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Organochlorine Compounds in Lake Superior: Chiral Polychlorinated Biphenyls and Biotransformation in the Aquatic Food Web

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The enantiomeric composition of seven chiral PCB congeners was measured in the Lake Superior aquatic food web sampled in 1998, to determine the extent of enantioselective biotransformation in aquatic biota. All chiral PCB congeners studied (CBs 91, 95, 136, 149, 174, 176, and 183) biomagnified in the Lake Superior aquatic food web, based on biomagnification and food web magnification factors greater than unity. PCB atropisomers were racemic in phytoplankton and zooplankton, suggesting no biotransformation potential toward PCBs for these low trophic level organisms. However, *Diporeia* and mysids had significantly nonracemic residues for most chiral congeners studied. This observation suggests that these macrozooplankton can stereoselectively metabolize chiral congeners. Alternatively, macrozooplankton obtained nonracemic residues from feeding on organic-rich suspended particles and sediments, which would imply that stereoselective microbial PCB biotransformation may be occurring in Lake Superior sediments at PCB concentrations far lower than that previously associated with such activity. Widely nonracemic PCB residues in forage fish (lake herring, rainbow smelt, and slimy sculpin) and lake trout suggest a combination of both in vivo biotransformation and uptake of nonracemic residues from prey for these species. Minimum biotransformation rates, calculated from enantiomer mass balances between predators and prey, suggest metabolic half-lives on the order of 8 yr for CB 136 in lake trout and 2.6 yr for CB 95 in sculpins. This result suggests that significant biotransformation may occur for metabolizable PCB congeners over the lifespan of these biota. This study highlights the potential of chiral analysis to study biotransformation processes in food webs.

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Introduction

Polychlorinated biphenyls (PCBs) are persistent, bioaccumulative compounds that remain a potential health hazard in aquatic ecosystems long after they are used. Although PCB concentrations dropped dramatically in the years after they were banned (*1*−*3*), these decreases have leveled off after the mid-1980s or so (*4*−*6*). PCB concentrations may be affected by many factors. Nonpoint sources of contamination, such as atmospheric deposition and benthic recycling of sediment-sorbed contaminants (*7*, *8*), may maintain concentrations in the aquatic food web due to chemical cycling among environmental compartments. In aquatic biota, parameters such as lipid content (*8*, *9*), trophic position (*10*, *11*), changes in diet (*4*, *10*, *12*) and food web length (*13*) may also affect contaminant concentrations. These many competing factors make it very difficult to determine long term declines in contaminants in aquatic ecosystems. For example, Stow et al. (*4*) noted that thousands of individual fish samples may be needed to detect current changes in PCB levels due to high variability in individual specimens. Given the recalcitrance of PCBs, it is likely that biotransformation is a major long-term ultimate removal process for these compounds. Therefore, difficulties in determining long-term PCB trends are due in part to the lack of a tracer for biological degradation that is also unaffected by abiotic processes, crucial for understanding long-term environmental fate of persistent compounds.

Chiral pollutants exist as pairs of mirror-image enantiomers that have identical physical−chemical properties and abiotic degradation rates but may have different biological and toxicological properties and therefore different biotransformation rates. Enantiomer analysis has been used recently to study aquatic food web biotransformation dynamics of α-HCH, chiral chlordane compounds, and toxaphene congeners (*14*−*18*). However, to our knowledge no such study has been done for the 19 stable chiral PCB congeners despite recent reports of nonracemic amounts of these atropisomers (also referred to as enantiomers in this paper) in a number of aquatic species suggesting biotransformation (*19*−*21*). Many of these reports have been on single species or with incomplete food webs, making it difficult to determine how much of the nonracemic residues observed were due to in vivo biotransformation and how much were due to uptake from prey.

In addition, previous analysis of chiral contaminants in aquatic food webs have focused on food webs that included mammals or birds, which generally have higher capability to biotransform persistent contaminants (*22*). Lower aquatic species, such as fish and invertebrates, are generally considered to have poor biotransformation capability toward persistent xenobiotic compounds due to lower levels and activities of cytochrome P-450 1A and 2B isozymes (*23*, *24*). Observations that fish can eliminate some chiral contaminants stereoselectively (*25*, *26*) suggest that these lower organisms may have greater capability to biodegrade "persistent" pollutants than previously thought. Thus, chiral analysis can be a useful tool to understand contaminant dynamics in food webs.

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In this paper, we report the enantiomer composition of several chiral PCB congeners in the Lake Superior aquatic food web. This information in conjunction with achiral PCB measurements and nitrogen and carbon stable isotope analysis of biota is used to determine the extent of stereoselective biotransformation by aquatic biota, to apportion enantiomer signatures in biota between in vivo metabolism and uptake from prey, and to estimate biotransformation rates in the aquatic food web.

Experimental Section

Samples from the Lake Superior aquatic food web were collected from the Apostle Islands area in the western part of the lake in summer 1998 (*27*) from the R/V *Lake Guardian*, R/V *Siscowet*, and CCGS *Limnos*: water, phytoplankton, bulk zooplankton (>102 μm), *Mysis relicta*, amphipods (*Diporeia hoyi*), lake herring (*Coregonus artedii*), rainbow smelt (*Osmerus mordax*), slimy sculpin (*Cottus cognatus*), and lake trout (*Salvelinus namaycush*). Individual lake trout were extracted separately, while forage fish (FF) samples were composites of 8 individuals each for smelt and herring and 6 for sculpin. Mysids and *Diporeia* were composites with 6.5−17 g total wet weight. Details on extraction and achiral analysis by gas chromatography with electron capture detection are described elsewhere (*28*). A subset of samples that had been analyzed for toxaphene enantiomers (*27*) were also analyzed for enantiomers of chiral chlorobiphenyls (CBs) 91, 95, 136, 149, 174, 176, and 183 by chiral gas chromatography/mass spectrometry (GC/MS) using methods described elsewhere (*29, 30*). Water samples were not analyzed for chiral PCBs as concentrations were low enough ($\sum$PCB = 0.45 ng/L; *27*) that individual enantiomers were unquantifiable by GC/MS. Lake herring samples from the 1998 study were not available for chiral PCB analysis, so lake herring samples from a 1993−1996 study of toxaphene in eastern Lake Superior (*12*) were analyzed for chiral PCBs instead.

Chiral PCB compositions are expressed as enantiomer fractions (EFs) (*31*), defined as

$$\mathrm{EF} = \frac{A}{A+B} \qquad (1)$$

where A and B are the (+) and (−) enantiomer concentrations, respectively, for CB 136, 149, 174, and 176 and the first-eluting (E1) and second-eluting (E2) enantiomers on Chirasil-Dex for CBs 91 and 95 (*19*) and on BGB-172 BSCD for CB 183 (*29*). EFs of racemic standards were between 0.496 ± 0.004 and 0.505 ± 0.008 (σ) for all congeners. Enantiomer separation and precision of EFs was verified by periodic analysis of chiral PCBs in standardized reference materials (*29*). A conservative measure of EF precision of 0.032 (95% confidence) based on EF measurements on different chiral columns for this study was used for significance testing (*20*) of samples as compared to racemic standards. EF distributions among different species were compared by using analysis of variance (ANOVA) on data ranks, with Tukey honestly significant difference tests run on all significantly different groups (overall $P < 0.05$).

Results

PCB concentrations increased with trophic level as expected (Table 1). A detailed discussion of achiral PCB food web dynamics is presented elsewhere (*32*). There were no statistical differences in total PCB concentrations between the subset of samples reanalyzed for enantiomers and the full sample set for all species. A comparison of lake herring PCB concentrations and congener profiles between the 1993−1996 study (analyzed for chiral PCBs) and the 1998 herring showed no differences.

