SPATIAL AND TEMPORAL DISTRIBUTION OF POLYBROMINATED DIPHENYL ETHERS IN LAKE TROUT FROM THE GREAT LAKES

Jennifer M. Luross, Mehran Alaee*, David B. Sergeant**, D. Michael Whittle**, Keith R. Solomon

University of Guelph, Guelph, Ontario. N1G 2W1 Canada
* National Water Research Institute, 867 Lakeshore Road, P.O. Box 5050 Burlington, Ontario. L7R 4A6 Canada
** Great Lakes Laboratory for Fisheries and Aquatic Sciences, 867 Lakeshore Road, P.O. Box 5050 Burlington, Ontario. L7R 4A6 Canada

Introduction

Flame retardants are substances that are added to materials in order to suppress, significantly reduce or delay combustion [1]. There are four major groups of flame retardants: inorganic, halogenated, organophosporus, and nitrogen-based that can be either reactive (chemically built into a polymer molecule) or additive (added to the polymer). This study involves the halogenated group of flame retardants which generates 25% of the total worldwide production[2]. From this group, the main focus was on brominated flame retardants, more specifically, polybrominated diphenyl ethers (PBDEs). Commonly used as an additive flame retardant, PBDEs are found in a wide-range of products including paints, plastics, textiles, and electrical devices. With the extensive use of these products, the concern for human and environmental health intensifies to the extent that PBDEs are now being classified as emerging persistent organic pollutants (POPs). The majority of the research is from Europe where Swedish studies have shown bioaccumulation of PBDEs in fish, seals, and birds[3,4,5,6]. PBDE congeners were also observed in marine fish and shellfish from Japan[7]. Norén and Meironyté have shown that concentrations of PBDEs in human breast milk have doubled every five years over the last 25 years[8]. Initially, there was limited information on the environmental fate and distribution of PBDEs in North America yet current studies assessing the impact of these compounds have shown detectable levels in marine mammals from the Arctic[9] and the Gulf of St. Lawrence[10], as well as wildlife tissues and eggs[11].

In order to establish current levels of PBDEs in the Great Lakes, lake trout (*Salvelinus namaycush*), a top predator species in the Great Lakes food web, were obtained from Lake Ontario, Lake Huron, Lake Erie, and Lake Superior in 1997. Lake trout from Lake Ontario collected in 1978, 1983, 1988, 1993, and 1998 were used to assess the historical inputs of PBDEs to the Great Lakes. These specimens have been archived as whole fish homogenates by the Great Lakes Laboratory for Fisheries and Aquatic Sciences, Department of Fisheries and Oceans, Canada. Ten fish per lake were analyzed using a method that was recently developed involving high-resolution mass spectrometry and certified reference materials[12]. This method, when applied to the lake trout samples, identified a pattern for the distribution of PBDEs and their concentrations in North American lake trout.

Materials and Methods

A 10g aliquot of whole fish homogenate was ground using a mortar and pestle with 130g of anhydrous sodium sulphate until free flowing. The mixture was transferred into a large chromatography column, spiked with a chlorinated diphenyl ether surrogate mixture, and eluted with 300mL of methylene chloride (DCM). Samples were concentrated by a combination of rotary evaporation and nitrogen evaporation prior to gel permeation chromatography (GPC) for bulk lipid removal. The GPC unit was an automated ABC Laboratories Autoprep model 1002A. The column was packed with 60g of Bio Beads S-X3, 200-400 mesh (Bio-Rad Laboratories, Richmond, CA) in a 25mm X 600mm glass column. The elution solvent was 300mL of 1:1

