Global Assessment of Polybrominated Diphenyl Ethers in Farmed and Wild Salmon

RONALD A. HITES,*,† JEFFERY A. FORAN,‡ STEVEN J. SCHWAGER,§ BARBARA A. KNUTH,∥ M. COREEN HAMILTON,⊥ AND DAVID O. CARPENTER∥
School of Public and Environmental Affairs, Indiana University, Bloomington, Indiana 47405, Midwest Center for Environmental Science and Public Policy, Milwaukee, Wisconsin 53202, Department of Biological Statistics and Computational Biology and Department of Natural Resources, Cornell University, Ithaca, New York 14853, AXYS Analytical Services Ltd., P.O. Box 2219, 2045 Mills Road, Sidney, British Columbia, Canada V8L 3S8, and Institute for Health and the Environment, University at Albany, Rensselaer, New York 12144

We have shown recently that levels of persistent, bioaccumulative contaminants (polychlorinated biphenyls, dioxins, and several chlorinated pesticides) are significantly higher in farmed than in wild salmon and that European farm-raised salmon have significantly greater toxic contaminant loads than those raised in North and South America. In this paper, we extend these results to polybrominated diphenyl ethers (PBDEs) and show that farm-raised salmon have higher levels of these compounds than wild salmon. We also show that farm-raised salmon from Europe have higher PBDE levels than those raised in North America and that both European and North American farm-raised salmon have higher PBDE levels than those farmed in Chile. Among the species of wild salmon, chinook had significantly elevated PBDE levels relative to the other wild species. These elevated PBDE levels may be related to chinook’s feeding behavior and trophic level. Among all of the wild species we studied, chinook tend to feed higher in the food web throughout their adult life and grow to be larger individuals.

Introduction
Polybrominated diphenyl ethers (PBDEs) serve as flame retardants in a wide variety of commercial and household products. For example, polyurethane foam, which is used widely in upholstered furniture, is flammable unless it is treated with suitable flame retardants such as PBDEs. Because many governments now have regulations requiring household products to be flame resistant, PBDEs have become an important commercial substance. Not surprisingly, the use of PBDEs has increased over the years, and global annual sales are now ~70 000 t (1). PBDEs are available commercially as three products, two of which are mixtures of several congeners. The so-called penta-product contains 2,2′,4,4′-tetrabromodiphenyl ether (BDE-47), 2,2′,4,4′,5-pentabromodiphenyl ether (BDE-99), 2,2′,4,4′,6-pentabromodiphenyl ether (BDE-100), 2,2′,4,4′,5,5′-hexabromodiphenyl ether (BDE-153), and 2,2′,4,4′,5,6,6′-hexabromodiphenyl ether (BDE-154) in a ratio of about 9:12:2:1:1:1 (1). Over 95% of the penta-product produced worldwide is now used in the United States and Canada. The octa-product contains several hexa- to nona-brominated congeners, and the deca-product is composed almost entirely of decabromodiphenyl ether (BDE-209).

Despite their societal benefits, PBDEs seem to be migrating from the products in which they are used and entering the environment and people. PBDEs are now ubiquitous; they can be found in air, water, fish, birds, marine mammals, and people; in many cases, the concentrations of these compounds have increased over the last 20 years (1). One source of PBDEs in people is their food supply, and an increasingly important food is salmon, which is nutritious and high in beneficial fats.

Between 1987 and 1999, salmon consumption increased annually at a rate of 14% in the European Union and 23% in the United States (2). Currently, over half the salmon sold globally is farm-raised in Northern Europe, Chile, Canada, and the United States; the annual global production of farmed salmon (predominantly Atlantic salmon) has risen from ~27 000 to over 1 million t during the past two decades (3). The health benefits of eating fish such as salmon have been well-documented (4, 5). However, both farmed and wild salmon have been shown to bioaccumulate contaminants, albeit to different levels (6).

In a previous study, we reported that levels of persistent, bioaccumulative contaminants (such as PCBs, dioxins, and several chlorinated pesticides) are significantly higher in farm-raised salmon than in wild Pacific salmon and that salmon raised on European farms have significantly greater toxic contaminant loads than those raised on North and South American farms (6). In this paper, we provide results on polybrominated diphenyl ether (PBDE) concentrations in salmon purchased from farms in eight major salmon farming regions, in five species of wild Pacific salmon, and in salmon fillets purchased at supermarkets in North America and Europe.

