Polychlorinated biphenyls (PCBs) have been well studied for possible effects on newborns and infants after it was determined that PCBs could effectively pass through the placental barrier and that they were associated with lower birth weights (1). Jacobson et al. (2) found that children exposed in utero to PCBs had delayed central nervous system functioning. Subsequent studies then confirmed that for this same cohort, reductions in cognitive function were associated with higher in utero PCB exposure at 4 years of age (3), followed by lower IQs at 11 years of age (4). These studies seem to indicate a potential link between PCBs and neurodevelopment.

Although many theories exist about how PCBs affect neurodevelopment, the main hypothesis involves disruption of thyroid hormone homeostasis (5). Thyroid hormones regulate neuronal proliferation and cell migration and differentiation, including control over when differentiation begins and when cell proliferation ends (6). Studies in the rat showed that transport of thyroid hormones to the brain requires thyroxine (T₄) to pass through the blood–brain barrier bound to the thyroid hormone transport protein transthyretin (TTR) (7). Although PCBs show some binding affinity for TTR (8), hydroxylated metabolites of PCBs (HO-PCBs) have much higher in vitro binding affinities that can be as high as 12 times the TTR protein affinity (9). The main metabolites that have been previously identified in plasma (10). Thus, circulating retinol concentrations can also be affected by PCB and HO-PCB exposure (18).

Similar toxicological properties to HO-PCBs (10,14,15). A recent review described the formation and retention of HO-PCBs and the main metabolites that have been previously identified in plasma (16). HO-PCBs decrease circulating levels of thyroid hormones in rats through competitive binding to TTR (17). TTR is also responsible for retinol transport by forming a dimer with retinol-binding protein. Thus, circulating retinol concentrations can also be affected by PCB and HO-PCB exposure (18).

The fetus may be especially vulnerable to PCB and HO-PCB exposure. When fetal mice were exposed in utero to 4′-HO-CB79, a metabolite of polychlorinated biphenyl congener number 77 (CB77), both maternal and fetal plasma T₄ levels decreased significantly compared to controls (19). In this same study, fetal plasma had twice the 4′-HO-CB79 concentration of the maternal plasma (20). These experiments were recently repeated on pregnant rats orally exposed to 4-HO-CB107 (21), one of the main HO-PCBs found in human plasma (12,13). In that study, both maternal and fetal plasma concentrations of thyroid hormones were reduced by exposure to 4-HO-CB107. Fetal total T₄ concentrations decreased by 89% of that of the controls (21). The decreased plasma T₄ levels also decreased forebrain and cerebellum T₄ concentrations compared to controls (21), which may lead to a neurodevelopmental effect. PCB also decreases brain T₄ availability in rats (22). Another interesting finding of the 4-HO-CB107 rat dosing study was

