PCB Congener Profile in the Serum of Humans Consuming Great Lakes Fish

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The State of Michigan has a long history of research into human exposure to environmental contaminants through consumption of recreationally caught fish. A large cohort of Lake Michigan residents who eat fish (fish-eaters) and those who do not eat fish (nonfish-eaters) established in 1980 served as the basis for the congener-specific polychlorinated biphenyl (PCB) exposure evaluation reported here. In this paper we present the serum PCB congener profile for a subset of this cohort who were over 50 years of age. Serum samples were collected in 1993–1995 and were evaluated by a dual column capillary column gas chromatography procedure capable of detecting over 90 PCB congeners. This evaluation demonstrated significant PCB exposure in the fish-eaters (mean serum PCB of 14.26 ppb; *n* = 101). This elevated exposure allowed the establishment of a detailed profile of the PCB congeners found in humans exposed by this route. Twenty-two congeners of varying concentrations were the most prevalent and constituted over 95% of the total PCB present in most subjects. Four congeners, 138/163 (2,2′,3,4,4′,5-PCB/2,3,3′,4′,5,6-PCB), 180 (2,2′,3,4,4′,5,5′-PCB), and 153 (2,2′,4,4′,5,5′-PCB), accounted for 55–64% of the total PCB load. Other congeners, some of toxicologic significance, were also detected by this analytical protocol. Nonfish-eaters had lower total serum PCB levels (mean = 4.56; *n* = 78), but the same general pattern of PCB congeners was present. It was demonstrated that careful selection of a subset of prevalent PCB congeners could provide a cost-effective assessment of exposure without losing critical scientific information. *Key words:* **aging, capillary column gas chromatography, fish-eaters, Great Lakes, Lake Michigan, polychlorinated biphenyl (PCB) congener profile.** *Environ Health Perspect* **108:167–172 (2000). [Online 10 January 2000]**
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Recreational fishing in the Great Lakes became a growth industry after the successful introduction of several desirable salmon species in the late 1960s. The discovery that aquatic chemical pollutants such as DDT and polychlorinated biphenyls (PCBs) biomagnified in these edible species necessitated an assessment of the public health risks from eating sport-caught Great Lakes fish. To evaluate this, the Michigan Department of Public Health established an 11-community cohort study of Lake Michigan recreational fish-eaters in 1979–1982 (*1,2*). This cohort was unique because of its size (572 fish-eaters and 419 controls), age stratification (18–79 years), elevated fish consumption (24–270 lb/year; median 38.5 lb/year), and era of recruitment (10–20 ppm DDT and PCB levels in trout and salmon). Reported serum levels, based on packed column analytical methodology, showed significant PCB exposure in fish-eaters (median 21.4 ppb) as compared to control subjects who ate little or no sport-caught fish (median 6.6 ppb) (*1,2*). This confirmed earlier pilot work which indicated that contaminated fish represented the most significant route of PCB exposure for humans and that age, duration of consumption, and amount consumed correlated with exposure (*3*). Retesting of a portion of this cohort in 1989 showed no significant decline in average serum PCB levels (*4*). In contrast, fish-eaters recruited more recently have been found to have serum PCB levels far below those in the Michigan cohort (*5–7*). Although the Michigan cohort does appear to be more highly exposed than other more recently recruited cohorts, it is difficult to make direct comparisons between older studies in which PCBs were determined via packed column chromatography and more recent studies in which PCBs were quantitated via capillary gas chromatography.

The size, historical database, and high degree of exposure of the Michigan fish-eater cohort provide an excellent opportunity to evaluate a PCB-exposed population as it ages. This includes the dynamics of serum PCB concentrations over time, as well as detailed identification of individual PCB congeners—especially those typically present at low levels. In this paper we present the PCB congener profile for a subset of the Michigan fish-eater cohort who were over age 50 at the time of reassessment in 1993–1995. A future report will focus on changes in total serum PCB concentrations over time and relate those changes to changes in fish contamination levels and fish consumption practices.