Materials and Methods
PBDEs were measured in about 700 farmed and wild salmon (totaling ~2 t) collected from around the world. Farmed Atlantic salmon (Salmo salar) were purchased from wholesale suppliers in the United States, United Kingdom, Norway, and Canada between March and December 2002. These suppliers provided farmed salmon from eight major salmon farming regions: Norway, Chile, Scotland, British Columbia, Eastern Canada, the Faroe Islands, Maine, and the state of Washington. These are the top farmed salmon-producing regions (by weight) in the world—with the exception of Maine and Washington, which lag behind Ireland in production. Maine and Washington were chosen instead of Ireland because of their interest to U.S. consumers.

One to three wholesale suppliers provided fish from 2 to 10 farms in each region for a total of 51 different salmon farms, with representation from each of these eight regions. Suppliers provided information on the origin of the fish...
(region and farm) by including the original labels from the farm source where possible or by filling in labels we provided to them for this purpose. When necessary for clarification, we confirmed the written information verbally or by e-mail with the suppliers. The farms chosen reflect those from which suppliers could obtain farmed Atlantic salmon of the appropriate size within the sampling period. Ten fish were obtained from each farm, nine of which were grouped randomly into three composites of three fish each. Most individual fish weighed ~4–6 kg. A total of 459 farmed salmon from wholesalers was used to produce 153 samples for analysis. Viscera and gills from all whole fish were removed before they were shipped; the heads were left on.

Between September 2001 and August 2002, other suppliers provided 135 wild fish representing five wild species of Pacific salmon: chum (Oncorhynchus keta), coho (O. kisutch), chinook (O. tshawytscha), pink (O. gorbuscha), and sockeye (O. nerka). Samples of each species were purchased from different geographic regions, including Kodiak, AK; Southeast Alaska; British Columbia; and Oregon. Three composites of three fish for each species at each location resulted in a total of 45 samples for analysis. We did not analyze wild Atlantic salmon because few are available commercially; nor did we analyze farmed Pacific salmon because they are not raised in any substantial amounts (3, 7).

Between March and November 2002, we purchased an additional 144 salmon fillets (three whole fillet samples from each of three retail outlets) from supermarkets in Boston, Chicago, Denver, Edinburgh, Frankfurt, London, Los Angeles, New Orleans, New York, Oslo, Paris, San Francisco, Seattle, Toronto, Vancouver, and Washington, DC. Purchasers asked specifically for farmed salmon at fish counters and were instructed not to purchase any other type of salmon. Fillets from supermarkets were composited by the retail outlet where they were purchased. Composite samples consisted of three fillets per retail outlet, for a total of 48 samples.

We also analyzed 13 samples of salmon feed purchased from May to December 2002 from the European, North American, and South American outlets of the two major feed companies, which together have ~80% of the global market for fish feed (8). These samples are not necessarily the specific feeds fed to the farmed fish we sampled; our intention was to identify contaminant trends in feed available typically in the global market. For the first company, two samples of feed, purchased 3–4 months apart, were obtained from facilities in Scotland, Eastern Canada, British Columbia, and Chile. For the second company, two samples of feed, purchased 3–4 months apart, were obtained from facilities in Scotland and British Columbia, and one sample was obtained from a facility in Chile. Where possible, two samples per location were purchased several months apart to account for possible seasonal variations in the feed formulation.

All samples were shipped to the analytical laboratory (AXYS Analytical in Sidney, BC) fresh or frozen on ice or gel-packs. Fish were thawed and inspected by a fisheries biologist to verify species. Each fish was weighed, its length was measured, and it was filleted to give two skin-on fillets. We analyzed skin-on fillets because most salmon are sold at retail outlets with the skin on. In each case, the fillets from three fish were ground and re-ground together to make a homogeneous composite.