Concentrations of polychlorinated biphenyls (PCBs), hydroxylated metabolites of PCBs (HO-PCBs) and octachlorostyrene (4-HO-HpCS), and pentachlorophenol (PCP) were determined in umbilical cord plasma samples from three different regions of Québec. The regions studied included two coastal areas where exposure to PCBs is high because of marine-food–based diets—Nunavik (Inuit people) and the Lower North Shore of the Gulf of St. Lawrence (subsistence fishermen)—and a southern Québec urban center where PCB exposure is at background levels (Québec City). The main chlorinated phenolic compound in all regions was PCB. Concentrations of PCP were not significantly different among regions (geometric mean concentration 1,670 pg/g, range 628–7,680 pg/g wet weight in plasma). The ratio of PCP to polychlorinated biphenyl congeners number 153 (CB153) concentration ranged from 0.72 to 42.3. Sum HO-PCB (ΣHO-PCBs) concentrations were different among regions, with geometric mean concentrations of 553 (range 238–1,750), 286 (103–788), and 234 (147–464) pg/g wet weight plasma for the Lower North Shore, Nunavik, and the southern Québec groups, respectively. Lower North Shore samples also had the highest geometric mean concentration of sum PCBs (ΣPCBs) 2,710 (525–7,720) pg/g wet weight plasma. Sum PCB concentrations for Nunavik samples and southern samples were 1,510 (309–6,230) and 843 (290–1,650) pg/g wet weight plasma. Concentrations (log transformed) of ΣHO-PCBs and ΣPCBs were significantly correlated (r = 0.62, p < 0.001), as were concentrations of all major individual PCB congeners and individual PCB congeners. In Nunavik and Lower North Shore samples, free thyroxine (T₄) concentrations (log transformed) were negatively correlated with the sum of quantitated chlorinated phenolic compounds (sum PCB and ΣHO-PCBs; r = −0.47, p = 0.01, n = 20) and were not correlated with any PCB congener or ΣPCBs. This suggests that PCB and HO-PCBs are possibly altering thyroid hormone status in newborns, which could lead to neurodevelopmental effects in infants. Further studies are needed to examine the effects of chlorinated phenolic compounds on thyroid hormone status in newborns. Key words: hydroxylated metabolites, pentachlorophenol, polychlorinated biphenyls, retinol, thyroxine, umbilical cord plasma. Environ Health Perspect 110:411–417 (2002). [online 12 March 2002] http://ehpnet1.niehs.nih.gov/docs/2002/110/p411-417/sandau.abstract.html
an accumulation of 4-HO-CB107 in fetal plasma, liver, and brain (21).

Thus, prenatal exposure to PCBs, HO-PCBs, and PCP may all lead to thyroid hormone disruption and possibly neurodevelopmental effects. Analysis of umbilical cord plasma is of special interest because it provides a direct indication of in utero exposure to developmental toxicants. PCBs have been measured previously in umbilical cord plasma (23,24), but the present study, to our knowledge, is one of the first studies to examine chlorinated phenolic compounds in this biological medium. Participants were from populations with different PCB exposures caused by differences in dietary habits. Retinol and thyroid hormone status [triiodothyronine (T3), free T4, thyroid-stimulating hormone (TSH), and thyroxine-binding globulin (TBG)] were determined in samples from remote maritime populations, so the relationship between chlorinated phenolic compounds and these biological markers could be explored.

Materials and Methods

Samples. Plasma samples were obtained during various umbilical cord blood surveys conducted from 1993 to 1996 in Quebec (25,26). These surveys took place in Nunavik (northern Quebec), the Lower North Shore of the Gulf of St. Lawrence, and southern Quebec (Quebec City; Figure 1). The population in the Quebec City area receives background PCB exposure similar to that of the general population of Canada, whereas the former two coastal areas comprise small settlements of people with unusually high PCB exposure. The traditional diet of Nunavik Inuit includes seal and beluga blubber, which contain concentrations of PCBs in the order of several milligrams per kilogram (27,28). The diet of the Lower North Shore subsistence fish-eating population includes fish, sea mammals, and seabird eggs (28). Ten samples from each region were randomly selected for chlorinated phenolic compound and PCB residue analysis from all samples collected during the surveys. Nunavik samples were all from Inuit newborns, southern Quebec samples from Caucasian newborns, and Lower North Shore samples from three Caucasians and seven aboriginal neonates.

Standards and chemicals. PCBs are numbered according to the numbering scheme as described by Ballschmiter and Zell (29). Hydroxylated PCBs and their methoxy derivatives are given the appropriate Ballschmiter PCB number according to their chlorination pattern. The HO- or MeO-functional groups are numbered thereafter, as described by Letcher et al. (16). Note that the numbering of two congeners in our previous publication (12) has changed: 4-HO-CB109 is now 4-HO-CB107, and 4-HO-CB107 is now 4-HO-CB108.

