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Hydroxylated PCB metabolites (OH-PCBs) in archived serum from 1950–60's California mothers: A pilot study

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Abstract

We are studying participants selected from the Child Health and Development Studies (CHDS), a longitudinal birth cohort of over 20,000 California pregnancies between 1959 and 1967, for associations between maternal body burden of organochlorine contaminants and thyroid function. We designed a pilot study using 30 samples selected among samples with high and low PCB concentrations to evaluate the feasibility of measuring OH-PCBs in the larger study population. GC-ECD and GC-NCI/MS were used to determine PCBs and OH-PCBs as methyl derivatives, respectively. Maternal serum levels of Σ_{11} PCBs and Σ_8 OH-PCB metabolites varied from 0.74 to 7.99 ng/mL wet wt. with a median of 3.05 ng/mL, and from 0.12 to 0.98 ng/mL wet wt. with a median of 0.39 ng/mL, respectively. Average concentrations of Σ_8 OH-PCB metabolites in the high PCB group were significantly higher than those in the low PCB group ($p < 0.05$). The levels of OH-PCB metabolites were dependent on PCB levels ($r = 0.58$, $p < 0.05$) but approximately an order of magnitude lower ($p < 0.05$). The average ratio of Σ_8 OH-PCBs to Σ_{11} PCBs was 0.14 ± 0.08 . The primary metabolite was 4-OH-CB187 followed by 4-OH-CB107. Both of these metabolites interfere with the thyroid system in *in vitro*, animal, and human studies. OH-PCBs were detectable in all archived sera analyzed, supporting the feasibility to measure OH-PCB metabolites in the entire cohort.

1. Introduction

Since the 1970s–1980s worldwide ban on the production and use of PCBs (polychlorinated biphenyls), concentrations in biota, in general, gradually decreased (Bignert et al., 1998; Norén and Meironyté, 2000). This decrease, however, leveled off in human populations over the last decade (Weihe et al., 2003). Mainly due to their pronounced lipophilicity, and to continued exposure via dietary intake and ambient air inhalation, PCBs persist in the environment and continue to threaten human health, particularly child development. Toxicological associations with endocrine disruption, immune dysfunction, and neurodevelopmental deficits are well documented in the literature. PCBs have also been suggested to interfere with the reproductive system (Richthoff et al., 2003; Rignell-Hydbom et al., 2004), and the auditory system in rats (Kenet et al., 2007) and in children (Longnecker et al., 2004) and thus may be associated with

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developmental disorders. PCBs may also be associated with the occurrence of diabetes (Longnecker et al., 2001; Codru et al., 2007).

Recent evidence suggests that some of the hydroxylated metabolites of PCBs (OH-PCBs) may be more toxic than their parent compounds. These metabolites are formed *in vivo* by cytochrome P450 enzymes which oxidize PCBs to more water soluble forms (e.g., OH-PCBs) via a 1,2 shift (NIH shift) or direct oxygen insertion (Letcher et al., 2000). However, not all OH-PCBs are excreted; several circulate in blood (Park et al., 2007) and may exert toxicological effects, particularly on the thyroid system. OH-PCBs compete with and replace thyroid hormone (thyroxine, T4) on one of its transport proteins (transthyretin, TTR), a possible indication of endocrine disruption associated with hypothyroidism (Lans et al., 1993; Meerts et al., 2002). Neonatal hypothyroidism, if not corrected with replacement hormone, results in cognitive neurodevelopmental deficits (Builee and Hatherill, 2004). Strikingly, one study found that the concentrations of OH-PCBs in cord blood were similar to those of PCBs, indicating that similar levels of OH-PCBs and of their parent PCBs were transferred to the fetus (Park et al., 2008). Methylsulfone PCBs, other PCB metabolites, were also detected in the cord blood, but only in trace amounts (Linderholm et al., 2007a), suggesting that the placenta preferentially transports the OH-PCB metabolites.

Access to archived serum samples collected between 1959 and 1967 from pregnant women participating in the Child Health and Development Studies (CHDS) provided us an opportunity to investigate levels of OH-PCB metabolites. The time period reflected in these samples is prior to the ban on PCBs and, therefore, PCB levels are expected to be higher than they are now. Limited information on OH-PCB metabolites is available from other sources. Thus the primary purpose of this study was to assess the feasibility of measuring specific OH-PCB metabolites in the limited volume (1 mL) of archived sera to determine if the levels measured would likely be toxicologically significant, prior to investigating the relationship between OH-PCBs and the parent PCBs in the entire cohort. Our ultimate goal for the larger study population is to explore associations between OH-PCBs, their parent compounds, and health outcomes over the life course of the mothers and their offspring.