EF profiles (Figure 1) indicated racemic amounts of chiral

TABLE 1. Mean Lipid Content, δ^{13}C (‰), δ^{15}N (‰), and Concentrations and Standard Deviations (ng/g lipid) of Total PCBs, CB 153, and Chiral Congeners[a] in Lake Superior Biota Analyzed in This Study[b]

species	*n*	% lipid	δ^{13}C	δ^{15}N	$\sum$PCB	153	91	95	136	149	174	176	183
trout	29	11.8 ± 4.8	−25.6 ± 1.6	9.9 ± 1.0	5440 ± 3870	680 ± 560	17 ± 14	55 ± 34	20 ± 13	120 ± 80	42 ± 28	42 ± 66	55 ± 65
herring	8	4.8	na[c]	na[d]	2190 ± 600	150 ± 38	7.5 ± 2.7	45 ± 15 [f]	5.9 ± 2.5	71 ± 23[g]	23 ± 7	23 ± 7[h]	21 ± 8
smelt	5	4.7 ± 0.4	−24.4 ± 0.3	6.1 ± 0.7	1120 ± 740	130 ± 120	4.1 ± 2.2	14 ± 5	2.9 ± 0.9	40 ± 27	13 ± 9	2.5 ± 2.9	15 ± 14
sculpin	6	4.7 ± 1.8	−23.5 ± 1.6	7.4 ± 0.6	1000 ± 210	120 ± 30	2.6 ± 1.0	17 ± 6	3.4 ± 0.8	12 ± 2	5.3 ± 0.5	1.3 ± 0.4	14 ± 4
Mysis	3	4.9 ± 1.0	−28.8 ± 2.7	5.9 ± 0.1	280 ± 40	30 ± 2	1.2 ± 0.1	2.7 ± 0.1	0.5 ± 0.1	9.5 ± 0.7	2.7 ± 0.1	0.1 ± 0.1	3.8 ± 0.3
Diporeia	3	4.3 ± 1.5	na	na	250 ± 120	19 ± 8	1.1 ± 0.6	4.7 ± 3.2	1.1 ± 0.7	8.1 ± 3.9	3.1 ± 1.7	0.3 ± 0.3	2.4 ± 1.0
zoo	5	4.2 ± 1.5	−30.5 ± 0.7	4.8 ± 1.7	370 ± 290	21 ± 19	3.2 ± 2.2	13 ± 8	2.2 ± 1.5	15 ± 11	4.2 ± 4.1	0.7 ± 0.4	2.8 ± 3.0
phyto[e]	2		−27.9	2.3	480 ± 170	23 ± 10	3.1 ± 1.8	18 ± 10	2.5 ± 1.0	24 ± 6	5.2 ± 4.0	1.0 ± 0.3	3.3 ± 2.6

[a] Sum of (+) and (−) enantiomers for CBs 91, 95, 136, 149, 174, 176, and 183. [b] n = number of samples, na = not analyzed. [c] δ^{13}C in herring from 1998 study was −24.8‰. [d] δ^{15}N in herring from 1998 study was 5.6‰. [e] Concentrations in phytoplankton are in ng/g dry weight. [f] Sum of congeners 95 + 66. [g] Sum of congeners 149 + 133. [h] Sum of congeners 176 + 130.

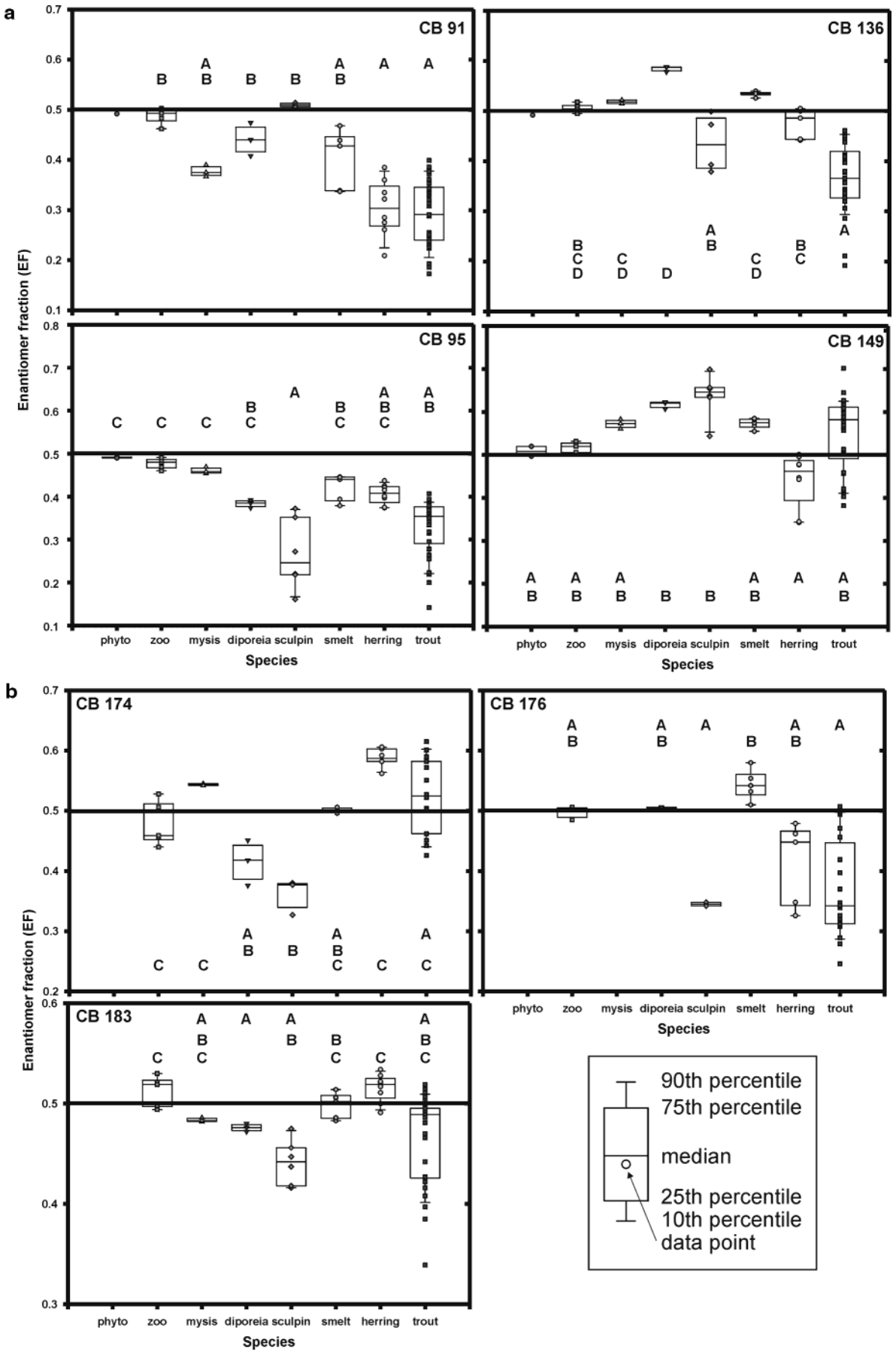

FIGURE 1. Box plots of enantiomer fractions (EFs) for chiral penta- and hexachlorobiphenyls (subfigure a) and heptachlorobiphenyls (subfigure b) measured in Lake Superior biota. The horizontal line across each panel indicates racemic EF of 0.5. "Phyto" = phytoplankton, "Zoo" = zooplankton. Distributions with the same letter are not statistically different.