Methods

Sample. The original Michigan fish-eater cohort was recruited in 1979–1982 by field visits to sites of fishing activity (e.g., docks, marinas, bait shops) in 11 Lake Michigan shoreline communities. Both direct contact and referrals were used in the field to identify candidates for the study. An initial screening interview was used to select fish-eaters who ate 26 or more pounds of sport-caught fish annually. A total of 572 individuals met this criterion and was enrolled in the study as fish-eaters. A relatively unexposed comparison group was identified by random digit dialing from a pool of 1,000 households in the same communities. A telephone-screening interview was used to select subjects who ate 0–6 lb sport-caught fish annually. A total of 419 individuals who were age- and region-matched to the fish-eaters were enrolled in the study (*1*).

In this paper we evaluate a subsample of the cohort who agreed to participate in a neuropsychologic evaluation of subjects ≥ 50 years of age, which was initiated in 1992. A pool of 549 age-eligible subjects was divided into 12 cells representing age (50–59 years, 60–69 years , and ≥ 70 years), sex, and fish consumption. Subjects were then randomly selected and recruited from each cell, with a participation goal of 13–17 from each cell. A total of 188 eligible subjects agreed to participate. Informed consent was obtained from all subjects prior to participation. Blood serum analyses were performed on 179 of the 188 participants: 101 fish-eaters and 78 controls. Details of the recruitment and characterization of this cohort subsample have been described elsewhere (*8*).

Blood collection. Blood for analytical laboratory analysis was collected at the subjects'

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homes by a public health department phlebotomist via antecubical venapuncture. Two 15-mL vacutainer tubes of blood were collected and centrifuged within 4 hr. Serum was transferred into hexane-rinsed glass specimen vials, labeled with the participant identification number, and frozen at -20°C until analysis. Duplicate samples were drawn from 10% of the subjects for laboratory quality assurance procedures. The blood samples were batched for analysis with randomized insertion of duplicates and bench controls. Samples were identified only by laboratory number to blind the laboratory personnel to the fish-eating status of the subject.

Analytical procedure. Congener-specific quantitation of PCBs was performed by the Health Risk Assessment Laboratory at the Michigan Department of Community Health (Lansing, MI). This laboratory has supported epidemiologic studies for over 25 years and has performed all previous analytical work for the Michigan fish-eater cohort. The laboratory follows both intralaboratory and interlaboratory quality assurance procedures and has participated in analytical procedure development for congener-specific gas chromatographic analysis of PCBs.

Extraction and fractionation. We extracted and fractionated serum specimens and quality control materials by a modification of procedures established by the U.S. Environmental Protection Agency (EPA) (*9*). Frozen serum specimens and quality control materials were removed from the freezer and allowed to warm to room temperature. Methanol (2.0 mL) was added to 4.0 mL serum with gentle mixing. The PCB congeners were extracted into 5.0 mL diethyl ether/hexane (1:1, v/v) with rotary mixing. The extraction step was repeated twice. The resulting diethyl ether/hexane extract (15 mL) was concentrated to a final volume of 1.0 mL under nitrogen at 35°C. The concentrated diethyl ether/hexane extract was then applied to a Florisil column (7 mm i.d.; Floridin Company, Pittsburgh, PA) containing 2.5 g Florisil prerinsed with 10 mL hexane. PCB congeners were eluted from the Florisil column with 20 mL 6% diethyl ether in hexane, concentrated to a final volume of 1.0 mL under nitrogen at 35°C, and applied to a Silica Gel 60 column (9 mm i.d.; Merck and Company, Rahway NY) containing 5.0 g Silica Gel 60 prerinsed with 10 mL hexane. The Silica Gel 60 column was washed with 14 mL hexane. The PCB congeners were eluted from the column with 20 mL hexane followed by 15 mL benzene. The PCB fraction consisted of the 20 mL hexane eluate along with the hexane displaced from the column by benzene (approximately 8 mL). This fraction was concentrated to 0.6 mL under nitrogen at 35°C and then diluted to a final volume of 2.5 mL with isooctane. Two microliters of the resulting solution was analyzed by capillary gas chromatography.

