA was only detected in American eels). This concurred with distributions reported in previous studies (Shen et al. 2010). The variability among samples, on

the other hand, was higher for European eels, whereas a greater number of Dechloranes were detected in American eels (Figure 3).

The high contribution of Dec-602 in European eels was unexpected, because it is not produced or imported to the EU. Even in North America (close to production facilities), it is only listed in the Non-domestic Substances List published by Environment Canada (http://www.ec.gc.ca/CEPARegistry/subs_list/NonDomestic.cfm). This indicated that Dec-602 is used internationally, but to date is not considered a substance of high priority or concern. However, Dec-602 has been reported to have a high bioaccumulation potential (higher for example than DP) and to be very bioavailable (Shen et al. 2011). Glass eels did not contain detectable concentrations of Dec-602, but it was the predominant Dechlorane in all other life cycle stages, suggesting little uptake during the oceanic phase of the eel. To determine how quickly Dec-602 becomes the major Dechlorane contaminant, the results of glass and adult eels were compared with the concentration in young American yellow eels and concentrations previously found in European elvers (Sühring et al. 2013). Elvers that had been in freshwater for less than a year already showed a predominance of Dec-602 (59% of total Dechlorane contamination). In American eels a similar progression was observed with no Dec-602 in glass eels and a relative contribution of 56% Dec-602 to total Dechlorane contamination in young yellow eels. This indicated a rapid uptake when juvenile eels enter their freshwater phase (Figure 3).

DP was detected in all analysed life stages of the European eel and all adult American eels (Figure 3). Of the two stereoisomers (syn- and antiDP), synDP was predominant in glass and yellow eels, with 96% relative contribution in European and 72% relative contribution in American yellow eels, respectively. These findings matched observations from previous studies indicating that synDP bioaccumulates and biomagnifies in fish to a greater extent than antiDP (Shen et al. 2011; Wu et al. 2010). However, the two isomers had a similar relative contribution to sum DP in European and American silver eels (60% and 56% respectively). This significant change in the isomer ratio over the life cycle of eels and from the technical product (75% antiDP; Sverko et al. 2011) has several implications. It confirms the assumption that synDP is the more bioaccumulative isomer in yellow eels. In contrast, when eels have stopped feeding in their silver phase, there seems to be either an uptake of antiDP via gills and skin, or a faster elimination of synDP from muscle tissue. Elimination could be induced by metabolism, excretion or redistribution of synDP to other fatty tissues such as gonads (Peng et al. 2012).

DPMA was detected in American yellow eels only, but was reported in American silver eels from a similar area (Byer et al. 2013a).

4. Conclusions

This study described the bioaccumulation of PBDEs over the life cycle of both American and European eels. Additionally, it was concluded that concentrations of alternate BFRs and Dechloranes were mostly driven by location and not by life stage. Contamination of American eels was likely caused by point sources in Lake Ontario or the upper Saint Lawrence River. In contrast, European eels seemed to be exposed primarily to diffuse sources, with no specific trend in the contamination pattern. In both American and European eels DPTE was a major contaminant, indicating existing sources and a continued release to the environment. Bans on the use of Penta-PBDE in the EU are effectively reducing PBDE contamination of juvenile eels. A significant increase of Dec-602 concentrations in the eel's freshwater phase was observed consistent with its high bioavailability and bioaccumulation potential.

In general, this study showed the relevance of continued monitoring of PBDE contamination in eels, and the emerging importance of contamination by alternate BFRs and Dechloranes. Further research is needed to identify the sources of contamination of compounds with no official record on production or application such as DPTE and Dec-602. It should also be investigated if the contaminations found in juvenile eels were caused by maternal transfer, as the transfer of BFRs to offspring could be a critical reason for concern.

Supporting Information

Tables on the samples, used standards, Blank values, LODs, LOQs, as well as a detailed list of the results are available in the supporting information.

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References

Belpaire, C. and G. Goemans. 2007. “Eels: contaminant cocktails pinpointing environmental contamination.” *ICES Journal of Marine Science* 64 (7): 1423-1436. doi:10.1093/icesjms/fsm121. http://icesjms.oxfordjournals.org/cgi/doi/10.1093/icesjms/fsm121.

Belpaire, C., Goemans, G., Geeraerts, C., Quataert, P., Parmentier, K., Hagel, P., De Boer, J. 2009 “Decreasing eel stocks: survival of the fattest?” *Ecology of Freshwater Fish* 2009: 18: 197–214. doi: 10.1111/j.1600-0633.2008.00337.x

BSEF. Bromine Science and Environmental Forum, http://www.bsef.com/, accessed 27. June 2013.