PCBs in lower trophic level organisms and significantly nonracemic amounts in higher species. CB 176 concentrations were low in most species, so EFs were not quantifiable in phytoplankton and mysids. Likewise, EFs for the three chiral heptachlorobiphenyls analyzed were not measurable in phytoplankton. EFs were racemic in all phytoplankton and bulk zooplankton samples, indicating no stereoselective bioprocessing in these species. On the other hand, EFs for the macroplankton were significantly nonracemic for a number of congeners: CBs 91, 149, and 174 for mysids and CBs 91, 95, 136, 149, and 174 for *Diporeia*. The EF patterns were markedly different between the two species for several congeners and were reversed for CB 174 with (+)-CB 174 prevalent in mysids and (−)-CB 174 in *Diporeia*.

All four of the fish species had widely nonracemic residues of chiral PCB congeners. Depletion of E1-CB 91 was evident in all fish except sculpin, which was racemic. These results are consistent with previous reports in other fish species in U.S. waters (*20*) except for sculpin, which were enriched in E1-CB 91. E1-CB 95 was depleted in all species. CB 136 was mostly racemic in smelt and herring but depleted in (+)-CB 136 in sculpins and lake trout. This depletion is consistent with the EFs < 0.5 observed in bass, bluegill, suckers, and sculpins in U.S. fish (*20*). CB 149 was nonracemic but in different directions, with the (+) enantiomer prevalent in sculpin, smelt, and some of the lake trout population and (−) in herring and the rest of the lake trout. These mixed results are in keeping with other field measurements on chiral PCBs in fish (*26*), in which (+)-CB 149 dominated in sculpins, while CB 149 in bluegill populations resembled those of lake trout in this study.

Most species had racemic amounts of CB 183, but EFs are <0.5 for sculpin and trout (average EFs = 0.442 and 0.466, respectively). The EF direction is opposite to that observed for sculpins from U.S. streams (average EF = 0.55) (*20*) but is the same magnitude as those in this study if the elution order for CB 183 enantiomers is reversed between the BGB-172 BSCD column and the B-PH column used in the earlier study.

There was a significant (all $P < 0.05$ in this discussion) but weak correlation between lake trout age and EF for CB 91 ($r^2 = 0.14$) as well as correlations between lake trout lipid-normalized congener concentrations and EFs for CBs 91, 136, and 149 ($r^2 = 0.35$, 0.40, and 0.16, respectively). In addition, correlations between recalcitrant CB 153 lipid-normalized concentrations in trout and EFs were significant for CBs 91, 95, 136, and 176 (Figure 2), suggesting that trout with heavier PCB contamination also had higher amounts of biotransformation of these chiral congeners. There were no significant correlations between lake trout weight and EFs for any chiral congener. Biomagnification factors (BMFs) for individual enantiomers of a congener differed from each other and from the sum total but were greater than unity in all cases (Table 2), indicating that enantiomers were biomagnifying in the aquatic food web. This observation is supported by the food web magnification factors (FWMFs), which did not differ much among enantiomers of a congener (Table 2) and were greater than unity except for CB 176.

Discussion

Achiral Biotransformation Indicators versus EFs. The difficulty in using models based on achiral analysis to detect PCB biotransformation in field studies is illustrated by the relative concentration ratios of chiral congeners in Lake Superior trout (Figure 3). The concentration of a compound (C) as compared to a recalcitrant contaminant, such as CB 153 (C_{CB153}), should be less than unity if the compound is biotransformed and greater than unity if it is bioaccumulated and not biotransformed. This amount can be expressed as relative ratios (RR) (*22*, *33*), which have been widely used to

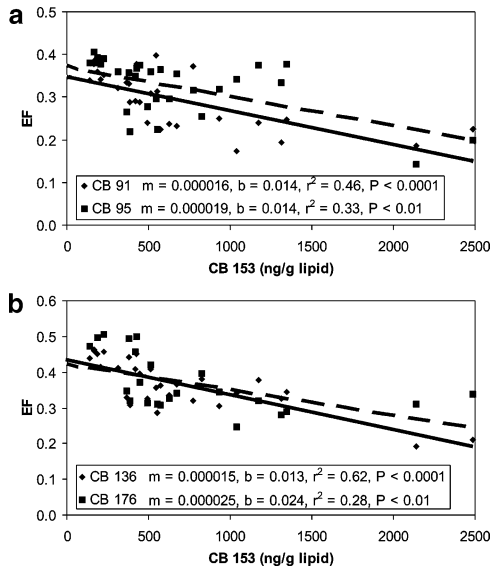

FIGURE 2. Lipid-normalized CB 153 concentrations vs EFs of CBs 91 and 95 (panel a) and CBs 136 and 176 (panel b) in lake trout. Regression lines (solid for CBs 91 and 136, dashed for CBs 95 and 176) include slope (*m*), *x*-intercept (*b*), coefficient of variation (r^2), and *P* value for slope.

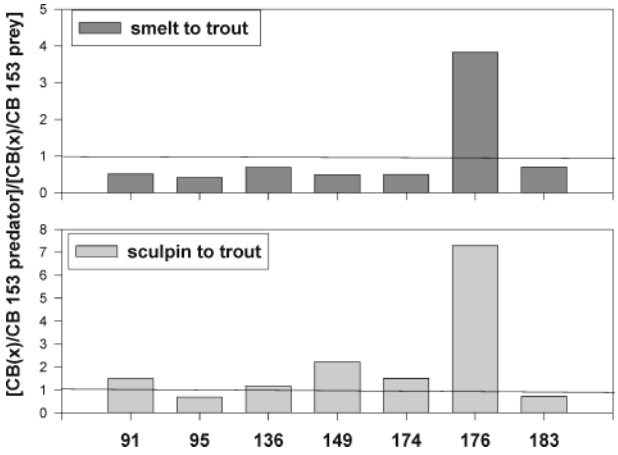

FIGURE 3. Relative concentration ratios of chiral PCB congeners (sum of both enantiomers) in Lake Superior lake trout.

infer field biotransformation of persistent xenobiotic compounds:

$$\mathrm{RR} = \frac{\dfrac{C}{\mathrm{CB\ 153_{predator}}}}{\dfrac{C}{\mathrm{CB\ 153_{prey}}}} \qquad (2)$$

The RR values for the chiral congeners measured in this study are mostly less than unity for trout (predator) consuming rainbow smelt (prey) but are mostly larger than unity for trout−slimy sculpin (Figure 3). High values for CB 176 are due largely to low concentrations of both enantiomers in all species. There are two explanations for this discrepancy. First, the calculated RR values are for trout solely consuming a single species. In reality, lake trout in Lake Superior will consume a variety of prey. The second reason is that RR values infer biotransformation but do not actually detect or measure it and are influenced by competing nonmetabolic mechanisms. For example, if a compound is being biotransformed by an organism at a rate slower than that of uptake by prey, then RR values will be larger than unity and will miss the presence of biotransformation activity.