Gas chromatography. We performed capillary gas chromatography of the fraction containing PCB congeners using a Varian 3500 gas chromatograph (Varian Instrument Company, Walnut Creek, CA) equipped with a Varian 8100 autosampler, a Varian 1093/1094 septum-programmable injector with a Y-connector to two high resolution-fused silica capillary columns (column 1, 60 m × 0.25 µm i.d. DB-5, film thickness 0.25 µm; column 2, 60 m × 0.25 i.d. DB-1701, film thickness 0.25 µm; J & W Scientific, Folsom, CA), two nickel-63 electron capture detectors, and a Varian DS-654 data station. The carrier gas was hydrogen (1.8 mL/min) and the make-up gas was nitrogen (20 mL/min). The injector temperature was 275°C. The column temperature ranged from an initial temperature of 110°C to a final temperature of 260°C. The temperature program was as follows: 110°C for 1 min, 110–170°C at 6°C/min, 170–230°C at 1.5°C/min, 230–260°C at 3°C/min, and then held at 260°C for 16 min. The total run time was 77 min. The detector temperature was at 320°C.

Quantitation. Individual PCB congeners were quantified using an Aroclor mixture as a standard according to the method of Mullin and colleagues (*10,11*). The standard mixture was composed of Aroclors 1232, 1248, and 1262 (25:18:18, v/v/v) and PCB congener 169 [International Union of Pure and Applied Chemistry (IUPAC) number; 3,3′,4,4′,5,5′-PCB]. The use of the high-resolution dual capillary column method allows quantification of 83 individual PCB congeners and 7 coeluting congener pairs (Table 1). We report the coeluting pairs as a single value, for a total of 90 values reported. In cases where both columns yielded a PCB value, we used the DB-5 column value in the analysis.

Quality control. In addition to duplicate sample analysis, quality control involved two distinct sets of control materials that were incorporated into each batch of 10 unknowns. The first set was prepared using bovine sera (PCB free) spiked with Aroclor 1260 at levels of 10, 30, and 100 ppb (µg/L) to generate three levels of quality control material. These controls were treated in the same manner as human serum specimens. We used data from these control materials to calculate percent recoveries, which ranged from 86% to 109%, depending on concentration. The second set of external control materials consisted of mixtures of individual PCB congeners in isooctane. We prepared two control mixtures in the laboratory; each consisted of 10 individual PCB congeners at 4 ppb each. In addition, we evaluated a mixture of 20 individual PCB congeners in

Table 1. PCB congeners quantitated by capillary column gas chromatography.

IUPAC no.	DL (ppb)	IUPAC no.	DL (ppb)	IUPAC no.	DL (ppb)
005	0.20	076	0.10	158	0.08
006	0.20	077	0.05	169	0.20
008	1.30	041/071	0.30	**138/163**	**0.50**
015	0.50	066/095	1.00	**171**	**0.02**
007/009	0.30	082	0.05	**172**	**0.15**
016	0.30	084	0.10	174	0.09
017	0.70	085	0.05	175	0.20
018	1.20	087	0.25	176	0.20
022	0.50	091	0.20	**177**	**0.10**
025	0.20	092	0.80	178	0.85
026	0.30	097	0.10	179	0.12
028	0.70	**099**	**0.25**	**180**	**0.20**
031	0.80	100	0.10	**182**	**0.16**
032	0.70	101	0.85	**183**	**0.05**
033	0.60	**105**	**0.30**	185	0.20
037/042	0.30	110	0.60	**187**	**0.03**
040	0.10	**118**	**0.60**	**193**	**0.10**
044	0.60	123	0.05	**170/190**	**0.05**
045	0.10	128	0.10	**194**	**0.06**
046	0.40	132	0.15	**195**	**0.03**
047	0.10	135	0.20	198	0.10
048	0.30	136	0.20	200	0.08
049	0.20	137	0.05	201	0.20
052	0.40	141	0.07	**199**	**0.14**
056	0.20	144	0.08	202	0.07
060	0.50	**146**	**0.12**	205	0.15
063	0.10	149	0.30	**196/203**	**0.12**
064	0.20	151	0.10	**206**	**0.03**
070	0.30	**153**	**0.30**	207	0.06
074	**0.20**	157	0.20	**208**	**0.02**