DCM:hexane in which 150mL was collected. Fractionation was accomplished with 3% deactivated silica gel columns; eluted with 100mL of DCM and activated micro-alumina (Woelm N-Super 1, type W 200, Universal Scientific Incorporated, Atlanta, Georgia) columns; eluted with 5mL hexane and 10mL toluene. Samples evaporated to dryness at room temperature in autosampler vials and 20uL of performance standard was added for analysis by high resolution mass spectrometry. Analyses of PBDEs were carried out on a VG AutoSpec-Q mass spectrometer connected to a Hewlett-Packard 5890 GC equipped with a CTC A200s autosampler. The GC injection port was configured for 1uL on-column injections, with an initial temperature of 160°C, held for 1 minute, and ramped at 100°C/min to 280°C for 55 minutes. Gas chromatographic separation prior to MS was achieved using a 60m X 0.25mm X 0.25μm Restek Rt_x5 capillary column. The GC conditions were the same as above with the exception of the final hold was extended to 60 minutes. Total run time was 90.7 min. Sample ionization was performed by electron ionization (EI) at an electron voltage ranging from 30 to 40eV depending on the optimization parameters of the instrument. Source temperature was 270°C and the resolving power of the analyzer was 10 000. The mass spectrometer was operated in SIM mode using a total of 8 descriptors to analyze the 23 PBDE congeners. Samples were quantified by an internal standard using Excel and 20/20 spreadsheet, following EPA 8290 QA/QC protocols. Analytical, surrogate spiking, and performance standards were purchased from Cambridge Isotope Laboratories in Andover, Massachusetts.

Results and Discussion

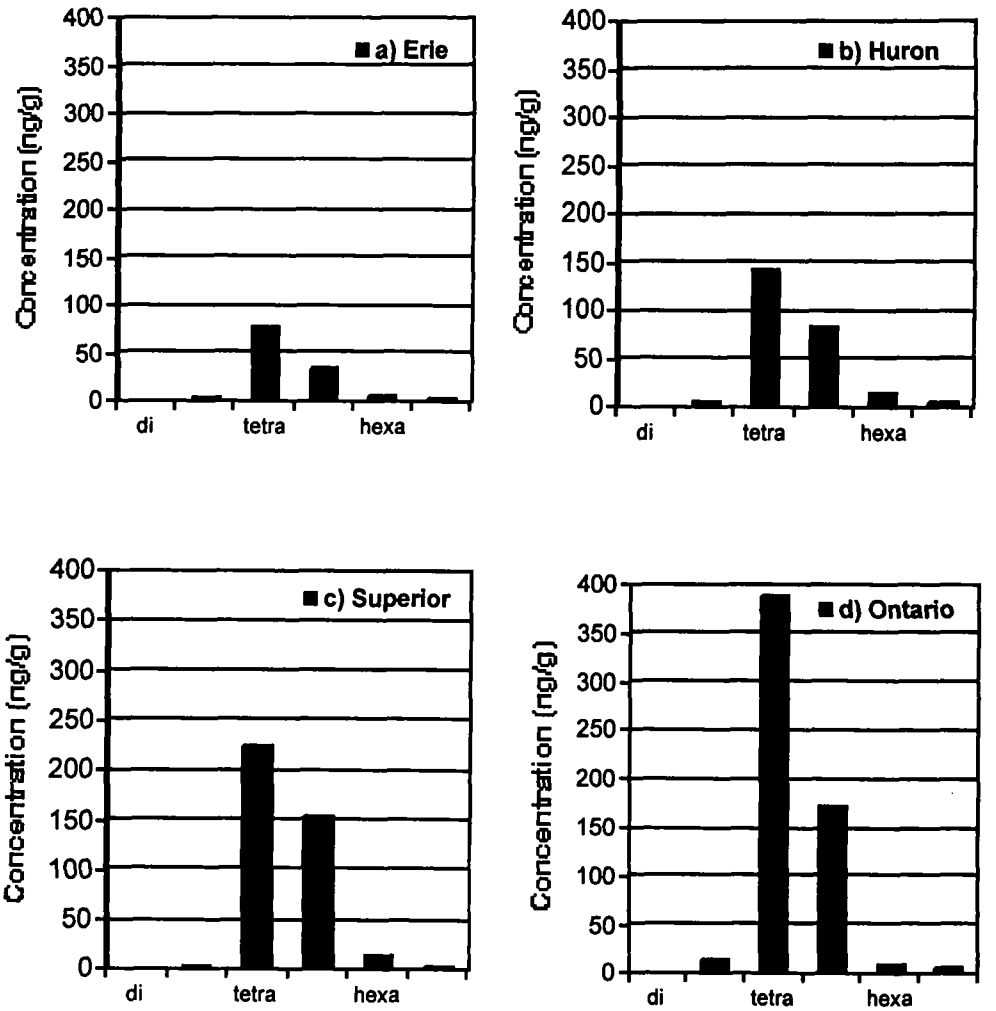
Spatial:
Figure 1 shows similar homologue patterns in lake trout from the Great Lakes, with the highest levels in the tetra group followed by penta and then hexa. The ratio of the penta homologue group in comparison to the tetra homologue group for lake trout from Lake Ontario and Lake Erie are the equal at 44%, while Lake Huron and Lake Superior lake trout have a larger penta to tetra ratio at 59% and 68%, respectively. Overall fish collected from Lake Ontario, a heavily industrialized and urban area, had the highest concentrations of total PBDEs (604ng/g lipid), followed by fish from Lake Superior (392ng/g lipid), then Lake Huron (247ng/g lipid), and Lake Erie (117ng/g lipid).