TABLE 2. Biomagnification (BMF) and Food Web Biomagnification Factors (FWMF) for Chiral PCBs in the Lake Superior Aquatic Food Web

			biomagnification factors (BMF)						
	slope[a]	FWMF[b]	trout herring	trout smelt	trout sculpin	trout FF[c]	sculpin *Diporeia*	herring zoo	herring *Mysis*
E1-CB 91	0.24 ± 0.11 (0.40)	1.3	1.9	2.8	4.3	2.7	2.1	1.5	5.3
E2-CB 91	0.37 ± 0.13 (0.54)	1.5	2.4	4.8	12.7	4.2	1.7	3.2	7.2
(±)-CB 91	0.32 ± 0.12 (0.51)	1.4	2.2	4.0	6.4	3.5	2.4	2.4	6.5
E1-CB 95	0.30 ± 0.11 (0.51)	1.4	1.0	2.9	3.7	1.8	2.6	2.9	14.9
E2-CB 95	0.40 ± 0.11 (0.63)	1.5	1.4	4.4	3.1	2.4	4.2	3.8	18.6
(±)-CB 95	0.37 ± 0.10 (0.61)	1.4	1.2	3.8	3.3	2.2	3.6	3.4	16.9
(+)-CB 136	0.37 ± 0.10 (0.65)	1.4	2.4	4.3	5.0	3.5	2.1	2.5	9.7
(−)-CB 136	0.48 ± 0.12 (0.69)	1.6	4.2	9.6	7.5	6.3	3.8	2.9	11.6
(±)-CB 136	0.42 ± 0.11 (0.67)	1.5	3.4	6.7	5.9	4.9	3.1	2.7	10.8
(+)-CB 149	0.42 ± 0.11 (0.63)	1.5	2.1	2.9	8.8	3.2	1.5	4.1	5.8
(−)-CB 149	0.42 ± 0.12 (0.59)	1.5	1.5	3.4	13.3	2.9	1.4	5.5	9.8
(±)-CB 149	0.42 ± 0.12 (0.62)	1.5	1.7	3.1	10.4	3.0	1.5	4.8	7.5
(+)-CB 174	0.53 ± 0.14 (0.79)	1.7	2.2	8.5	16.2	4.7	1.5	6.7	9.3
(−)-CB 174	0.51 ± 0.14 (0.78)	1.7	2.9	7.8	8.4	5.0	1.8	4.6	7.7
(±)-CB 174	0.53 ± 0.14 (0.79)	1.7	1.8	3.1	7.8	3.0	1.7	5.4	8.4
(+)-CB 176	−0.43 ± 0.41 (0.16)	0.7	1.3	10.1	28.9	3.2	2.4	36.2	–
(−)-CB 176	−0.33 ± 0.37 (0.12)	0.7	1.8	25.1	30.7	4.8	4.6	50.6	–
(±)-CB 176	−0.37 ± 0.39 (0.14)	0.7	1.8	16.6	32.3	4.6	4.3	36.0	221.8
E1-CB 183	0.52 ± 0.13 (0.68)	1.7	2.3	3.2	4.1	3.0	5.3	7.6	6.0
E2-CB 183	0.57 ± 0.14 (0.69)	1.8	3.0	4.0	3.9	3.5	6.2	7.3	5.3
(±)-CB 183	0.56 ± 0.14 (0.69)	1.8	2.6	3.6	4.0	3.3	5.8	7.4	5.6
CB 153	0.58 ± 0.13 (0.72)	1.8	4.7	5.3	5.7	5.2	6.3	6.9	4.8
∑PCB	0.47 ± 0.11 (0.69)	1.6	2.5	4.8	5.5	3.8	3.9	5.9	7.9

[a] Slope (with 95% confidence interval) calculated from the model ln concentration = $a + (b \times {}^{15}N)$, excluding phytoplankton and species for which no ^{15}N was analyzed in 1998 sampling (lake herring, *Diporeia*). Coefficient of determination (r^2) shown in parentheses. All concentrations were lipid corrected. [b] FWMF = e^b (see ref *17*). [c] BMF for trout/forage fish (FF) is average concentration in lake trout divided by average concentration in forage fish (lake herring, smelt, and slimy sculpin).

These considerations affect all other such achiral-based techniques to infer biotransformation, such as lower measured biomagnification factors (BMFs), and lower retention of congeners expected to be metabolized based on structure−activity relations (*22*, *33*−*35*). None of these methods specifically trace stereoselective biological processes (e.g., biotransformation) without being influenced by nonstereoselective processes (i.e., nonmetabolic ones), so they do not provide conclusive evidence either for or against in vivo biotransformation. For example, BMFs for lake trout for chiral PCBs were all larger than unity (Table 2), indicating that these congeners were being accumulated despite biotransformation activity as evidenced by nonracemic EFs. This is consistent with laboratory observations of stereoselective CB elimination of bioaccumulative chiral congeners (*26*) as well as high BMFs for nonracemic PCBs in bowhead whales (*21*).

Chiral PCB EFs in Planktonic Species. The racemic amounts of all chiral PCBs observed in Lake Superior phytoplankon and bulk zooplankton suggest that these low trophic level organisms have negligible capability to biotransform persistent organic contaminants. This observation agrees with racemic PCBs measured in *Calanus* zooplankton, a major food source of cetaceans in the Arctic (*21*). The plankton samples in our Lake Superior study were composites of numerous individuals and species (e.g., copepods, calanoids, and cladocerans for zooplankton), so we cannot rule out the possibility that individual species may have nonracemic residues of chiral PCBs that are diluted by those of other organisms in the composites.

The nonracemic EFs in mysids and amphiphods is surprising and suggests two possible explanations. The first is that mysids and *Diporeia* have the capability to biotransform PCBs stereoselectively. Previous research on PCB bioaccumulation in mysids (*36*, *37*) and *Diporeia* (*38*) suggested that these organisms lacked metabolic capability toward PCBs. However, these studies focused on understanding PCB uptake and elimination kinetics (i.e., nonmetabolic processes) and therefore used recalcitrant CB 153 to eliminate the possibility of biotransformation from consideration. Radiolabeled parent compounds were also typically used, and the lack of radioactivity observed for polar extract fractions (*37*) was interpreted to suggest that the macrozooplankton did not produce metabolites. However, hydroxylated and methylsulfonyl PCBs, which are more polar than the parent compounds, may themselves still be hydrophobic and persistent. For example, 4-OH-CB 187, which has been observed at picogram per gram concentrations in Lake Superior lake trout blood plasma (*39*), has a log K_{ow} of 5.2 calculated from fragment constant methods (*40*). This value is in the range of biomagnifying compounds (*41*), and the heavy chlorination of this hydroxylated CB makes biotransformation of it unlikely. Many methylsulfonyl PCBs are known to be persistent in biota (*42*). So it may be possible that mysids and *Diporeia* are biotransforming chiral PCBs enantioselectively in Lake Superior.

An alternative explanation is that macrozooplankton had taken up nonracemic chiral PCBs from food sources. Mysids are omnivorous feeders, consuming zooplankton, amphipods, phytoplankton, sediment detritus, other mysids, chironomids, and fish larvae (*43*, *44*). *Diporeia* feed primarily on organic carbon in fine-grained particulate matter in the water column and sediments (*45*). Both macrozooplankton are exposed to sediments and fresh sedimentary particles settling through the water column. Sediments are the repository for much of the PCB burden in Lake Superior (*46*). If these organisms are obtaining nonracemic PCBs from their food sources, then stereoselective biotransformation of PCBs in those sediment and particulate organic carbon food sources mediated by microorganisms is likely in order to produce such enantiomer preferences.

Microbial biotransformation of PCBs under both aerobic (*47*) and anaerobic (*48*) conditions is well-studied, and at least some of the pathways for both mechanisms are

stereoselective (*19*, *49*). However, achiral studies suggest that microbial PCB biotransformation is not active to any appreciable degree below concentrations of about 1 μg/g in sediments (*50*). If our alternative hypothesis is true, then stereoselective microbial biotransformation of PCBs has taken place at concentrations far below this suggested cutoff point in Lake Superior. Typical concentrations of total PCBs in Lake Superior are 1−5 ng/g in sediments (*46*) and 5−10 ng/g in the benthic nepheloid layer (*51*). Thus, microbes may possibly biotransform PCBs at low concentrations at rates that are difficult to measure by achiral means but are readily detected by chiral analysis.