2. Materials and Methods

2.1. Study Population

The CHDS, a longitudinal birth cohort of over 20,000 births among members of the Northern California Kaiser Foundation Health Plan, enrolled pregnant women between 1959 and 1967. These women were followed throughout their pregnancies and serial serum samples were obtained in each trimester and postpartum. Following routine blood tests, these samples were appropriately frozen at -20°C , stored and archived. A subset of offspring was followed at ages 5, 9–11 and 15–17. All subjects gave oral informed consent consistent with practices in the early 1960's. This study was approved by the Institutional Review Board of Columbia University..

The study base for the present analyses is a subset of 600 offspring followed to adolescence (age 15–17), and randomly selected for an ongoing study of prenatal organochlorines, thyroid function and development. Frozen archived maternal serum samples were obtained from either the third trimester or postpartum as Longnecker et al. have demonstrated correspondence of these compounds during pregnancy (Longnecker et al., 1999). These samples were stored at the National Cancer Institute (Fredrick, MD). All maternal samples were analyzed for PCBs and organochlorine pesticides. For this pilot study, we selected 15 of the samples with the highest PCB concentrations and 15 of the lowest based on the distribution of total PCB concentration in the first 100 samples to be analyzed.

2.2. Standards and Chemicals

All PCB standards (PCB-66, 74, 99, 118, 138, 153, 170, 180, 187, 194, and 203) were purchased from AccuStandard, Inc (New Haven, CT, USA). The following hydroxylated PCBs (OH-PCBs) were purchased from Wellington Laboratory (TerraChem Inc., Shawnee Mission, KS, USA) and used as authentic reference standards for the identification and quantification of the analytes: 2,3,3',4',5-pentachlorobiphenyl-4-ol (4-OH-CB107), 2,2',3,4',5,5'-hexachlorobiphenyl-4-ol (4-OH-CB146), 2,2',3',4,4',5-hexachlorobiphenyl-3-ol (3'-OH-CB138), 2,2',3,3',4',5-hexachlorobiphenyl-4-ol (4'-OH-CB130), 2,2',3,4',5,5',6-heptachlorobiphenyl-4-ol (4-OH-CB187), 2,2',3',4,4',5,5'-heptachlorobiphenyl-3-ol (3'-OH-CB180), and 2,2',3,3',4',5,5'-heptachlorobiphenyl-4-ol (4'-OH-CB172). 2,2',4,4',5,5'-hexachlorobiphenyl-3-ol (3-OH-CB153) was provided by Professor Åke Bergman (Stockholm University, Sweden). The numbering of PCBs and OH-PCBs is based on that specified by Ballschmitter and Zell (1980) and Letcher et al (2000), respectively. We used PCB-14, 65 and 166 as the surrogate internal standards for PCB analysis (1.0 ng) and 4'-OH-CB159 for OH-PCB measurement (1.0 ng). We used PCB-30, 204 and 209 (1.5 ng) as injection standards for PCB analyses and only PCB-209 (2.0 ng) for OH-PCB analyses. Diazomethane was synthesized in hexane by using N-nitroso-N-methylurea (Sigma-Aldrich, USA) as described elsewhere (Sandau, 2000). Other chemicals and solvents used for the analysis include dichloromethane and hexane (trace analysis, Burdick and Jackson), methanol, methyl-tert butyl ether, and water (HPLC grade, Fisher Sci., USA), 2-propanol (99.9%, pesticide grade, Fisher Sci., USA), hydrochloric acid, sulfuric acid (98%), potassium hydroxide, potassium chloride, sodium hydroxide, and ethyl alcohol (94–96%, 200 proof) (Fisher Sci., USA), silica (200–400 mesh) (Sigma-Aldrich, USA).