The following [13C12]-labeled standards were acquired from Wellington Laboratories (Guelph, ON, Canada) and were used as an internal recovery standard mixture: 4´-HO-CB120, 4´-HO-CB159, 4´-HO-CB172, and 4-HO-CB187. [13C6]PCP was purchased from Cambridge Isotope Laboratories (Andover, MA, USA) and was used for PCP quantitation. Labeled PCBs ([13C12]CB-118, 153, 180, and 194) were used as internal recovery standards, and [13C12]CB-138 was used as the performance standard for PCB analysis. [13C12]PCB standards

| Compound | Nunavik | Lower North Shore | Southern Quebec |
|----------|---------|----------------|---------------|
| PCP      | 1,870   | 28             | 1,740         |
| 4-HO-HpCS| 31      | 13             | 12            |
| HO-PCBs  |         |                 |               |
| 4-HO-CB187| 47      | 13             | 10            |
| 4-HO-CB146| 37      | 13             | 12            |
| 4-HO-CB153| 19      | 6              | 6             |
| 4-HO-CB107| 12      | 3              | 11            |
| 4-HO-CB108| 10      | 3              | 3             |
| 4-HO-CB172| 10      | 3              | 4             |
| 4-HO-CB187| 6       | 3              | 4             |
| 3-HO-CB187| 4       | 3              | 3             |
| 4-HO-CB193| 3       | 2              | 2             |
| 4-HO-CB208| 2       | 1              | 1             |
| 3-HO-CB193| 2       | 1              | 1             |
| 4-HO-CB199| 1       | 1              | 1             |

Table 1. Concentrations (picograms per gram wet weight plasma) of halogenated phenolic compounds and 2PCBs in umbilical cord plasma from three regions in Quebec (n = 10 for each region).

Figure 1. Sample locations for the three populations: Nunavik (northern Quebec), Lower North Shore (Gulf of St. Lawrence), and southern Quebec (Quebec City).
Table 2. Concentrations (picograms per gram wet weight plasma) of 49 PCB congeners in umbilical cord plasma from three regions in Québec (n = 10 for each region).