DL, detection limits. The 22 congener peaks are indicated in bold.

isooctane (2 ppb), which we obtained from the EPA. The congener mixtures were used to assess analytical performance (accuracy and precision) of the gas chromatographic analysis. Relative standard deviations were between 11.9 and 24.7%, depending on concentration. The laboratory also participated in an interlaboratory proficiency testing program under the Great Lakes Human Health Effects Research Program administered by the Agency for Toxic Substances and Disease Registry (Atlanta, GA).

Data analysis. We compared the PCB congener profiles in the 101 fish-eaters and 78 nonfish-eaters by focusing on a subset of 22 peaks representing 25 congeners (shown in bold in Table 1) previously shown to account for > 90% of the total PCBs in the serum of Lake Michigan fish-eaters. For each individual subject, we determined the proportion of the total serum PCB concentration accounted for by each individual peak and the total proportion accounted for by the sum of the 22 peaks. Arithmetic mean proportions for the fish-eaters and nonfish-eaters were then determined. Nondetectable values were set to zero for these analyses. We compare estimates of total PCBs based on these 22 congener peaks to estimates based on previously reported protocols reporting fewer congener peaks (*12*).

Results

This now-aging subset of the original Michigan Great Lakes fish-eater cohort continues to have elevated PCB levels in comparison to nonfish-eaters (Table 2). The 14.26 ppb overall average serum PCB for fish-eaters, 13–15 years after original enrollment, verifies that this cohort was unique in terms of the extent of exposure to contaminants found in fish. The higher exposure of the Michigan fish-eater cohort allows a more thorough assessment of the presence of PCB congeners than would be possible with enrollees in more recent studies who have significantly less PCB exposure (*5–7,13*). Combined with an analytical technique capable of reporting 90 elution peak values (Table 1), this data set allows construction of the PCB congener profile for Lake Michigan fish-eaters (Table 2, Figure 1). As shown in Table 3, the total serum PCB level for the 22 most prevalent congener peaks is only slightly lower than that for all 90 congener peaks.

The 22 congener peaks identified as prevalent in Lake Michigan fish-eaters accounted for, on average, 99% of the total PCB (range 86–100%). The 22 congeners accounted for < 90% of the total PCB in only one individual with detectable PCB levels. Five other individuals, two fish-eaters and three controls, had no detectable PCBs in their serum.

The proportion each congener contributed to the total PCBs in fish-eaters and nonfish-eaters is shown in Table 2 and illustrated in Figure 1. Three peaks representing four PCB congeners, PCBs 138/163 (2,2′,3,4,4′,5-PCB/2,3,3′,4′,5,6-PCB), 180 (2,2′,3,4,4′,5,5′-PCB), and 153 (2,2′,4,4′,5,5′-PCB), accounted for, on average, 55% of the total PCBs among fish-eaters and 64% of the total PCBs among nonfish-eaters. All of these are hexa- and heptachlorinated, ortho-substituted PCBs. Although the fish-eaters had significantly higher serum PCB concentrations, the same general pattern of congeners was observed in both fish-eaters and nonfish-eaters from Michigan.

Discussion

Congener profile. The Michigan Department of Public Health laboratory (Lansing, MI) uses a dual capillary column gas chromatographic technique capable of quantitating 83 single congeners and 7 coeluting congener pairs. In 1993, an analysis was made to determine which of the 90 congener peaks were consistently detectable in fish-eaters from the Michigan Great Lakes fish-eater study. Samples from 38 participants with total serum PCB levels > 20 ppb (as determined by Association of Official Analytical Chemists packed column analysis) were evaluated. This cutoff was selected because we believed that the maximum number of PCB congeners present would be detected, whereas at lower total serum PCB levels some peaks would be below detection levels and therefore lost to analysis. The analysis of samples from these higher exposure fish-eaters revealed that 22 peaks representing 25 congeners (those shown in Table 2 and Figure 1) consistently accounted for > 90% of the total PCB present in serum. Accordingly, these 22 congener peaks provide the basis for the comparative analyses in this paper.