Unfortunately, we were unable to measure enantiomer compositions of chiral PCBs in Lake Superior sediments sampled in the 1998 cruise as concentrations were too low. For similar reasons, we also cannot rule out the possibility that the zooplankton accumulated dissolved-phase nonracemic residues produced by microbial activity. This latter mechanism explained similar nonracemic α-HCH signatures between water and plankton in the Northwater Polynya (*17*). Thus, we cannot distinguish between the various possible explanations for nonracemic PCBs in mysids and *Diporeia*. More research is needed to understand pharmacokinetics of PCBs in low trophic level organisms. *Diporeia* and bulk zooplankton also had nonracemic residues of chiral toxaphene congeners (*27*), indicating that the processes responsible for nonracemic PCBs in zooplankton may also be acting on toxaphene as well.

Chiral PCB EFs in Forage Fish. The presence of nonracemic chiral PCBs in sculpins compared to their prey suggests these fish are stereoselectively biotransforming some congeners. Over 93% of the diet of slimy sculpins in Lake Superior consisted of *Diporeia*, with mysids making up most of the remaining fraction (*52*). However, the EFs of CBs 95 and 136 in slimy sculpin were significantly different from those of *Diporeia* (Figure 1). If the diet of the predator is well-characterized, then the most likely mechanism by which EFs can change between predator and prey is enantioselective biotransformation (*21*, *25*, *26*). Although mysids made up much of the diet of slimy sculpins in parts of the Apostle Islands region (*53*), the EFs for these congeners for mysids are also similar to those of *Diporeia* and different from those of slimy sculpins (Figure 1) so they do not change our suggestion that sculpins have stereoselectively biotransformed CBs 95 and 136. Our results are consistent with literature reports of methylsulfonyl PCB metabolites present in saltwater (*54*) and freshwater (*55*) sculpins, including those of chiral CBs 132, 149, and 174. Nonracemic PCBs in freshwater sculpins in U.S. rivers have also been observed (*20*), supporting our suggestion of stereoselective biotransformation by sculpin. Because sculpins are benthic fish and are constantly exposed to sediment that can be ingested during feeding, they may have evolved the metabolic capability to eliminate xenobiotic compounds found in sediments and are thus able to transform anthropogenic contaminants such as PCBs.

The EF signatures in lake herring suggest both in vivo metabolism of some PCB congeners and uptake from prey of others. EFs for CBs 91 in lake herring were significantly different from those of their prey (Figure 1), which in Lake Superior consists mostly of zooplankton (copepods and cladocerans) and minor amounts of mysids (*56*). As with the case for sculpins, this observation suggests in vivo enantioselective metabolism of these congeners by lake herring and indicates that PCB metabolic capacity in fish may extend to lower trophic species. The EFs of other congeners were within the range of EFs in the prey species, suggesting uptake of such residues.

Rainbow smelt have nonracemic EFs of chiral PCBs as well. But unlike the other forage fish species, none were significantly different than EFs in their prey (Figure 1), which consists mainly of mysids and copepods with some cladocerans, *Diporeia*, and small fish (*56*). Unlike sculpins and lake herring, there is no evidence that rainbow smelt have any metabolic capability to biotransform PCBs, as nonracemic residues appear to be due to uptake from prey. However, we cannot rule out the possibility that smelt may be enantioselectively biotransforming chiral PCBs within the range of EFs observed in their prey.

Chiral PCB EFs in Lake Trout. The chiral PCB compositions in lake trout are likely due to a combination of uptake of nonracemic residues from prey and in vivo metabolism. For CBs 91, 95, 149, and 174, the EFs were nonracemic in lake trout but were not statistically different from those of the various major prey species (Figure 1). The diet of lean lake trout in Lake Superior consists largely of coregonids (including lake herring) and rainbow smelt, with lesser amounts of sculpins (*57*, *58*). This general observation holds despite changes in relative proportions of coregonids and smelt consumed, from factors such as predator age (*59*), forage fish population changes over time (*12*, *60*−*62*), and whether trout feed nearshore (i.e., lean lake trout) or offshore (i.e., siscowet lake trout) (*57*, *58*, *63*). The nonracemic amounts of these congeners in lake trout are probably due to consumption of nonracemic residues from prey such as the forage fish species, which may have themselves enantioselectively metabolized the congeners, as we have discussed earlier. Our field results for lake trout are also consistent with laboratory studies showing that rainbow trout, a closely related salmonid, did not eliminate CB 95 enantioselectively (*26*).

In contrast, CB 136 EFs suggest enantioselective in vivo biotransformation of this congener by lake trout, as the EFs were significantly different than those of lake herring and smelt (Figure 1). Lake trout EFs for CB 136 were not statistically different than that of sculpin; however, this FF makes up only a minor part of the lean lake trout diet (*58*) to which most of our specimens belong. As with the case for slimy sculpins and their prey, the most likely explanation for the different CB 136 signatures in lake trout as compared to their prey is preferential metabolism of (+)-CB 136 over its opposite enantiomer. This suggestion is consistent with laboratory studies showing that rainbow trout eliminated (+)-CB 136 preferentially (*26*).

The reversals in CB 149 enantiomer preferences in lake trout may be due to changes in diet composition for this species in Lake Superior. (+)-CB 149 is enriched in sculpins and smelt, whereas (−)-CB 149 is enriched in lake herring (Figure 1). This observation suggests that those lake trout with more (+)-CB 149 may have fed more on sculpins and smelt and those with more (−)-CB 149 on lake herring. Whittle et al. (*12*) observed changes in toxaphene congener distributions in Lake Superior lake trout over time, which they attributed to shifts in lake trout diet. The population of lake herring in Lake Superior rapidly increased during 1984−1990, while rainbow smelt populations generally declined. This shift is likely reflected in the congener distribution changes (*12*), and perhaps as well in the EFs of CB 149. However, the lack of correlation between CB 149 EFs and age indicate that other factors are also important in determining CB 149 enantiomer composition in Lake Superior lake trout.

The nonracemic presence of CB 183 in some lake trout and in sculpins (Figure 1) is also surprising. This congener (2,2′,3,4,4′,5′,6-heptachlorobiphenyl) has no vicinal hydrogen atoms. PCB congeners with vicinal meta, para hydrogen atoms are more suspectible to attack by cytochrome P450 2B isozymes (*22*, *34*), so CB 183, much like CB 153, should be recalcitrant. However, it is clear that this congener was enantioselectively bioprocessed somewhere in the Lake

TABLE 3. Examples of Enantiomer Mass Balances between Predators and Prey in Lake Superior[a]