| Congener | Nunavik | Lower North Shore | Southern Québec |
|----------|---------|-------------------|-----------------|
|         | GM      | Min | Max | GM      | Min | Max | GM      | Min | Max |
| CB92     | 9       | ND  | 60  | 7       | ND  | 197 | 9       | ND  | 13  |
| CB84     | 27      | 5   | 141 | 36      | 6   | 251 | 16      | 9   | 31  |
| CB101/90 | 49      | 11  | 262 | 87      | 13  | 786 | 44      | 17  | 181 |
| CB199    | 100     | 16  | 1,120| 174     | 17  | 1,630| 38      | 16  | 31  |
| CB97     | 10      | ND  | 167 | 12      | ND  | 232 | 8       | 16  | 19  |
| CB87     | 28      | 5   | 236 | 63      | 8   | 436 | 13      | 8   | 27  |
| CB95     | 6       | 2   | 115 | 14      | 2   | 150 | 6       | 9   | 38  |
| CB110    | 42      | 8   | 403 | 79      | 10  | 709 | 44      | 18  | 502 |
| CB118    | 67      | 19  | 402 | 155     | 30  | 673 | 35      | 9   | 81  |
| CB105    | 19      | 6   | 300 | 37      | 7   | 155 | 11      | 6   | 31  |
| CB136    | 13      | 1   | 316 | 15      | 2   | 651 | 12      | 1   | 536 |
| CB151    | 7       | 3   | 34  | 11      | 4   | 68  | 8       | ND  | 14  |
| CB144/135| 17      | ND  | 97  | 27      | ND  | 713 | 13      | ND  | 22  |
| CB149    | 20      | 9   | 71  | 33      | ND  | 96  | 30      | 14  | 103 |
| CB134    | 8       | 1   | 35  | 11      | 2   | 86  | 3       | ND  | 18  |
| CB146    | 23      | 5   | 98  | 54      | 15  | 178 | 11      | 6   | 54  |
| CB153    | 262     | 49  | 1,340| 430     | 107 | 1,350| 104     | 30  | 199 |
| CB141    | 3       | 2   | 20  | 5       | 3   | 13  | 5       | 1   | 8   |
| CB130    | 6       | 2   | 20  | 9       | 3   | 30  | 2       | ND  | 4   |
| CB137    | 4       | 1   | 13  | 6       | 2   | 15  | 2       | ND  | 3   |
| CB138/163| 157     | 36  | 712 | 232     | 62  | 704 | 54      | 11  | 110 |
| CB158    | 5       | 2   | 18  | 8       | 2   | 17  | 3       | 1   | 6   |
| CB178    | 1       | ND  | 9   | 1       | ND  | 3   | 1       | ND  | 1   |
| CB128    | 7       | 3   | 27  | 16      | 6   | 47  | 4       | ND  | 7   |
| CB156    | 27      | 5   | 94  | 40      | 17  | 104 | 11      | 2   | 19  |
| CB157    | 8       | 2   | 26  | 17      | 7   | 45  | 5       | ND  | 8   |
| CB179    | 2       | 1   | 5   | 2       | ND  | 4   | 2       | ND  | 5   |
| CB176    | 1       | ND  | 2   | 1       | ND  | 1   | ND      |     |     |
| CB178    | 2 < 1  | 27  | 1 < 1| 20      | 1   | 14  | ND      |     |     |
| CB187/182| 39      | 7   | 146 | 102     | 24  | 297 | 38      | 13  | 228 |
| CB183    | 14      | 4   | 135 | 23      | 6   | 57  | 7       | 2   | 12  |
| CB195    | < 1     | < 1 | 2   | 1       | ND  | 1   | ND      |     |     |
| CB174    | 3       | 2   | 12  | 4       | 2   | 9   | 6       | 1   | 11  |
| CB175    | 5       | 2   | 13  | 10      | 4   | 18  | 4       | 1   | 7   |
| CB171    | 3       | 1   | 7   | 6       | 2   | 13  | 2       | 1   | 4   |
| CB172    | 2 < 1  | 10  | 6   | 2       | 14  | 1   | ND      |     |     |
| CB180    | 118     | 33  | 663 | 146     | 43  | 501 | 40      | 8   | 84  |
| CB193    | 3       | < 1 | 53  | 5       | 1   | 23  | 1       | < 1 |     |
| CB191    | < 1     | < 1 | 5   | 1 < 1  | 7   | 1   | < 1     | < 1 |     |
| CB170/190| 21      | 4   | 74  | 39      | 13  | 87  | 13      | 3   | 22  |
| CB202    | 4       | 2   | 21  | 5       | 3   | 10  | 2       | ND  | 3   |
| CB201    | 1 < 1  | 22  | 2   | 1       | 4   | 1   | ND      |     |     |
| CB199    | 1       | ND  | 4   | 1       | ND  | 4   | 2       | ND  | 6   |
| CB201    | 4       | 1   | 17  | 10      | 2   | 31  | 7       | 4   | 11  |
| CB196/203| 8       | 2   | 26  | 33      | 7   | 113 | 15      | 6   | 95  |
| CB195    | 3       | 1   | 26  | 5       | 1   | 32  | 2       | ND  | 2   |
| CB194    | 9       | 2   | 23  | 17      | 7   | 55  | 11      | 2   | 21  |
| CB206    | 3       | 1   | 10  | 6       | 3   | 12  | 1       | ND  | 2   |
| CB209    | 2       | < 1 | 1   | < 1     | 2   | < 1 | ND      |     |     |
| Sum PCBs | 1,510   | 309 | 6,230| 2,710   | 525 | 7,720| 843     | 290 | 1,650 |

Abbreviations: GM, geometric mean; Max, maximum; Min, minimum; ND, not detected.