2.3. Analysis and Clean Up

We analyzed maternal sera specimens for 11 PCB congeners and 8 OH-PCB metabolites (as described above) in the ultra clean laboratory of the Department of Toxic Substances Control, Berkeley, CA. PCB and OH-PCB congeners of interest were the most dominant ones found in Californian maternal sera from our previous studies (Rogers et al., 2004 for PCBs and unpublished screening data for OH-PCBs). The blood extraction method and analysis was adopted as described elsewhere (Park et al., 2007). In summary, we separated the PCBs and OH-PCBs from maternal serum by using MTBE:hexane (1:1,v/v), denaturation (6M HCl and 2-propanol), KCl (1%) wash, and KOH (0.5M) phase separation. After acidification and derivatization, the extracts in the phenolic fractions (i.e., containing OH-PCBs) were cleaned up by using concentrated H₂SO₄ (98%) and then a Pasteur pipette column packed with acidic silica gel (1:2, w/w) and activated silica gel. OH-PCB metabolites were determined as methyl derivatives (MeO-PCBs) by using a Varian 1200 gas chromatograph/mass spectrometer (Varian Inc., Walnut Creek, CA). The Mass Spectrometer (MS) was operated in negative chemical ionization (NCI) mode with electron energy of 70 eV. The gas chromatograph (GC) was equipped with a DB-5MS capillary column (30 m × 0.25 mm i.d., 0.25 µm film thickness, J&W Scientific, USA). Helium and methane were used as the carrier and the reagent gases, respectively. Injection (2 µL) was made in split/splitless mode with an injector temperature of 250 °C. The initial GC temperature was set to 80 °C and held for 2 min followed by a 50 °C/min increase to 200 °C, 1 °C/min to 230 °C, and 30 °C/min to 300 °C and held for 4 min. Post run was set to 320 °C for 1 min. The temperatures for both ion source and quadrupole were set to 150 °C. The GC temperature program completely resolved the possible co-elutions (e.g., 3'-OH-CB153/4-OH-CB146, 3'-OH-CB138/4'-OH-CB130, and 3'-OH-CB180/4'-OH-CB172). The most intense ions were monitored; [(M+2-CH₃)⁻] for 4-OH-CB187 and 4'-OH-CB172, and [(M-HCl)⁻] or [(M+2-HCl)⁻] for the rest of the congeners.

Sample clean up and instrumentation for PCB analysis have been described elsewhere (Rogers et al., 2004). In summary, after the extracts in neutral fractions were cleaned up by using

deactivated Florisil column chromatography, they were analyzed for 11 PCB congeners on a Varian 3800 GC-ECD (Varian Inc., Walnut Creek, CA) equipped with RTX-5MS capillary column (60m × 0.25 mm i.d., 0.25 µm thickness, Restek, Bellefonte, PA) and DB-XLB capillary column (60 m × 0.25 mm i.d., 0.25 µm thickness, JandW Scientific, Folsom, CA). Injection (2 µL) was made in split/splitless mode with an injector temperature of 280 °C. The initial GC temperature was set to 80 °C and held for 1.6 min followed by a 15 °C/min increase to 135 °C, 1 °C/min to 261 °C, 3 °C/min to 295 °C and 1 °C/min to 300 °C held for 3.5 min. Post run was set to 320 °C for 1 min.

2.4. Quantification and QA/QC

All glassware were washed, rinsed with acetone and hexane, and baked at 500 °C overnight. Each batch for the analysis of PCB and OH-PCB metabolites was comprised of one reagent blank (1% potassium chloride solution), one laboratory control sample (pooled serum), one Standard Reference Material (SRM1589a, National Institute of Standards and Technology, Gaithersburg, MD), and ten sera specimens. PCB-66, 118, 138, and 187 were quantitated on the RTX-5MS column while PCB-74, 99, 153, 170, 180, 194, and 203 were quantitated on the DB-XLB column by using Empower (Waters, Milford, MA) software application. External calibration standards for the OH-PCB quantification ranged from 0.1 to 48 pg/WL. For the OH-PCB quantification, we derivatized the external calibration standards simultaneously with the serum sample extracts for accurate quantification. Human serum (SRM 1589a) was used as a standard reference material for PCB analysis. Pooled serum samples were used as in-house control samples for both PCB and OH-PCB analyses as described elsewhere (Rogers et al., 2004; Park et al., 2007). Precision and accuracy of both PCBs and OH-PCBs from surrogate spikes, reference material, and control samples were within reasonable analytical error ranges (±25%), with the exception of six samples showing OH-PCB surrogate recoveries falling between 40–60%, possibly due to incomplete derivatization. However, those data were not excluded from this discussion since their contribution to the data summary and comparison did not alter statistically the overall conclusions.