The concentrations of the individual congeners varied between the lakes. Lake Huron fish had slightly higher levels of all the lower brominated compounds (di and tri-BDEs) and 2, 2', 4, 4', 5, 5'-hexa-BDPE (BDE-153) when compared to Lake Superior. Lake Ontario lake trout had slightly lower levels of BDE-77 and BDE-99 in comparison to Lake Superior. The difference in congener levels of PBDEs can be attributed to the variation in local sources versus atmospheric transport of these compounds. Elevated amounts of 2, 2', 4, 4'-tetra-BDPE (BDE-47) a predominant PBDE congener and major constituent of the commonly used fire retardant Bromkal 70-5DE®, were detected in all lake trout samples analyzed and contributed 55% to the total values for each lake. The second and third most predominant congeners were 2, 2', 4, 4', 5-penta-BDPE (BDE-99) (15% of total), and 2, 2', 4, 4', 5, 5'-hexa-BDPE (BDE-153) (4% of total), respectively. These congeners share the 2, 2', 4, 4' substitution that seems to be important for bioaccumulation in lake trout. These results suggest that PBDEs are ubiquitous pollutants in lake trout from the Great Lakes.

Temporal:
Results from the Lake Ontario temporal study indicated an increase in concentration of total PBDEs in lake trout over time. Lake trout analyzed in 1998 had the highest concentration of total PBDEs at 945ng/g lipid, followed by 1993 (434ng/g lipid), 1988 (171ng/g lipid), 1983 (8ng/g lipid), and finally in 1978 (3ng/g lipid). The same congeners as detected in the spatial segment of the study were again the most abundant with BDE-47, BDE-99, and BDE-153 making up 50%, 15%, and 5% of the totals, respectively. Individual congeners were similar to the total PBDE pattern with an exception for 4, 4'-DiBDPE which had higher concentrations in fish from 1978. Figure 2 a & b shows the temporal trend of BDE-47and BDE-99. The increase of PBDEs seen in

the early 1980's corresponds with the ban of polybrominated biphenyls (PBBs) and Mirex during the late 1970's. As a result of increased PBDE utilization, higher levels have been found in the environment. This rise in PBDE concentrations is consistent with the results of a similar study involving different biota which also showed increasing levels of BDE-47, -99, and –100 since the 1970's[13]. During the 1980's the calculated doubling rate for BDE-47 was every 2 years but the rate has since slowed in the 1990's to approximately 4 years. This trend has been seen in other studies where a general increase in the tetra-BDE congeners in biota occurred during the 1980's, but has now stabilized or started to decrease[14].

Figure 1 (a,b,c,& d): 1997 Homologue group distribution of PBDEs (ng/g lipid) in Lake Trout from the Great Lakes