Table 3. Concentrations of retinol, thyroid hormones, and TBG in umbilical cord plasma samples from three regions in Québec (n = 10 for each region).

| Nutrient  | Nunavik | Lower North Shore | Southern Québec |
|-----------|---------|-------------------|-----------------|
|           | GM      | Min | Max | GM      | Min | Max | GM      | Min | Max |
| Retinol (µg/L) | 160  | 61  | 250 | 160  | 89  | 290 | 190    | 110 | 330 |
| F<sub>T4</sub> (pmol/L) | 16  | 13  | 22 | 17  | 9.6 | 21 | 190    | 110 | 330 |
| T<sub>3</sub> (nmol/L) | 0.84 | 0.45 | 1.20 | 1.04 | 0.20 | 0.78 | 190    | 110 | 330 |
| T<sub>SH</sub> (µmol/L) | 7.7  | 3.9 | 19 | 6.7  | 3.9 | 15 | 190    | 110 | 330 |
| T<sub>BG</sub> (nmol/L) | 920 | 590 | 1,300 | 880 | 620 | 1,300 | 190 | 110 | 330 |

Abbreviations: GM, geometric mean; Max, maximum; Min, minimum; NA, not analyzed.
Results

Recoveries of the internal recovery standards ([13C6]PCP and [13C12]HO-PCBs and PCBs) were in the range of 75–104%. Mean recovery of phenolic internal standards was better than 87%. All concentrations were recovery corrected.

With the Liliefors test for normal distribution, the chemical residue data were not normally distributed. Thus, all data (including retinol and thyroid hormone concentrations) were log transformed before statistical analysis. The regional concentration data are summarized using geometric means along with minimum and maximum values (Tables 1–3).

Thirty compounds were characterized as HO-PCBs in the umbilical cord plasma samples. Concentrations of PCP and identified HO-PCB congeners are listed in Table 1. Two congeners, 4-HO-CB107 and 4′-HO-CB108, coelute and were quantitated as a single peak. The peak is likely 4-HO-CB107, as demonstrated in previous studies (16,30). ΣHO-PCBs represents a sum of all identified HO-PCBs and all compounds characterized as HO-PCBs. Unidentified HO-PCBs were quantitated using relative response factors as described previously (12). ΣHO-PCBs were analyzed for regional differences by multiple analysis of variance. Lower North Shore samples had the highest mean concentration of ΣHO-PCBs, which was significantly higher than concentrations in southern Quebec samples using the Sheffe test (p = 0.01). The Nunavik samples were not significantly different from southern samples (p = 0.8) or from Lower North Shore samples (p = 0.06). PCP concentrations were highest in Nunavik samples but were not significantly different among regions.

We also found another compound recently identified as a major chlorinated phenolic compound in polar bear plasma, 4-hydroxy-heptachlorostyrene (4-HO-HpCS) (31), in the human umbilical cord plasma samples (Table 1). This compound was determined in all umbilical cord plasma samples. No quantitative standard was available at the time of analysis, so we estimated concentrations of 4-HO-HpCS using the average heptachlorinated MeO-PCB response factor. The geometric mean concentrations in Nunavik and Lower North Shore samples were about six times higher than in southern Quebec samples.

Forty-nine PCB congeners with 5 or more chlorines were above the detection limit in most of the umbilical cord plasma samples. Concentrations of all 49 PCBs and ΣPCBs (sum of all congeners) are listed in Table 2. The ratio of ΣHO-PCBs to ΣPCBs is given in Table 1. ΣPCBs were highest in Lower North Shore plasma samples and Nunavik samples, but only Lower North Shore samples were significantly different (p = 0.01) from southern Quebec samples by the Sheffe test. The mean ratio of ΣHO-PCB metabolites to PCBs was highest in southern Quebec samples and lowest in Nunavik samples, but the ratio was not statistically different among regions. ΣHO-PCBs and ΣPCBs concentrations were highly correlated in umbilical cord plasma (r = 0.69, p < 0.001), as shown in Figure 2. Fractions of the main identified HO-PCBs of ΣHO-PCBs are shown in Figure 3. The PCBs from which the main metabolites may be formed are listed above each metabolite in Figure 3.

Figure 4 shows the correlation between one of the main HO-PCBs, 4-HO-CB1146, and its potential precursor PCBs. The metabolite was significantly correlated (p < 0.001) with all possible precursor PCBs and was significantly correlated with many non-related PCBs (not shown).

Mean retinol concentrations were lowest in Nunavik samples and highest in southern Quebec samples, but differences between the regions were not statistically significant (Table 3). No significant correlations were observed between concentrations of retinol and any individual HO-PCB or PCB congeners, ΣHO-PCBs, sum of all chlorinated phenolic compounds, or ΣPCBs.