Any values lower than the LOQ (~0.01 ng/mL wet wt.) were replaced by LOQ/2 for summary statistics. The concentrations of OH-PCBs are expressed as ng/mL since they are preferentially bound to blood protein rather than lipid (Table 2). Thus PCBs were also reported on a wet weight basis for ratio comparison.

2.5. Statistical analysis

The PCB and OH-PCB data were skewed (Shapiro-Wilk normality test, $p < 0.05$). We conducted non-parametric tests (e.g., Spearman correlation and Wilcoxon rank sum) for PCB and OH-PCB data to assess their relationships, and any differences between high and low PCB groups. All statistical tests were conducted by using SAS/STAT Software (Version 9).

3. Results and Discussion

3.1. Sociodemographic characteristics

Sociodemographic characteristics of the 30 pregnant women are shown in Table 1. Overall, approximately half the sample was white, but proportionally more non-whites (67%) were in the high PCB group than white (33%). Most women were high school graduates and approximately half worked outside the home. Most women had previous pregnancies. Forty percent of women were smoking during their pregnancy. Over one third of women resided on a farm before age 15, and proportionally more of these were in the high PCB group (47%).

3.2. PCBs and OH-PCBs: levels, distribution, and relationships

While measuring PCBs in the archived maternal serum samples (N=510) for the original CHDS study, we designed this pilot study to explore the feasibility of measuring OH-PCBs. We did this by selecting 30 samples with high and low PCBs and analyze them for OH-PCBs. The mean and median PCB levels of these subsamples (N=30) were similar to the respective mean and median of the entire cohort (N=510) and, in also similar to those reported in previously studied subsets of the same population (James et al., 2002), albeit our range was narrower.

We selected and analyzed 11 major PCB congeners as target analytes based on the information published from those previous studies. Of the many OH-PCB congeners identified in human blood (Hovander et al., 2002), we decided to monitor eight, covering penta, hexa, and hepta-OH-PCBs; 4-OH-CB107, 3-OH-CB153, 4-OH-CB146, 3'-OH-CB138, 4'-OH-CB130, 4-OH-CB187, 3'-OH-CB180 and 4'-OH-CB172 that were reported to be detected predominantly from human blood (Sandau et al., 2000; Fängström et al., 2002; Park et al 2007; 2008).

We were able to measure OH-PCBs in small volumes (1 mL) of stored sera. 4-OH-CB107, and 4-OH-CB187 were measured significantly above the method detection limit from almost all samples (>98%), followed by 4-OH-CB146 (>77%). 3'-OH-CB138 and 3-OH-CB153 were measured in about half of samples (40% and 50%, respectively). 4'-OH-CB172 were only detected in 20% of the samples while 4'-OH-CB130, 3'-OH-CB180 were rarely detected. Therefore, some of the summations of OH-PCB concentrations shown in Table 2 may have been biased by using LOQ/2.

Maternal serum levels of Σ_{11} PCBs and Σ_8 OH-PCB metabolites varied from 0.74 to 7.99 ng/mL wet wt. with a median of 3.05 ng/mL, and from 0.12 to 0.98 ng/mL wet wt. with a median of 0.39 ng/mL, respectively (Table 2). Average concentrations of Σ_8 OH-PCB metabolites in the high PCB group were significantly higher than those in the low PCB group ($p < 0.05$). With the exception of 4-OH-CB107 ($p > 0.05$), the levels of measurable OH-PCB congeners including 3-OH-CB153, 4-OH-CB146, 3'-OH-CB138, 4-OH-CB187, and 4'-OH-CB172 showed statistically significant difference ($p < 0.05$) between low and high PCB groups. Maternal serum OH-PCBs have been reported in only a few studies from Sweden (Gruenewald et al., 2003), The Netherlands (Soechitram et al., 2004), Faroe Islands (Fängström et al., 2002; Fängström et al., 2005), Slovakia (Park et al., 2007), and Japan (Enomoto et al., 2007) (Table 3). The median of the sum of the six major congeners (4-OH-CB107, 3-OH-CB153, 4-OH-CB146, 3'-OH-CB138, 4-OH-CB187, and 4'-OH-CB172) measured from this study was comparable or higher to other maternal groups with the exception of those from the Faroe Islands. In that study, pregnant women had high levels of fish and pilot whale blubber consumption, and their median blood OH-PCB level was higher by about one order of magnitude.