Ten grams of wet fish tissue or fish feed was mixed with known amounts of nine individual, fully 13C-labeled brominated diphenyl ethers before extraction, ground with anhydrous Na2SO4, and Soxhlet extracted with dichloromethane for 16 h. The extract was cleaned up by gel permeation chromatography on BioBeads SX-3 and fractionated on Florisil, silica, and alumina. Gas chromatographic mass spectrometric analysis of PBDEs was accomplished using a Micromass Autospec Ultima magnetic sector high-resolution mass spectrometer equipped with a Hewlett-Packard 6890 gas chromatograph. The mass spectrometer was operated at a static mass resolution of 5000–6000. Chromatographic separation was achieved using a Durabond DB-5HT high-temperature column (30 m x 250 μm i.d., 0.10 μm film thickness). The PBDE concentrations were obtained by isotope dilution quantification using the 13C-labeled internal standards.

All analyses were conducted in accordance with the AXYS accredited QA/QC program. Regular participation in international inter-laboratory calibration programs for PBDE analysis, such as those of Quasimeme and the University of Umea, are key components of this program. Each analysis batch of nine samples also included a procedural blank, a “known” or laboratory control sample, and an analysis duplicate. The sample results were reviewed and evaluated in relation to the QA/QC samples worked up at the same time. The sample internal standard recoveries and detection limits, procedural blank data, and laboratory control sample data were evaluated against method criteria to ensure data quality. All instrument QA specifications for EPA methods were adhered to and applied to all analyses conducted for this study. All data met the QA/QC specifications. In general, duplicate measurements differed from each other by <15%. For most reported congeners, blank concentrations were below the detection limits; small concentrations of a few congeners were detected in the blanks but in such cases the observed levels were significantly less (more than 2 orders of magnitude) than in the samples; hence, blank values were not subtracted from the sample measurements.

PBDE concentrations are reported as the sum of the concentrations of the following PBDE congeners: 1, 2, 3, 7, 8, 10, 11, 12, 13, 15, 17, 25, 28, 30, 32, 33, 35, 37, 47, 49, 66, 71, 75, 77, 85, 99, 100, 105, 116, 119, 126, 138, 140, 153, 154, 155, 166, 181, 183, 190, 206, 207, and 208. Detection limits ranged from less than 1 pg/g up to ~10 pg/g, depending on the congener.

PBDE levels in farmed and wild salmon were compared by analysis of variance. In comparing wild and farmed salmon, we considered farmed salmon as a single group. In addition, regions in which salmon were farmed were compared by analysis of variance with multiple comparisons of means to test for differences among locations in PBDE levels. In all analyses of variance, the replicate composites from each source were not assumed to be independent observations. Differences between farmed and wild salmon and differences among farming locations were consistently substantial and highly significant.

Results and Discussion

Figure 1 shows that total PBDEs in the farmed salmon were significantly more concentrated as a group (red bars) than in the wild salmon (green bar) [F = 31.05, p < 0.0001, with df = (1, 64)]. PBDE concentrations were significantly higher in farmed salmon from Europe than from North America, in farmed salmon from North America than from Chile, and in farmed salmon from Chile than in wild salmon [F = 135.16, p < 0.0001 with df = (1, 62) for all]. PBDE concentrations in the salmon purchased from retail outlets (yellow bars) in Europe were significantly higher than those in salmon purchased in stores in North America [F = 5.69, p = 0.0317 with df = (1, 14)], but both types of store-bought samples had average PBDE concentrations much higher than in wild salmon. The PBDE concentrations we observed were in the same range as those measured by Jacobs et al. (9), who found PBDE levels of ~5 ng/g wet weight in 13 samples from European salmon farms.

Figure 2 shows the PBDE concentrations as a function of location. Salmon fillets obtained from commercial outlets in the various cities (yellow bars) generally clustered with the
farmed samples and not with the wild samples. PBDE concentrations were highest in wild chinook from British Columbia and in farmed salmon from Scotland and western Canada and lowest in farmed salmon from Chile and Washington. Salmon fillets purchased from supermarkets in Edinburgh and London were generally the most contaminated with PBDEs, and those purchased in Washington, DC, and New Orleans were the least contaminated of the store-bought samples. Most of the salmon sold in European stores comes from European farms, which produce the more contaminated salmon, while most of the salmon sold in U.S. stores comes from Chile and Canada (10, 11).