Byer, J.D., 2013a “ORGANOHALOGENATED PERSISTENT ORGANIC POLLUTANTS IN AMERICAN EEL (ANGUILLA ROSTRATA) CAPTURED IN EASTERN CANADA” Chapter 5; available at http://qspace.library.queensu.ca/bitstream/1974/8036/1/Byer_Jonathan_D_201305_PhD.pdf, accessed 23/07/2013

Byer, J.D., Lebeuf, M., Alaee, M., Brown, R.S., Trottier, S., Backus, S., Keir, M., Casselman, J., and Hodson, P. V. (2013b) Spatial trends of organochlorinated pesticides, polychlorinated biphenyls, and polybrominated diphenyl ethers in Atlantic anguillid eels. Chemosphere. 90, (5), 1719-1728.

Canadian Geographic 2008. “Rivers of Canada –Saint Lawrence River (Mixedwood Plains)” *Canadian Geographic – The Canadian Atlas Online;* http://www.canadiangeographic.ca/atlas/themes.aspx?id=rivers&sub=rivers_east_stlawrence&lang=En, accessed 12/07/2013

Covaci, A.,Harrad, S., Abdallah, M. A.-E., Ali,N. , Law, R.J., Herzke, D., de Wit, C.A.(2011) „Novel brominated flame retardants: A review of their analysis, environmental fate and behaviour“. Environment International 37 (2011) 532–556

Csiszar, S.A., Daggupaty, S.M., Verkoeyen, S., Giang, A., Diamond M.L.(2013) „ SO-MUM: A Coupled Atmospheric Transport and Multimedia Model Used to Predict Intraurban-Scale PCB and PBDE Emissions and Fate“. *Environmental science & technology* (47) pp. 436−445 doi: 0.1021/es3033023

Darnerud, P. O. Eriksen, G. S. Jóhannesson, T. Larsen, P. B. and Viluksela, M. 2001. “Polybrominated Diphenyl Ethers : Occurrence, Dietary Exposure, and Toxicology Chemical and Physical Properties of PBDEs”. Environmental Health Perspectives, 109, 49-68.

Dekker, W. 2000. The fractal geometry of the European eel stock. – ICES Journal of Marine Science, 57: 109–121.

De Wit, C.A., 2002. An overview of brominated flame retardants in the environment. Chemosphere 46, 583–624. <http://www.elsevier.com/locate/chemosphere>.

Domínguez, A.A., Law, R.J., Herzke, D., de Boer, J., 2011. Bioaccumulation of brominated flame retardants. In: Eljarrat, E., Barceló, D. (Eds.), Brominated Flame Retardants. The Handbook of Environmental Chemistry 16, 141-185. doi: 10.1007/698_2010_95. Springer-Verlag Berlin Heidelberg, Germany.

Durif, C., Dufour, S. and Elie, P.. The silvering process of Anguilla anguilla: a new classification from the yellow resident to the silver migrating stage. *Journal of Fish Biology*,66, 2005.

Eljarrat, E., Feo, M.L., Barceló, D., 2011. Degradation of brominated flame retardants. In: Eljarrat, E., Barceló, D. (Eds.), Brominated Flame Retardants. The Handbook of Environmental Chemistry 16, pp. 187–202. doi: 10.1007/698_2010_96. Springer-Verlag Berlin Heidelberg, Germany.

Environment Canada 2009 “Flows of the St. Lawrence River and its Main Tributaries http://www.ec.gc.ca/stl/default.asp?lang=En&n=B82B3625-1” accessed 26. June 2013

European Court of Justice. 2008. “Court of justice.” *Official Journal of the European Union*: C 116/2 - C 116/3.

Fisheries Forum. 2003. “Québec Declaration of Concern.” *Fisheries (Bethesda)* 28 (12): 28-30. www.fisheries.org.

Fisk, P., Girling, A.E., Wildey, R.J. 2003. Prioritisation of flame retardants for environmetal risk assessment. *Environment*. http://www.environment-agency.gov.uk/.

Van Ginneken, V.J.T, Maes, G.E. 2005. „The European eel (Anguilla anguilla, Linnaeus), its lifecycle, evolution and reproduction: a literature review" Reviews in Fish Biology and Fisheries 15:367–398. doi:10.1007/s11160-006-0005-8

ICES. 2008. "European eel." *ICES Advice 2008* Book 9: 123-129.

Mikael Harju, Eldbjørg S. Heimstad Dorte Herzke Torkjel Sandanger Stefan Posner and Frank Wania: Emerging "new" Brominated flame retardants in flame retarded products and the environment: Current State of Knowledge and Monitoring requirements, checked on 27/09/2012.

Ministry of Natural Resources 2007. "American eel in Ontario" *State of resources reporting.* February 2007; 978-1-4435-2056-0 PDF http://www.mnr.gov.on.ca/stdprodconsume/groups/lr/@mnr/@sorr/documents/document/stel02_166010.pdf, accessed 12/07/2013

Möller, A., Xie, Z., Sturm, R., and Ebinghaus, R. 2010. "Large-scale distribution of dechlorane plus in air and seawater from the Arctic to Antarctica." *Environmental science & technology* 44 (23): 8977-82. doi:10.1021/es103047n. http://www.ncbi.nlm.nih.gov/pubmed/21047104.