The distribution of individual OH-PCBs in human serum varies geographically as summarized elsewhere (Park et al., 2007). Typically, studies find 4-OH-CB187 as the predominant OH-PCB congener followed by 4-OH-CB107, 3-OH-CB153, 4-OH-CB146, and 3'-OH-CB138. We found 4-OH-CB187 as the primary metabolite followed by 4-OH-CB107 and then 4-OH-CB146. The sum of these three congeners accounted for $91 \pm 9\%$ on average of the Σ_8 OH-PCB. All but one of our samples showed this profile; that one exception showing higher level of 4-OH-CB107 than 4-OH-CB187. Four samples showed higher concentrations of 4-OH-CB146 than 4-OH-CB107. Unique profiles were also observed in other studies; 4-OH-CB107 > 4-OH-CB187 > 4-OH-CB146 in Canadian female Inuit (Sandau et al., 2000) and 4-OH-CB146 > 4-OH-CB107 > 4-OH-CB187 in elderly Swedish fishermen's wives (Weiss et al., 2006). 4-OH-CB120, possibly formed from PCB-118, was also reported as a major metabolite from Yusho population (Linderholm et al., 2007b). These comparisons suggest that biological variability, or variability based on other environmental factors, may be responsible for the metabolite

fingerprints. Indeed, in our study, the average concentration of 4-OH-CB187 was significantly higher than the concentration of some PCB congeners such as PCB-74, 66, 99, 187, 170, 203, and 194 (all $p < 0.05$). We also observed more para-hydroxylated OH-PCB metabolites (e.g. 4-OH-CB107, 4-OH-CB146, and 4-OH-CB187) relative to meta-positioned ones (e.g., 3-OH-CB153, 3'-OH-CB138, and 3'-OH-CB180). This indicates that the formation and/or the retention of para-hydroxylated metabolites is prevalent relative to meta-positioned ones.

Compared to serum from pregnant women living in Michalovce, Slovakia during the period of 2002–2004 (Park et al., 2007), our archived California serum showed proportionally higher concentrations of 4-OH-CB107 and its possible PCB precursor, PCB-118. However, although PCB-118 might contribute to the formation of 4-OH-CB107, the correlation was weaker than expected ($r = 0.32$, $p = 0.06$) (Figure 1). Other pairs of possible PCB precursors and OH-PCB metabolites (PCB-153 and 3-OH-CB153, PCB-153+138 and 4-OH-CB146, and PCB-187 and 4-OH-CB187) showed better correlations ($r = 0.52$ to 0.60 , $p < 0.05$). Weaker correlations of 4-OH-CB107 and PCB-118 compared to other pairs were also observed in other studies, particularly in Faroe Island mothers ($R^2 = 0.13$). 4-OH-CB107, a metabolite of PCB-118, seems unstable, in agreement with the findings of shorter half life of 4-OH-CB107 (e.g., 3.5 days in laboratory rat experiment) relative to the other congeners (e.g., 15 days for 4-OH-CB187) (Malmberg et al., 2004). In addition, it is possible that 4-OH-CB107 is biotransformed from other PCBs such as PCB-105 (Letcher et al., 2000) that were not selected for the list of target PCB congeners in this study.

As expected, the blood levels of OH-PCB metabolites were dependent on PCB levels ($r = 0.58$, $p < 0.05$) but about an order of magnitude lower ($p < 0.05$). The average ratio of Σ_8 OH-PCBs to Σ_{11} PCBs was 0.16 ± 0.09 . The possible PCB precursors were distributed in the following order of abundance; PCB-153 > PCB-138 > PCB-180 > PCB-118 > PCB-170 > PCB-187. However, OH-PCB metabolites distributed as: 4-OH-CB187 > 4-OH-CB107 > 4-OH-CB146 > 3'-OH-CB138 > 3-OH-CB153 > 4'-OH-CB172 suggesting that a 1,2 shift mechanism was a major mechanism for the hydroxylation of PCBs, rather than direct oxygen insertion. 4-OH-CB187 may be biotransformed directly from PCB-187. Recent studies also showed that PCB-183 is biotransformed to 4-OH-CB187 via 1,2 shift, however, in a subsequent study, the authors corrected their previous result claiming that this pathway was unlikely, based on analysis with a special GC capillary column (Ohta et al., 2007). We observed that 4'-OH-CB172 was detected only in trace levels although its possible precursors such as PCB-180 and PCB-170 were present in considerably high levels. This lends more evidence to the idea that the metabolic capacity and pathways of PCB metabolism are complex in humans.