The relatively high PBDE concentration in the chinook samples from British Columbia and Oregon (see Figure 2) was interesting. In fact, an analysis of variance for total PBDE concentrations in the 45 samples of wild salmon, treating the three composites from each source as replicates, showed significant differences among the species \( F = 3.93, p = 0.0360, \) df = 3, 10, with the chinook elevated relative to the other species. The average total PBDE concentration in the 9 individual chinook samples was \( 2.258 \pm 0.565 \) ng/g as compared to \( 0.130 \pm 0.020 \) ng/g in the 36 others. The elevated PBDE levels found in the wild chinook may be related to their feeding behavior and trophic level. Among all of the wild species we studied, chinook tend to feed higher in the food web throughout their adult life stage, feeding mainly on fish, and grow to be larger individuals on average (12, 13), whereas other species’ diets rely more on invertebrates and zooplankton (14–17).

The large differences between the farmed and wild salmon PBDE concentrations are most likely a function of their diet. Farmed salmon are fed a concentrated feed high in fish oils and fish meal, which is obtained primarily from small pelagic fishes (18). We analyzed 13 samples of commercial salmon feed (see Table 1). Although the PBDE concentrations in these feed samples are quite variable, they are generally similar to or greater than those in the farmed salmon. There were no significant differences among the four locations (British Columbia, Eastern Canada, Chile, and Scotland) where the feed was purchased, among the three continents (North America, South America, and Europe) where the feed was purchased, or between the two purchase periods. However, there was a significant difference between the two feed companies \( p = 0.0242 \). Given the largely unknown sources of the pelagic fish that make up the fish meal from which the feed is produced and the proprietary nature of the fish feed recipes, it is difficult to speculate about why the two companies’ products have different PBDE levels.

The congener profiles of PBDEs in these salmon are shown in Figure 3. Because we purchased farmed (not wild) salmon from the retail outlets, it is not surprising that the profiles of these two types of samples are virtually identical (compare Figure 3 top and middle). In these samples, BDE-47 dominates, just as it does in almost all ambient environmental samples and in people (1). The next most abundant con-
TABLE 1. Total PBDE Concentrations in Fish Feed Purchased from Various Global Suppliers

<table>
<thead>
<tr>
<th>purchase date (in 2002)</th>
<th>sampling period*</th>
<th>location</th>
<th>lipid concn (%)</th>
<th>total PBDE (ng/g wet wt)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B May 21</td>
<td>1</td>
<td>Scotland</td>
<td>32.85</td>
<td>10.92</td>
</tr>
<tr>
<td>A Sep 26</td>
<td>2</td>
<td>BC</td>
<td>30.23</td>
<td>9.27</td>
</tr>
<tr>
<td>A Apr 30</td>
<td>1</td>
<td>BC</td>
<td>31.69</td>
<td>8.88</td>
</tr>
<tr>
<td>A Jul 24</td>
<td>1</td>
<td>Scotland</td>
<td>33.41</td>
<td>7.67</td>
</tr>
<tr>
<td>A Oct 21</td>
<td>2</td>
<td>BC</td>
<td>28.25</td>
<td>6.75</td>
</tr>
<tr>
<td>B May 17</td>
<td>1</td>
<td>EC</td>
<td>36.32</td>
<td>5.68</td>
</tr>
<tr>
<td>B Dec 24</td>
<td>2</td>
<td>Chile</td>
<td>33.23</td>
<td>5.16</td>
</tr>
<tr>
<td>B Sep 20</td>
<td>2</td>
<td>Scotland</td>
<td>34.38</td>
<td>5.24</td>
</tr>
<tr>
<td>B Sep 6</td>
<td>2</td>
<td>EC</td>
<td>33.65</td>
<td>5.96</td>
</tr>
<tr>
<td>B Oct 7</td>
<td>2</td>
<td>BC</td>
<td>32.17</td>
<td>0.98</td>
</tr>
<tr>
<td>B Dec 24</td>
<td>2</td>
<td>Chile</td>
<td>31.57</td>
<td>0.60</td>
</tr>
<tr>
<td>B Jun 6</td>
<td>1</td>
<td>Chile</td>
<td>38.84</td>
<td>0.50</td>
</tr>
<tr>
<td>B Jul 10</td>
<td>1</td>
<td>BC</td>
<td>34.99</td>
<td>0.49</td>
</tr>
</tbody>
</table>

*The samples were assigned to group 1 if they were purchased between April and July and to group 2 if they were purchased between September and December.