Table 2. Mean concentration, proportion of total, and percent of participants with detectable levels for 22 PCB congener peaks in fish-eaters and nonfish-eaters (controls).

PCB congener[a]	Chlorine substitution pattern	Maximum concentration (ppb)		Mean concentration (ppb)		Proportion of total		Percent of participants with detectable levels	
		Fish-eater	Control	Fish-eater	Control	Fish-eater	Control	Fish-eater	Control
138/163	2,2′,3,4,4′,5/2,3,3′,4′,5,6	14.70	6.19	3.13	1.12	0.25	0.25	97.03	85.90
153	2,2′,4,4′,5,5′	10.48	4.37	2.13	0.75	0.15	0.16	92.08	83.33
180	2,2′,3,4,4′,5,5′	10.35	4.24	2.00	0.79	0.15	0.23	96.04	96.15
170/190	2,2′,3,3′,4,4′,5/2,3,3′4,4′,5,6	3.88	1.61	0.73	0.28	0.06	0.07	98.02	92.31
199	2,2′,3,3′,4,5,5′,6	3.68	1.67	0.70	0.27	0.05	0.06	92.08	73.08
74	2,4,4′,5	2.82	1.04	0.64	0.22	0.05	0.04	84.16	57.69
118	2,3,4,4′,5	5.96	2.51	0.83	0.16	0.04	0.02	55.45	15.39
146	2,2′,3,4,5,5′	3.29	0.91	0.62	0.13	0.04	0.02	89.11	44.87
187	2,2′,3,4,5,5′,6	2.78	1.11	0.58	0.19	0.04	0.05	98.02	92.31
99	2,2′,4,4′,5	4.00	1.22	0.54	0.16	0.04	0.03	59.41	30.77
196/203	2,2′,3,3′,4,4′,5,6/ 2,2′,3,4,4′,5,5′,6	1.94	1.25	0.42	0.13	0.03	0.02	77.23	42.31
194	2,2′,3,3′,4,4′,5,5′	1.60	0.77	0.33	0.14	0.03	0.04	94.06	83.33
105	2,3,3′,4,4′	2.20	0.78	0.26	0.02	0.01	<0.01	36.63	3.85
183	2,2′,3,4,4′,5,6′	1.70	0.60	0.26	0.05	0.01	0.01	73.27	34.62
172	2,2′,3,3′,4,5,5′	1.46	0.53	0.22	0.03	0.01	<0.01	53.47	8.97
177	2,2′,3,3′,4,5,6	1.35	0.65	0.21	0.03	0.01	<0.01	54.46	12.82
193	2,3,3′,4,5,5′,6	0.89	0.43	0.13	0.02	0.01	<0.01	42.57	8.97
195	2,2′,3,3′,4,4′,5,6	0.35	0.29	0.08	0.03	0.01	<0.01	82.18	52.56
171	2,2′,3,3′,4,4′,6	0.31	0.22	0.06	0.01	<0.01	<0.01	66.34	26.92
182	2,2′,3,4,4′,5,6	0.78	0.00	0.05	0.00	<0.01	<0.01	8.91	0.00
206	2,2′,3,3′,4,4′,5,5′,6	0.14	0.08	0.01	0.01	<0.01	0.01	18.81	17.95
208	2,2′,3,3′,4,4′,5,5′,6,6′	0.13	0.10	0.02	0.01	<0.01	<0.01	37.62	20.51
All				13.96	4.55				

[a]IUPAC numbers.