				CB 91		CB 95		CB 136		CB 174	
predator	**prey**	f_i		***C***	**EF**	***C***	**EF**	***C***	**EF**	***C***	**EF**
sculpin[b]	*Diporeia*	0.93				4.7	0.383	3.4	0.436	5.3	0.361
	Mysis	0.03				2.7	0.460	0.5	0.518	2.7	0.543
			EF_{mb}				0.385		0.583		0.418
			mean EF				0.286		0.436		0.361
			predator age (yr)[c]				2		2		2
			$k_{m,min}$ (yr^{-1})				0.27		0.297		0.120
			half-life (yr)				2.6		2.3		5.8
herring[d]	zooplank.	0.85		3.2	0.489						
	Mysis	0.15		1.2	0.377						
			EF_{mb}		0.482						
			mean EF		0.299						
			predator age (yr)[e]		8						
			$k_{m,min}$ (yr^{-1})		0.097						
			half-life (yr)		7.1						
lake trout[f]	smelt	0.71						2.9	0.534		
	herring	0.15						5.9	0.476		
	sculpin	0.02						3.4	0.436		
	mysis	0.01						0.5	0.518		
	zooplankton	0.01						2.2	0.505		
	other[g]	0.1						2.2	0.505		
			^{EF}mb						0.515		
			mean EF						0.367		
			predator age (yr)[h]						6.9		
			$k_{m,min}$ (yr^{-1})						0.088		
			half-life (yr)						7.9		
lake trout[i]	smelt	0.71						2.9	0.534		
	herring	0.15						5.9	0.476		
	sculpin	0.02						3.4	0.436		
	mysis	0.01						0.5	0.518		
	zooplankton	0.01						2.2	0.505		
			^{EF}mb						0.514		
			mean EF						0.367		
			predator age (yr)[h]						6.9		
			$k_{m,min}$ (yr^{-1})						0.087		
			half-life (yr)						8.0		

[a] f_i = fraction of prey in predator's diet in Lake Superior. *C* and EF are mean concentrations (ng/g lipid) and EFs in prey species for respective congeners. $EF_{mb} = EF_{predator,mass\ bal}$ from eq 6. Mean EF = mean of measured EF in predator. Mass balances are calculated only for cases in which EFs of predators differ significantly from its dominant prey species (Figure 1). [b] f_i from ref *52*. Remaining 4% of diet consists of chironomids and is ignored in these calculations. [c] Based on age−length relationship from ref *52* and mean length of 58 ± 13 mm (σ) of sculpins in this study. [d] f_i from ref *56*. [e] Based on maximum lifespan of 8 yr for herring. [f] f_i from ref *56*. [g] "other" prey consists of other fish and invertebrates and is assumed to have same CB 136 concentration and EF as zooplankton. [h] Based on otolith measurements from this study. [i] f_i from ref *56* and ignoring remaining 10% of diet contribution from "other" prey.

Superior food web. This observation suggests that there may be a number of biochemical pathways aside from P450 2B that can degrade CB 183 present in biota. This hypothesis is supported by the presence of nonracemic amounts of CB 183 in freshwater bivalves (*20*) and bowhead whales (*21*), suggesting that PCB biotransformation is more complex than one would suspect without the insights given by chiral analysis. It is important to recognize, though, that we do not know which biochemical pathways are actually responsible for producing the EFs observed in this study based on our data.

The narrow distribution of many congener EFs in many species is consistent with the observation of fairly constant enantiomer signatures in field samples for many species and analytes (*25*, *64*). Vetter et al. (*25*) suggested that this constancy may be due to equilibrium between uptake and elimination processes, consistent with our suggestion that EFs in Lake Superior biota are due to combinations of uptake from prey and in vivo biotransformation.

Minimum Biotransformation Rates of PCBs by Lake Superior Biota. Estimates of minimum PCB biotransformation rates in Lake Superior biota are possible based on EF profiles and knowledge of predator−prey relations in the aquatic food web. If an organism is not stereoselectively bioprocessing a chiral contaminant, then the EF of that analyte in the organism ($EF_{predator,mass\ bal}$) should be a mass balance of the EFs (*31*) in each prey species *i* (EF_i), weighted to the analyte concentration in each prey species (C_i), the proportion of the analyte from each prey that is absorbed by the predator (abs_i), and the proportion of each prey species in the predator's diet (f_i):

$$EF_{predator,mass\ bal} = \frac{\sum(EF_i \cdot C_i \cdot abs_i \cdot f_i)}{\sum(C_i \cdot abs_i \cdot f_i)} \quad (3)$$

If the measured EF in the predator is different from that calculated from eq 3 and the predator's age is known, then the difference between the two can used to calculate minimum first-order biotransformation rates using the following equation for doing so based on changing EF profiles over time (*26*):

$$EF = \frac{1}{1 + \frac{(-)_o}{(+)_o} e^{(k_{m,+} - k_{m,-})t}} \quad (4)$$

where *t* is the predator's age; $k_{m,+}$ and $k_{m,-}$ are the biotransformation rate constants for the (+) and (−) enantiomers, respectively, in the predator; and $(+)_o$ and $(-)_o$ are the concentration of the (+) and (−) enantiomers, respectively,

in the predator if there were no biotransformation. The ratio of the concentrations is the enantiomer ratio (ER), which is related to EF (*31*):

$$\mathrm{EF} = \frac{\mathrm{ER}}{1 + \mathrm{ER}} \tag{5}$$

Neither $k_{m,+}$ nor $k_{m,-}$ are known, but a minimum value for the overall biotransformation rate constant ($k_{m,min}$) can be calculated by setting one of the enantiomer rates to zero (i.e., assuming EF changes are due solely to metabolism of one enantiomer with the other enantiomer unmetabolized; *26*). Taking these into account and rearranging eqs 4 and 5 yields

$$\mathrm{EF}_{\mathrm{predator,measured}} = \frac{1}{1 + \dfrac{1 - \mathrm{EF}_{\mathrm{predator,mass\ bal}}}{\mathrm{EF}_{\mathrm{predator,mass\ bal}}} e^{-k_{m,min}t}} \tag{6}$$

In calculating minimum biotransformation rates, abs_i must be determined for each prey species in the predator's diet. The amount of contaminant absorbed across the gastrointestinal tract depends on a number of factors, such as the prey's nutritional digestibility (*65*, *66*) and lipid content and the hydrophobicity and molecular size of the contaminant (*41*). This last factor is the same across all prey for a given chemical (i.e., hydrophobicity and molecular size for a given compound is the same regardless of whatever prey it is in). Therefore differences in chemical absorption in different prey would depend on factors relating to the prey organism. These factors appear to be closely related to the amount of lipid in the prey, as lipid-rich food may result in higher absorption of contaminants by predators (*66*). Because all the forage fish and planktonic species have fairly similar lipid contents (ca. 3−5%, Table 2), abs_i can be set equal for all prey as a first approximation. Therefore the term drops out of eq 3. Fish ages were the average of those measured in lake trout analyzed for chiral PCBs (6.9 ± 1.3 yr), the calculated average age for sculpins (2 yr) based on age−length relations (*52*) and the maximum age for lake herring (8 yr), as age was not determined for these fish and such values would represent conservative estimates. Predator diets (f_i) from Lake Superior were taken from published values from the 1980s (*52*, *56*). These proportions may have changed to some degree since those studies were done. In particular, lake trout appeared to have consumed greater proportions of lake herring starting in the mid- to late-1990s in response to the increase of this forage fish over rainbow smelt (*12*, *62*). However, our calculations serve as a useful illustration of the use of enantiomers to determine biotransformation rates in the field.

Example calculations of minimum PCB biotransformation rates (Table 3) suggest that chiral PCB congeners are metabolized slowly but measurably by Lake Superior fish. Biotransformation half-lives range from 2.6 yr for CB 95 in sculpin to 8 yr for CB 136 in lake trout (Table 3). These half-lives are slower than that measured for CB 136 elimination by rainbow trout in the laboratory of approximately 1 yr (*26*). However, the fish in that laboratory experiment received much higher CB concentrations in their diet (ppm levels) than fish in Lake Superior are likely to eat and were at higher temperatures (9 vs 4 °C, respectively). The shorter half-life for CB 136 in sculpins (2.3 yr, Table 3) as compared to trout (7.9 years) suggests that sculpins are more adept at metabolizing this congener than trout. The much longer half-life for CB 174 in sculpins (5.8 yr) than CB 95 and CB 136 (2.3 yr) may be due to the heavier chlorination of CB 174, which has only one set of vicinal meta, para hydrogen atoms available for CYP2B attack instead of two (as is the case for CBs 95 and 136).