The most dominant OH-PCB congeners, 4-OH-CB187 and 4-OH-CB107, found in the present study are biologically active (Meerts et al., 2002; Negishi et al., 2007; Otake et al., 2007; Park et al., 2009). In the Slovakian population, concentrations of 4-OH-CB107 were 4-times lower than in the present study, and were significantly related to neurodevelopmental deficits at age 16 months (Park et al., 2009). Since 4-OH-CB107 is known to be labile, these results may be puzzling. This congener was also found to lower the brain and blood thyroid hormone levels of mother and fetus in laboratory rats (Meerts et al., 2002) although the level of 4-OH-CB107 dosed was much higher than levels found in this study or in the Slovakia mothers. Interestingly, 7 year old children showed higher levels of 4-OH-CB107 relative to their mothers in Faroe Island (Fängström et al., 2005). Further, 4-OH-CB107 was the predominant metabolite measured in quantifiable levels from the livers of harbor seal pups, stranded along San Francisco Bay (Park et al., 2009). There is no evidence that 4-OH-CB107 is preferentially transferred via the placenta relative to other hydroxylated metabolites. Thus mechanisms of 4-OH-CB107 biotransformation need to be further investigated, including the difference in the enzymatic capability of precursor PCBs (e.g., PCB-105 and PCB-118) between young offsprings and adults (Ginsberg et al., 2004).

Although the majority of OH-PCBs are known to be driven by the biotransformation of PCBs in the body, external sources such as fish intake and the abiotic environment possibly contribute to the body burden of OH-PCB metabolites (Campbell et al., 2003; Hasegawa et al., 2007; Ueno et al., 2007). Ratios of Σ_8 OH-PCBs to Σ_{11} PCBs range from 0.05 to 0.21 and vary geographically (Table 3). A recent study on Yusho population showed the highest ratio (0.35 on average) (Linderholm et al., 2007b), which the authors suggested was possibly due to past exposure to PCBs or induction of cytochrome P450 enzymes. More information will come from our follow up investigation of the entire cohort (about 500 samples). In the follow up study we will also investigate the relationships between other organochlorines and their metabolites, particularly DDT that is also known as a cytochrome P450 inducer.

This is, to our knowledge, the first study to assess OH-PCB metabolite levels in a U.S. population, particularly 1950–60s' mothers exposed to environmental PCBs before regulatory actions to restrict or ban the production and use of PCBs. This study demonstrates that PCBs and their OH-PCB metabolites are still measurable in sera archived from 1950–60s' California mothers. Although maternal serum concentrations of OH-PCB metabolites were lower than parent PCBs, they were comparable to other studies that showed toxicological effects of these metabolites. Certain chemical structure is required to bind to TTR, which involves a hydroxyl group in either the para- or meta-position of a biphenyl ring, adjacent to chlorine atoms on both sides (Lans et al., 1993; Letcher et al., 2000). We found that 1950–60s' California mothers' sera had higher para-positioned OH-PCB metabolites (4-OH-CB107, 4-OH-CB146 and 4-OH-CB187) than meta-positioned OH-PCBs (e.g., 3-OH-CB153, and 3'-OH-CB138, 3'-OH-CB180). Particularly, 4-OH-CB107 and 4-OH-CB187 are of increasing concern due to their thyroid related toxicological effects reported from animal and/or human studies.

In summary, OH-PCBs were detectable in archived sera collected from pregnant California women in the 1960s, showing the feasibility to measure OH-PCB metabolites in the larger cohort. We modified our technique to ensure complete derivatization to improve lower OH-PCB surrogate recoveries. Our future work with the larger study population will focus on the associations between adult health and prenatal exposure to such PCB metabolites that were reported to be toxicologically active.

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Fig. 1.
Correlations between OH-PCBs and suggested PCB precursors.