As discussed above, four congeners (PCBs 138/163, 180, and 153) accounted for 55–64% of the total PCBs in both fish-eaters and nonfish-eaters, and the 22 previously identified peaks representing 25 PCB congeners accounted for > 99% of the total PCBs in both groups. In most analytical procedures, congeners 138 and 163 coelute. However, in a few studies the two congeners have been resolved and PCB 138 has been found to predominate, with PCB 163 accounting for only 10–24% of the peak (*14*). Congeners 138, 180, and 153 are the most consistently detected and quantitatively dominant congeners in human tissue worldwide, and frequently account for 40–60% of the total PCBs (*15*). These congeners have been reported as major contributers to total PCBs in maternal serum from New York (*16*) and in pooled serum samples from children born to women who regularly ate Lake Michigan fish (*17*). Other congeners that frequently make large contributions to the total PCBs include PCBs 28 (2,4,4′-PCB), 118 (2,3′,4,4′,5-PCB), and 170 (2,2′,3,3′,4,4′,5-PCB) (*15*). PCBs 118 and 170/190 (2,3,3′,4,4′,5,6-PCB) each accounted for 5–6% of the total PCBs in our sample. PCB 28 does not appear to be a major contributor to the total PCBs in our sample. However, this could be because of the relatively high detection limit for that congener (0.70 ppb). A few other congeners with relatively high detection limits (e.g., IUPAC numbers 8, 18, 31, 32, 92, 66/95, 101, and 178) could also be underrepresented in our sample.

An evaluation of reported human serum PCB levels from various fish-eater studies (*5–7,12,13*) shows a trend of PCB exposure well below that reported in the Michigan cohort (*1,4,8*). The reason for this could be 2-fold: *a*) the original Michigan study enrolled high consumers of more highly contaminated Great Lakes fish, and *b*) the laboratory method more thoroughly evaluated PCB exposure. To investigate the significance of the latter, we evaluated the average blood serum data of the 101 fish-eaters and 78 comparison participants in this study by three quantitation protocols—the 90 congener peaks routinely reported by our laboratory, the 22 congener peaks identified above as the primary peaks in Lake Michigan fish-eaters, and the 13 congeners used by Sonzogni et al. (*12*) in an earlier study of Lake Michigan fish-eaters. As shown in Table 3, there is little difference in total PCB between the first two protocols, but there is a significant reduction of 36.4% with the third, which quantitates on the basis of only 13 congeners. This analysis confirms the laboratory's preliminary evaluation that little accuracy of estimation was lost when the 25 most prevalent congeners found in highly exposed fish-eaters were used for the quantitation protocol. It also demonstrates that quantitation protocols using fewer congeners run the risk of underreporting total PCB exposure.

Selection of a PCB quantitation protocol, if used consistently, does not affect within-study validity. However, it could result in underestimation of total PCB burdens, and it does become an important factor when data from various studies or regions of the Great Lakes are compared for exposure analysis or risk assessment. The differences in quantitation protocols demonstrated here underscore the wisdom and importance of establishing an interlaboratory quality assurance scheme for research programs involving multiple institutions and a regional scope.

The selection of PCB congeners for analysis represents a critical decision in study design. For screening surveys, reduced analytical costs may be a persuasive reason for quantitating fewer congeners, but then the selection criteria for inclusion of congeners becomes very critical (*13*). Inappropriate choices could lead to underestimation of total PCB burdens. For hypothesis testing or interstudy comparisons, a larger number of congeners may be critical. For example, a 13-congener protocol used in the past by Sonzogni et al. (*12*) to evaluate Wisconsin fish-eaters excluded 16 congeners that together accounted for, on average, 4.8 ppb (34%) of the total PCB load in our sample of Lake Michigan fish-eaters. That laboratory has since revised their analytical protocol to include a much more extensive group of congeners. The subset of 25 congeners (22 congener peaks) selected in the Michigan protocol were identified based on prevalence in both fish-eaters and nonfish-eaters. With the exception of two congeners that occurred in < 20% of fish-eaters, all had a prevalence of ≥ 37%. In contrast, the 13 congeners selected in the no longer used Wisconsin protocol included 6 congeners with a prevalence < 1% in our Lake Michigan fish-eaters. Some of the 6 may have been selected based on their toxicologic significance or commercial availability, but they are not persistent in the environment and do not usually contribute significantly to the total PCB burden. These comparisons illustrate the importance of using appropriate selection criteria when only a subset of congeners will be assessed.