It is important to recognize that, although these biotransformation rates are slow, a substantial amount of metabolizable PCBs in biota may be degraded over time. Lake trout can live to be over 20 years old, while slimy sculpins can live for 5 yr (*52*). These lifespans are several biotransformation half-lives. These rates are higher than the 10% total PCB metabolic loss estimated in sculpin based on production of measured methylsulfonyl CB metabolites and regression of the ratio of several metabolizable congeners to CB 153 (*55*). However, rates were not directly calculated in that study. Achiral literature studies have suggested that PCB congeners with structures amenable to biotransformation have shorter half-lives than more recalcitrant congeners (*35*, *67*, *68*). Our estimates suggest that much of the elimination of these congeners in biota may be due to biotransformation and not physical elimination. Nonracemic contaminants in dead organisms would thus be transported to the sediments, at least to some extent. This process may lead to pools of nonracemic analytes in sediments and the benthic nepheloid layer, both of which are important in the cycling of organic pollutants in Lake Superior (*51*). We were unable to determine PCB enantiomer compositions in Lake Superior water and sediment; such information is crucial in order to understand the extent and significance of biotransformation on contaminant cycling in aquatic ecosystems.

Our estimates of minimum biotransformation rates are necessarily somewhat crude, as they incorporate a number of parameters with some variability (e.g., concentrations, EFs, diet compositions, etc.); do not take into account changes in diet over the lifespan of the predator (e.g., young-of-the-year lake trout eat mostly zooplankton and mysids; *53*); or account for differences in metabolic capability due to age, sex, and other physiological factors. In the case of lake trout, there is also the fact that we have no PCB measurements for the 10% of the diet not sampled in this study (e.g., other forage fish, chironomids, insects, etc.). However, if this fraction is assumed to be similar to zooplankton in concentration and EFs, the calculated rates change only by a few percent (Table 3). Despite these complications, our PCB biotransformation rates are the first to be determined for fish in a field study to our knowledge. Most studies have implicitly or explicitly set biotransformation rates to zero (*69*, *70*). Those field studies with estimates of PCB biotransformation often calculate these rates by curve-fitting in modeling efforts (*71*) due to lack of available data. Despite these potential difficulties, our estimates highlight the utility of EFs to understand biological processes affecting chiral contaminants. There may be more biotransformation of persistent contaminants by aquatic biota than has been previously assumed.

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Literature Cited

(1) Borgmann, U.; Whittle, D. *J. Great Lakes Res.* **1991**, *17*, 368−381.