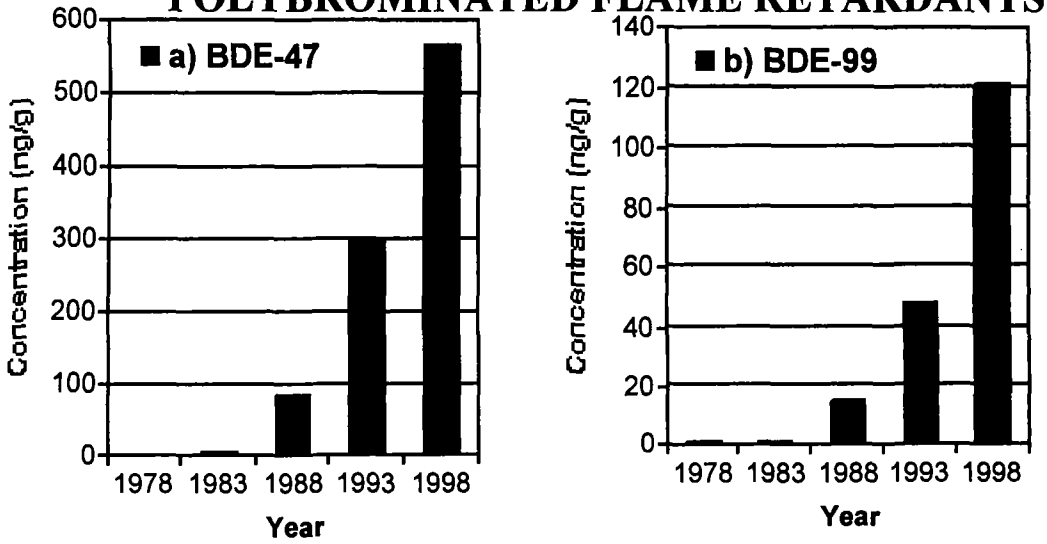


Figure 2 (a & b): Concentration of BDE-47 and BDE-99 (ng/g lipid) in Lake Trout from the Great Lakes between 1978 and 1998.

Acknowledgements

The Department of Fisheries and Oceans under the Toxic Chemical Program and TSRI have funded this research. Special thanks to Bob Wilkinson for his mass spec assistance and the staff of the Ultra-Trace Laboratory: Cam MacEachen, Susan Blunt, and Mary Malecki.

References

1. IPCS, Environmental Health Criteria 192: Flame Retardants: A General Introduction, World Health Organization: Geneva, Switzerland, (1997).
2. IPCS, Environmental Health Criteria 162: Brominated diphenyl ethers, World Health Organization: Geneva, Switzerland, (1994).
3. Andersson, O., and Blomkvist, G. (1981). *Chemosphere* 10 (9): 1051-1060.
4. Jansson, B., Asplund, L., and Olsson, M. (1987). *Chemosphere* 16 (10-12): 2343-2349.
5. Jansson, B., Andersson, R., Asplund, L., Litzén, K., Nylund, K., Sellström, U., Uvemo, U., Wahlberg, C., Wideqvist, U., Odsjö, T., and Olsson, M. (1993). *Environmental Toxicology and Chemistry* 12: 1163-1174.
6. Sellström, U., Jansson, B., Kierkegaard, A., de Wit, C., Odsjö, T., and Olsson, M. (1993). *Chemosphere* 26 (9): 1703-1718.
7. Watanabe, I., Kashimoto, T., and Tatsukawa, R. (1987). *Chemosphere* 16 (10-12): 2389-2396.
8. Norén, K., and Meironyté, D. (1998). Organohalogen Compounds 38: 1-4.
9. Alaee, M., Luross, J.M., Sergeant, D.B., Muir, D.C.G., Whittle, D.M., and Solomon, K.R. (1999) Organohalogen Compounds 40: 347-350.
10. Lebeuf, M. (1999) Workshop: Polybrominated Diphenyl Ethers in the Environment. NWRI Contribution No. 00-153. Eds. M. Alaee, J.M. Luross, and D.B. Sergeant. Canada Centre for Inland Waters, Burlington, Ontario. Canada.
11. Simon, M., and Wakeford, B. (1999) Workshop: Polybrominated Diphenyl Ethers in the Environment. NWRI Contribution No. 00-153. Eds. M. Alaee, J.M. Luross, and D.B. Sergeant. Canada Centre for Inland Waters, Burlington, Ontario. Canada.
12. Sergeant, D.B., Alaee, M., Luross, J., and Ikonomou, M. (1998). Organohalogen Compounds 35: 379-383.
13. de Wit, C.A. (1999) Organohalogen Compounds 40: 329-331.
14. Renner, R. (2000) Environmental Science and Technology News. 223A-226A.