Netzband, A., Reincke, H. and Bergemann, M. 2002. „The River Elbe - A Case Study for the Ecological and Economical Chain of Sediments". *JSS – J Soils & Sediments* 2 (3) 112 – 116 (2002)

US EPA 2010. "An Exposure Assessment of Polybrominated Diphenyl Ethers". EPA/600/R-08/086F. Available at http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=210404; accessed 24.06.2013

Palstra, A.P., van Ginneken, V .J. T., Murk, A. J. and van den Thillart, G. E .E .J .M. 2006. "Are dioxin-like contaminants responsible for the eel (*Anguilla anguilla*) drama?" *Die Naturwissenschaften* 93 (3): 145-8. doi:10.1007/s00114-005-0080-z. http://www.ncbi.nlm.nih.gov/pubmed/16508793.

Peng, H., Zhang, K., Wan, Y., Hu, J. 2012. „Tissue Distribution, Maternal Transfer, and Age-Related Accumulation of Dechloranes in Chinese Sturgeon." *Environmental science & technology* 46, pp. 9907–9913; doi:10.1021/es3025879

SCOP. 2009. “Report of the Conference of the Parties of the Stockholm Convention on Persistent Organic Pollutants on the Work of Its Fourth Meeting; Stockholm Convention on Persistent Organic Pollutants: Geneva, 4 to 8 May 2009.” *World Trade* (May 2009). *Chemosphere* 78 (2010): 134–138

Shen, Li; Reiner, Eric J.; MacPherson, Karen A.; Kolic, Terry M.; Sverko, Ed; Helm, Paul A. et al. 2010. „Identification and Screening Analysis of Halogenated Norbornene Flame Retardants in the Laurentian Great Lakes: Dechloranes 602, 603, and 604”. *Environmental science & technology* 44 (2), pp. 760–766.

Shen, L., Reiner, E.J., Helm, P.A., Marvin, C.H., Hill, B., Zhang, X., MacPherson, K.A., Kolic, T.M., Tomy, G.T., and Brindle, I.D. 2011a. “Historic trends of dechloranes 602, 603, 604, dechlorane plus and other norbornene derivatives and their bioaccumulation potential in lake ontario.” *Environmental science & technology* 45 (8): 3333-40. doi:10.1021/es104328r. http://www.ncbi.nlm.nih.gov/pubmed/21434636.

Shen, L., Reiner, E.J.,Karen a MacPherson, MacPherson, K.A., Kolic, T.M., Richman, L.A., Marvin, C.H, Burniston, D.A., Hill, B., Brindle, I.D., McCrindle, R. and Chittim, B.G. 2011. “Dechloranes 602, 603, 604, Dechlorane Plus, and Chlordene Plus, a newly detected analogue, in tributary sediments of the Laurentian Great Lakes.” *Environmental science & technology* 45 (2) : 693-9. doi:10.1021/es1027844. http://www.ncbi.nlm.nih.gov/pubmed/21133428.

Sühring, R., Möller, A., Freese, M., Pohlmann, J., Wolschke H., Sturm,R., Xie, Z., Hanel R., Ebinghaus, R. 2013. „Brominated flame retardants and dechloranes in eels from German rivers*.” Chemosphere* 90 (2013) 118–124. http://dx.doi.org/10.1016/j.chemosphere.2012.08.016

Sverko, E., Tomy, G.T., Reiner, E.J., Li, Y., McCarry, B.E., Arnot, J.A., Law, R.J. and Hites, R.A. 2011. “Dechlorane Plus and Related Compounds in the Environment: A Review.” *Environmental science & technology* . doi:10.1021/es2003028. http://www.ncbi.nlm.nih.gov/pubmed/21574656.

Tesch, F.W., Thorpe, J.E. (Ed.) 2003 “The Eel” 3rd edition. p.122, p.152, ISBN 0-632-06389-0, Blackwell Science Ltd, a Blackwell Publishing Company, Oxford, UK

Vetter, W., von der Recke, R., Ostrowicz, P., Rosenfelder, N. 2010. „Liquid chromatographic enantioseparation of the brominated flame retardant 2,3-dibromopropyl-2,4,6-tribromophenyl ether (DPTE) and enantiomer fractions in seal blubber.” doi:10.1016/j.chemosphere.2009.09.071

Wu, J., Zhang, Y., Luo, X., Wang, J., Chen, S., Guan, Y., and Mai, B. 2010. “Isomer-specific bioaccumulation and trophic transfer of Dechlorane Plus in the freshwater food web from a highly contaminated site, South China.” *Environmental science & technology* 44 (2): 606-11. doi:10.1021/es902744b. http://www.ncbi.nlm.nih.gov/pubmed/19994895.