(2) Miller, M. A.; Madenjian, C. P.; Masnado, R. G. *J. Great Lakes Res.* **1992**, *18*, 742−754.
(3) DeVault, D. S.; Hesselberg, R.; Rodgers, P. W.; Feist, T. J. *J. Great Lakes Res.* **1996**, *22*, 884−895.
(4) Stow, C. A.; Carpenter, S. R.; Eby, L. A.; Amrhein, J. F.; Hesselberg, R. J. *Ecol. Appl.* **1995**, *5*, 248−260.
(5) Hebert, C. E.; Norstrom, R. J.; Weseloh, D. V. *Environ. Rev.* **1999**, *7*, 147−166.
(6) Braune, B. M.; Donaldson, G. M.; Hobson, K. A. *Environ. Pollut.* **2001**, *114*, 39−54.
(7) Baker, J. E.; Eisenreich, S. J.; Eadie, B. J. *Environ. Sci. Technol.* **1991**, *25*, 500−509.
(8) Kucklick, J. R.; Baker, J. E. *Environ. Sci. Technol.* **1998**, *32*, 1192−1198.
(9) Kiriluk, R. M.; Servos, M. R.; Whittle, D. M.; Cabana, G.; Rasmussen, J. B. *Can. J. Fish. Aquat. Sci.* **1995**, *52*, 2660−2674.
(10) Broman, D.; Näf, C.; Rolff, C.; Zebühr, Y.; Fry, B.; Hobbie, J. *Environ. Toxicol. Chem.* **1992**, *11*, 331−345.
(11) Kidd, K. A.; Schindler, D. W.; Hesslein, R. H.; Muir, D. C. G. *Can. J. Fish. Aquat. Sci.* **1998**, *55*, 869−881.
(12) Whittle, D. M.; Kiriluk, R. M.; Carswell, A. A.; Keir, M. J.; MacEachen, D. C. *Chemosphere* **2000**, *40*, 1221−1226.
(13) Rasmussen, J. B.; Rowan, D. J.; Lean, D. R. S.; Carey, J. H. *Can. J. Fish. Aquat. Sci.* **1990**, *47*, 2030−2038.
(14) Vetter, W.; Luckas, B. *Chemosphere* **2000**, *41*, 499−506.
(15) Wiberg, K.; Letcher, R. J.; Sandau, C. D.; Norstrom, R. J.; Tysklind, M.; Bidleman, T. F. *Environ. Sci. Technol.* **2000**, *34*, 2668−2674.
(16) Fisk, A. T.; Moisey, J.; Hobson, K. A.; Karnovsky, N. J.; Norstrom, R. J. *Environ. Pollut.* **2001**, *113*, 225−238.
(17) Moisey, J.; Fisk, A. T.; Hobson, K. A.; Norstrom, R. J. *Environ. Sci. Technol.* **2001**, *35*, 1920−1927.
(18) Fisk, A. T.; Holst, M.; Hobson, K. A.; Duffe, J.; Moisey, J.; Norstrom, R. J. *Arch. Environ. Contam. Toxicol.* **2002**, *42*, 118−226.
(19) Wong, C. S.; Garrison, A. W.; Foreman, W. T. *Environ. Sci. Technol.* **2001**, *35*, 33−39.
(20) Wong, C. S.; Garrison, A. W.; Smith, P. D.; Foreman, W. T. *Environ. Sci. Technol.* **2001**, *35*, 2448−2454.
(21) Hoekstra, P. F.; Wong, C. S.; O'Hara, T. M.; Solomon, K. R.; Mabury, S. A.; Muir, D. C. G. *Environ. Sci. Technol.* **2002**, *36*, 1419−1425.
(22) Boon, J. P.; Eijgenraam, F.; Everaarts, J. M.; Duinker, J. C. *Mar. Environ. Res.* **1989**, *27*, 159−176.
(23) Stegeman, J. J.; Klopper-Sams, P. J. *Environ. Health Perspect.* **1987**, *71*, 87−95.
(24) Kleinow, K. M.; Melancon, M. J.; Lech, J. J. *Environ. Health Perspect.* **1987**, *71*, 105−119.
(25) Vetter, W.; Smalling, K. L.; Maruya, K. A. *Environ. Sci. Technol.* **2001**, *35*, 4444−4448.
(26) Wong, C. S.; Lau, F.; Clark, M.; Mabury, S. A.; Muir, D. C. G. *Environ. Sci. Technol.* **2002**, *36*, 1257−1262.
(27) Muir, D. C. G. *Toxaphene in the food webs of Lake Superior and nearby lakes*; Final report to U.S. EPA Great Lakes National Program Office: 2001.
(28) National Laboratory for Environmental Testing. *Protocol for the fractionation and analysis of OCs/PCB/toxaphene*; Protocol 97-101; National Laboratory for Environmental Testing, Environment Canada: 1997.
(29) Wong, C. S.; Hoekstra, P. F.; Karlsson, H.; Backus, S. M.; Mabury, S. A.; Muir, D. C. G. *Chemosphere* **2002**, *49*, 1339−1347.
(30) Wong, C. S.; Garrison, A. W. *J. Chromatogr. A* **2000**, *866*, 213−220.
(31) Harner, T.; Wiberg, K.; Norstrom, R. *Environ. Sci. Technol.* **2000**, *34*, 218−220.
(32) Muir, D. C. G.; Wong, C. S.; Whittle, D. M.; Backus, S. M.; Teixeira, C.; DeVault, D. S. Polychlorinated biphenyls in Lake Superior: Aquatic food web dynamics and temporal trends; NWRI unpublished report.
(33) Kannan, N.; Reusch, T. B. H.; Schulz-Bull, D. E.; Petrick, G.; Duinker, J. C. *Environ. Sci. Technol.* **1995**, *29*, 1851−1859.
(34) McFarland, V. A.; Clarke, J. U. *Environ. Health Perspect.* **1989**, *81*, 225−239.
(35) Niimi, A. J. *Sci. Total Environ.* **1996**, *192*, 123−150.
(36) Klump, J. V.; Kaster, J. L.; Sierszen, M. E. *Can. J. Fish. Aquat. Sci.* **1991**, *48*, 284−289.
(37) Landrum, P. F.; Frez, W. A.; Simmons, M. S. *Chemosphere* **1992**, *25*, 397−415.
(38) Landrum, P. F.; Nalepa, T. F. *J. Great Lakes Res.* **1998**, *24*, 889−904.
(39) Campbell, L. M.; Muir, D. C. G.; Whittle, D. M.; Backus, S.; Norstrom, R. J.; Fisk, A. T. *Environ. Sci. Technol.* **2003**, *37*, 1720−1725.
(40) Lyman, W. J.; Reehl, W. F.; Rosenblatt, D. H. *Handbook of Chemical Property Estimation Methods*; American Chemical Society: Washington, DC, 1990.
(41) Fisk, A. T.; Norstrom, R. J.; Cymbalisty, C. D.; Muir, D. C. G. *Environ. Toxicol. Chem.* **1998**, *17*, 951−961.
(42) Letcher, R. J.; Klasson-Wehler, E.; Bergman, A. In *The Handbook of Environmental Chemistry*; Paasivirta, J., Ed.; Springer-Verlag: New York, 2000; pp 317−359.
(43) Grossnickle, N. E. *Hydrobiologia* **1982**, *93*, 101−107.
(44) Lester, D. C.; McIntosh, A. *Environ. Toxicol. Chem.* **1994**, *13*, 1825−1841.
(45) Harkey, G. A.; Lydy, M. J.; Kukkonen, J.; Landrum, P. F. *Environ. Toxicol. Chem.* **1994**, *13*, 1445−1455.
(46) Jeremiason, J. D.; Hornbuckle, K. C.; Eisenreich, S. J. *Environ. Sci. Technol.* **1994**, *28*, 903−914.
(47) Bedard, D. L.; Wagner, R. E.; Brennan, M. J.; Haberl, M. L.; Brown, J. F., Jr. *Appl. Environ. Microbiol.* **1987**, *53*, 1094−1102.
(48) Bedard, D. L.; Quensen, J. F., III In *Microbial Transformation and Degradation of Toxic Organic Contaminants*; Young, L. Y., Cerniglia, C. E., Eds.; Wiley-Liss: New York, 1995; pp 127−216.
(49) Singer, A. C.; Wong, C. S.; Crowley, D. E. *Appl. Environ. Microbiol.* **2002**, *68*, 5756−5759.
(50) Rhee, G.-Y.; Bush, B.; Bethoney, C. M.; DeNucci, A.; Oh, H.-M.; Sokol, R. C. *Environ. Toxicol. Chem.* **1993**, *12*, 1025−1032.
(51) Baker, J. E.; Eisenreich, S. J. *J. Great Lakes Res.* **1989**, *5*, 84−103.
(52) Selgeby, J. H. *J. Great Lakes Res.* **1998**, *14*, 44−51.
(53) Hudson, P. L.; Savino, J. F.; Bronte, C. R. *J. Great Lakes Res.* **1995**, *21*, 445−457.
(54) Bright, D. A.; Grundy, S. L.; Reimer, K. J. *Environ. Sci. Technol.* **1995**, *29*, 2504−2512.
(55) Stapleton, H. M.; Letcher, R. J.; Baker, J. E. *Environ. Sci. Technol.* **2001**, *35*, 4747−4752.
(56) Johnson, T. B.; Mason, D. M.; Bronte, C. R.; Kitchell, J. F. *Trans. Am. Fish. Soc.* **1998**, *127*, 496−506.
(57) Dryer, W. R.; Erkkila, L. F.; Tetzloff, C. L. *Trans. Am. Fish. Soc.* **1965**, *94*, 169−176.
(58) Conner, D. J.; Bronte, C. R.; Selgeby, J. H.; Collins, H. L. *Food of Salmonine Predators in Lake Superior, 1981−87*; Technical Report 59; Great Lakes Fishery Commission: Ann Arbor MI, 1993.
(59) Mason, D. M.; Johnson, T. B.; Kitchell, J. F. *Can. J. Fish. Aquat. Sci.* **1998**, *55*, 1273−1284.
(60) Selgeby, J. H. In *Presented papers from the Council of Lake Committees Plenary Session on Great Lakes predator-prey issues, March 20, 1985*; Eshenroder, R. L., Ed.; Great Lakes Fishery Commission Special Publication 85-3; Great Lakes Fishery Commission: Ann Arbor, MI, 1985; pp 3−12.
(61) Selgeby, J. H.; Bronte, C. R.; Slade, J. W. In *The State of Lake Superior in 1992*; Hansen, M. J., Ed.; Great Lakes Fishery Commission Special Publication 94-1; Great Lakes Fishery Commission: Ann Arbor, MI, 1994; pp 53−62.
(62) Bronte, C. R.; Ebener, M. P.; Schreiner, D. R.; DeVault, D. S.; Petzold, M. M.; Jensen, D. A.; Richards, C.; Lozano, S. J. *Can. J. Fish. Aquat. Sci.* (in press).
(63) Harvey, C. J.; Kitchell, J. F. *Can. J. Fish. Aquat. Sci.* **2000**, *57*, 1395−1403.
(64) Vetter, W.; Schurig, V. *J. Chromatogr. A* **1997**, *774*, 143−175.
(65) Gobas, F. A. P. C.; McCorquodale, J. R.; Haffner, G. D. *Environ. Toxicol. Chem.* **1993**, *12*, 567−576.
(66) Gobas, F. A. P. C.; Wilcockson, J. B.; Russell, R. W.; Haffner, G. D. *Environ. Sci. Technol.* **1999**, *33*, 133−141.
(67) Niimi, A. J.; Oliver, B. G. *Can. J. Fish. Aquat. Sci.* **1983**, *40*, 1388−1394.
(68) Coristine, S.; Haffner, G. D.; Ciborowski, J. J. H.; Lazar, R.; Nanni, M. E.; Metcalfe, C. D. *Environ. Toxicol. Chem.* **1996**, *15*, 1382−1387.
(69) Thomann, R. V. *Environ. Sci. Technol.* **1989**, *23*, 699−707.
(70) Gobas, F. A. P. C. *Ecol. Modell.* **1993**, *69*, 1−17.
(71) Morrison, H. A.; Whittle, D. M.; Metcalfe, C. D.; Niimi, A. J. *Can. J. Fish. Aquat. Sci.* **1999**, *56*, 1389−1400.

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