Toxicologic significance of exposure. Unfortunately, only a few of the 25 most prevalent congeners identified in this sample of Great Lakes fish-eaters have been evaluated for toxic effects in animal models. These include two mono-*ortho* congeners, PCBs 105 and 118, which are of interest to toxicologists because of their affinity for the aryl hydrocarbon (Ah) receptor, and one di-*ortho* congener, PCB 153. Almost no toxicologic

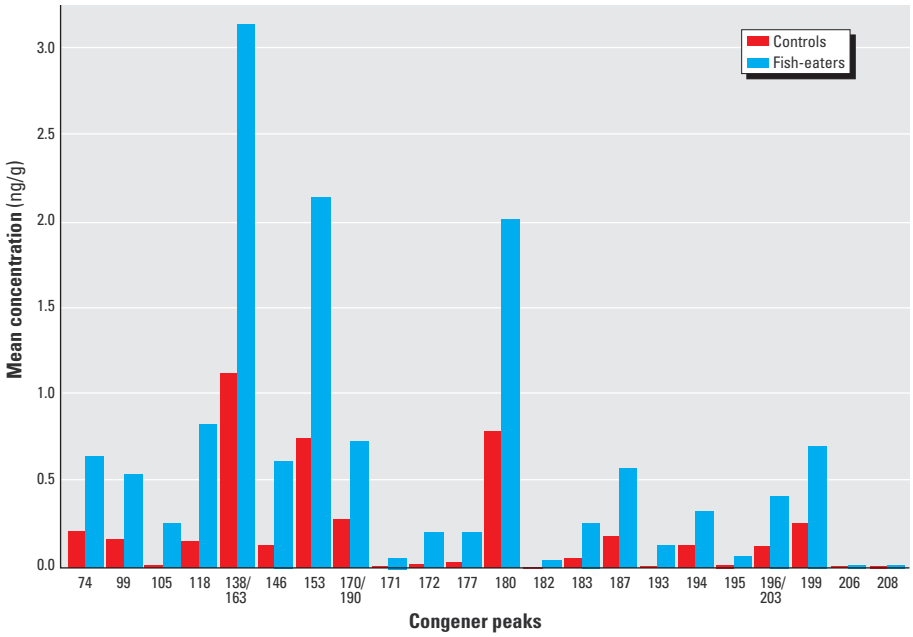

Figure 1. Mean concentrations (ng/g) of 22 PCB congener peaks in fish-eaters and controls. Peaks were quantitated using dual column capillary column gas chromatography. PCB congeners are identified by IUPAC number.

Table 3. Comparison of total PCBs calculated on the basis of all 90 congener peaks, the 22 most prevalent peaks, or 13 peaks used by Sonzogni et al. (*12*).

No. of congeners	Fish-eaters (n = 101)	Nonfish-eaters (n = 78)
90	14.26	4.56
22	13.96	4.55
13	9.42	3.29

data are available for the other 21 congeners. Both PCB 105 and PCB 118 have dramatic effects on thyroid hormones in animal models, producing very marked reductions in circulating thyroxine (T_4) levels (*18,19*). PCB 153 causes more moderate reductions in serum T_4 (*19*). Together, these three congeners account for approximately 23% of the total PCBs in Lake Michigan fish-eaters. This suggests that thyroid hormone levels are a relevant health outcome to assess in this population. A number of PCB congeners have also been evaluated for estrogenic activity, but only 2 of the 25 most prevalent congeners in the fish-eaters, PCBs 99 and 153, have been evaluated (*20,21*). Both were weakly estrogenic in the rat uterotrophic assay.