Table 1

Maternal characteristics of pilot subjects selected from Child Health and Development Studies (1959–1967)

Maternal Characteristic (%)		Maternal Characteristic (%)	
Race		Body Mass Index	
White	53	1st Quartile	0
Non-white	47	Quartiles 2–4	100
Black	30	4th Quartile	20
Non-Black	70	Quartiles 1–3	80
Parity		Farm Residence before Age 15 yrs.	
Primiparous	20	Yes	34
Multiparous	80	No	66
Marital Status		Maternal Smoking Status	
Married	100	Current	40
Unmarried	0	Not Current	60
Total Income			
<\$5,000	15	Ever	50
\$5,000+	85	Never	50
Education		Place of Birth	
<Grade 12	13	U.S. Born	87
Grade 12+	87	Born outside U.S.	13
Occupation			
Housewife	55	Born in California	40
Employed Outside Home	45	Born outside California	60

Table 2
Distributions of PCBs and OH-PCB metabolites in sera collected from 30 California mothers (1959–67)

	High End of PCBs (N=15)			Low End of PCBs (N=15)		
	min	med	max	min	med	Max
PCBs (ng/mL wet wt)						
PCB-74	0.13	0.24	0.33	0.04	0.15	0.23
PCB-66	0.01	0.10	0.17	<0.01	0.02	0.10
PCB-99	0.06	0.24	0.51	0.03	0.13	0.28
* PCB-118	0.35	0.49	1.35	0.14	0.29	0.49
* PCB-153	0.75	0.85	1.79	0.16	0.43	0.47
* PCB-138	0.66	0.82	1.62	0.16	0.40	0.45
* PCB-187	0.11	0.22	0.54	0.04	0.10	0.12
* PCB-180	0.51	0.60	1.03	0.10	0.31	0.40
* PCB-170	0.17	0.23	0.43	0.03	0.10	0.12
PCB-203	0.04	0.12	0.19	<0.01	0.06	0.14
PCB-194	0.07	0.11	0.20	<0.01	0.05	0.10
Σ_{11} PCB	3.42	3.85	7.99	0.74	2.08	2.67
OH-PCBs (ng/mL wet wt)						
4'-OH-CB107	<0.01	0.08	0.24	0.02	0.05	0.30
3-OH-CB153	<0.01	0.02	0.08	<0.01	<0.01	0.04
4-OH-CB146	<0.01	0.05	0.15	<0.01	0.03	0.06
3-OH-CB138	<0.01	0.03	0.09	<0.01	0.00	0.03
4-OH-CB130	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
4-OH-CB187	0.12	0.28	0.56	0.05	0.19	0.27
3'-OH-CB180	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
4'-OH-CB172	<0.01	<0.01	0.04	<0.01	<0.01	<0.01
Σ_8 OH-PCBs	0.16	0.44	0.98	0.12	0.25	0.69
OH-PCB/PCB	0.05	0.11	0.19	0.07	0.14	0.35

* Possible precursors of OH-PCB metabolites presented in this study.

<0.01 indicates below quantification limit (LOQ) for PCBs and method detection limit (MDL) for OH-PCBs.

Table 3
Median concentrations of OH-PCBs (ng/g wet wt.) and average ratios of OH-PCB to PCBs from worldwide maternal sera

	Netherlands ^a	Sweden ^b	Faroe Island ^c	Michalovec, Slovakia ^d	Svidnik, Slovakia ^d	Japan ^e	California ^f , USA
<i>OH-PCBs</i>							
4-OH-CB107	0.05	0.01	0.47	0.03	0.02	0.00	0.10
3-OH-CB153	0.03	0.01	0.29	0.07	0.04	0.00	0.02
4-OH-CB146	0.06	0.03	0.79	0.11	0.05	0.01	0.04
3'-OH-CB138	0.04	0.01	0.33	0.07	0.03	0.00	0.02
4-OH-CB187	0.12	0.04	1.50	0.20	0.11	0.01	0.23
4'-OH-CB172	0.01	0.00	n/a	0.04	0.02	0.00	<0.01
Sum	0.32	0.12	3.38	0.55	0.27	0.03	0.42
Ratio of OH-PCB/PCB	0.18	0.10	0.21	0.09	0.08	0.10	0.14

^aSoechitram et al, 2004,
^bGuvenuis et al, 2003,
^cFängström et al, 2005,
^dPark et al, 2007,
^eEnomoto et al, 2007,
^fThis study