FIGURE 3. PBDE congener profiles in farmed (top), retail (middle), and wild (bottom) salmon. The error bars represent standard errors.

geners, BDE-99 and BDE-100, are also relatively abundant in most other environmental samples. The PBDE congener profile of the wild salmon is similar to that of the farm-raised samples, indicating that all salmon, farmed or wild, are ultimately getting their dose of PBDEs from similar sources. The absolute PBDE dose is much higher for the farmed relative to the wild salmon. Decabromodiphenyl ether (BDE-209) was not included in the total PBDE concentrations reported above because it was not detected (LOD < 0.1 ng/g wet wt) in the salmon samples. BDE-209 was more prevalent in the feed samples, averaging 15 ± 5% of the total PBDEs.

Risk-based thresholds for PBDEs have not been established by the World Health Organization, the U.S. EPA, the U.S. FDA, or other regulatory and public health agencies. The European Parliament took action to ban penta-, octa-, and deca-BDEs in September 2001, with the agreement modified subsequently to ban penta- and octa-BDEs by August 2004 (19). Debate on the regulatory fate of PBDEs continues elsewhere because the toxicological database and literature are incomplete (20).

Highly brominated PBDEs are metabolically active compounds that induce hepatic cytochrome P450 1IB1 and IA1 (21, 22) and have weak or moderate binding affinity to the Ah receptor (23). PBDEs disrupt spontaneous behavior, impair learning and memory, and induce other neurotoxic effects in adult mice exposed during neonatal life (24). These effects occur in both a dose/response and time/response fashion, can worsen with age, and may be inducible during relatively short but critical periods of neonatal development (24–27). PBDEs are also endocrine disruptors, altering thyroid hormone homeostasis and causing a dose-dependent depletion of T4 (28), and PBDEs are weak agonists of estrogen receptors (both ERα and ERβ), an effect that may be enhanced by in vivo metabolism (29).

PBDEs have not been demonstrated to be carcinogenic in rodent bioassays. BDE-99, one of the few congeners tested, was negative in bacterial mutagenicity assays, did not cause structural chromosome aberrations, and showed no signs of cytotoxicity (30). However, some less-brominated congeners have induced genetic recombination in mammalian cells (31), and some concern for PBDE carcinogenesis continues to be raised (32).

Our data indicate that frequent consumption of farmed salmon and wild Pacific Chinook salmon will increase human dietary exposure to PBDEs much more so than consumption of most other wild Pacific salmon. It has been suggested that PBDE concentrations now observed in humans may leave little or no margin of safety (33); thus, prudent public health practice argues for the selective consumption of food, including many wild salmon species that contain comparatively lower concentrations of PBDEs as well as lower concentrations of many chlorinated organic contaminants (6). Salmon with lower PBDE concentrations still contain omega-3 fatty acids, and associated health benefits can be accrued by consumption of these fish with commensurately lower, contaminant-associated health risks.

This study demonstrates the importance of labeling salmon as farmed and identifying the country of origin. Additional studies of contaminant sources, particularly in feeds used for farmed, carnivorous species such as salmon, are needed to inform efforts to develop less contaminated feeds and food products. This conclusion is consistent with the recommendations of a recent panel of the Institute of Medicine (34), which called for major efforts to reduce the content of dioxin-like compounds in food given to animals and fish that are used for human consumption.

Acknowledgments

This research was initiated and supported by the Environmental Division of the Pew Charitable Trusts. We thank Amy Mathews Amos of Turnstone Consulting for superb project management and Suzanne Burrows for help obtaining the samples.

Note Added after ASAP Posting

This paper was released ASAP on August 10, 2004. Two sentences were added at the end of the fifth paragraph in the Results and Discussion section, and the corrected version was posted on August 27, 2004.

Literature Cited


Received for review March 24, 2004. Revised manuscript received July 7, 2004. Accepted July 19, 2004.

ES049548M