PCBs 118 and 153 have also been evaluated for effects on behavioral functioning in animal models (*22,23*). Gestational and lactational exposure to either congener was associated with later deficits in spatial learning and memory in female offspring (*22*). Male offspring did not show a similar deficit. PCB 153 also caused an increase in locomotor activity of both male and female offspring, whereas PCB 118 did not alter activity levels in either sex (*23*).

A broader range of PCB congeners have been evaluated using *in vitro* assays designed to assess neurotoxicity. Of particular interest is a recent study that evaluated the interaction of various highly chlorinated, *ortho*-substituted PCB congeners with ryanodine sensitive calcium channels (*24*). Previously, certain *ortho*-substituted PCB congeners were shown to interact with ryanodine sensitive calcium release channels in microsomal membranes of neurons and to alter cellular calcium signaling (*25*). These molecular events could be the underlying mechanism for the neurobehavioral effects that have been reported following PCB exposure. The recent study examined the structure–activity relationship among 20 environmentally relevant nonplanar, *ortho*-substituted PCBs towards two different isoforms of the ryanodine channel—one found mainly in skeletal muscle (RyR1) and one found mainly in heart and brain (RyR2). The brain isoform was found to be 2- to 5-fold more sensitive to several of the primary congeners detected in Lake Michigan fish-eaters including PCBs 138 and 170, both of which are detected in > 97% of the fish-eaters. Both of these congeners had median effective concentrations (EC_{50} values) in the nanomolar range (*24*). The neurotoxicity of these congeners in animal models has yet to be investigated.

Most laboratory animal studies have used PCB doses in the milligram per kilogram range, whereas human exposures are usually at least an order of magnitude lower. Furthermore, humans are exposed to complex environmental mixtures of PCB congeners, whereas animal studies have focused on individual PCB congeners. From a risk assessment perspective, the most useful approach to PCB toxicity testing is to assemble a test mixture of the most prevalent congeners found in human tissues. The detailed congener-specific PCB analysis reported here provides information that could be used to assemble such a test mixture.

Summary and Conclusions

In this study, we used an established cohort of persons with robust exposure to contaminants in recreationally caught Great Lakes fish to show that significant quantities of serum PCBs are still present 15 years after enrollment. The current levels of PCBs in this group are far above those found in enrollees of more recent fish-eater studies. The higher exposure of this cohort provided the basis for an accurate analysis of the profile of individual PCB congeners to which fish-eaters are exposed. Although 90 congener peaks were quantitated, the analysis revealed that 22 peaks representing 25 congeners comprised 99% of the total PCBs present in both fish-eaters and nonfish-eaters. Fish-eaters had significantly higher total serum PCB concentrations, but the same general pattern of PCB congeners was present in both groups. Thus, it was demonstrated that careful selection of a subset of prevalent PCB congeners could provide a cost-effective assessment of exposure without losing critical scientific information.

An examination of the prevalent PCB congeners found in fish-eaters revealed that few have been evaluated for toxic effects in animals. Of those that have, several which make significant contributions to the total serum PCBs are potentially toxic by either *in vivo* or *in vitro* testing. Toxicities include alterations in thyroid hormones, weak estrogenic effects, behavioral effects, and neurotoxicologic effects.

In conclusion, identification of the PCB congener profile in fish-eaters and nonfish-eaters reveals the presence of several congeners that have the potential to affect biologic or health outcomes, indicates some health outcomes that should be evaluated in exposed populations, and highlights the need for toxicologic evaluation of additional PCB congeners in animal models. Whether past or present PCB exposure levels are sufficient to elicit detectable health effects in adult humans remains to be seen. We are currently in the process of evaluating neuropsychologic function and thyroid function in the Lake Michigan fish-eaters for which we have PCB congener profiles. PCB exposure from contaminated fish does not appear to be associated with any reductions in fine motor function (*26*). The data from additional neuropsychologic and hormonal measurements are still under analysis and will be the subject of a latter report.

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