Plasma concentrations of T3, free T4, TSH, and TBG were not significantly different between Lower North Shore and Nunavik samples (Table 3). Concentrations of main HO-PCBs and PCBs were not significantly correlated with thyroid hormone markers. In contrast, PCB concentrations were negatively correlated with T3 (r = –0.55, p = 0.01), TBG (r = –0.44, p = 0.05), and free T4 levels (r = –0.51, p = 0.02). Figure 5 shows the statistically significant inverse correlation (r = –0.47, p = 0.01) between free T4 concentrations and log-transformed sum of all chlorinated phenolic compounds (sum of PCP and ΣHO-PCBs). The relationship was not improved using log-free T4 and log-sum molar concentration of phenolic compounds. The sum of all chlorinated phenolic compounds was also negatively associated with T1 concentrations (r = –0.48, p = 0.03). Concentrations of ΣPCBs and ΣHO-PCBs were both negatively correlated with TSH concentrations (r = –0.46, p = 0.04 and r = –0.45, p = 0.04, respectively).

Discussion

Hydroxylated metabolites and other chlorinated phenolic compounds, to our knowledge, have never been examined in umbilical
cord plasma. We found that PCP was the most abundant phenolic compound in all three regions, representing an average of 78%, 66%, and 82% of the concentration of the sum of all quantitated chlorinated phenolic compounds in the Nunavik, Lower North Shore, and southern Quebec groups, respectively. Mean PCP concentrations were similar among groups, and individual values ranged from 628 to 7,680 pg/g wet weight. We previously reported PCP as the dominant chlorinated phenolic compound in blood samples from Nunavik and southern Quebec adults (12). Thus, PCP may supersede HO-PCBs as the chlorinated phenolic compound of highest concern in humans.

PCP and its salts have been used extensively as wood preservatives, biocides, and disinfectants (32). PCP use has been curtailed since the late 1970s and has been banned in some countries, such as Sweden (1977) and Germany (1987) (32). The use of PCP has been restricted in Canada since 1981. The main exposure to PCP for nonoccupationally exposed individuals is through the diet (33). Another significant source of PCP may occur through the metabolism of hexachlorobenzene (34). Plasma is the most important compartment for PCP storage. In dosed rats, 99% of PCP is bound to plasma proteins (35). In human volunteers, the percentage of PCP bound to plasma proteins was estimated to be 96% (36).

PCP can induce deleterious effects on several organs or tissues. Increased lymphocyte responses were noted in patients with high PCP blood levels (37). PCP can be metabolized to reactive quinone metabolites (38) with possible covalent binding to crude liver homogenates and isolated liver proteins in vitro (39). PCP has twice the affinity of T₄ to TTR (10) and has been shown to decrease circulating T₄ levels in rams exposed from conception (40). PCP also affects thyroid hormone metabolism by competitively inhibiting iodothyronine sulfation in vitro (41). In the present study, the sum of plasma concentrations of phenolic compounds, the major part being PCP, were negatively correlated to free T₄ and T₃ plasma levels. This suggests that PCP and perhaps other chlorinated phenolic compounds can alter thyroid hormone status in newborns, which in turn could lead to adverse neurodevelopmental effects in infants.

Another chlorinated phenolic compound recently identified by our laboratory in polar bear plasma, 4-HO-HpCS (31), was also found in all umbilical cord plasma samples analyzed. This is the first time this compound has been shown to be present in human plasma. The likely precursor for this compound is octachlorostyrene, an industrial byproduct. The fact that lower concentrations of 4-HO-HpCs were found in the southern Quebec group than in the Nunavik and Lower North Shore groups suggests that the likely source of exposure is the consumption of species from the marine food chain. Sandau et al. (31) showed that this compound had an affinity similar to T₄ for binding to TTR, which is slightly less than PCP (10) and lower than most HO-PCBs that have been determined (42).

Concentrations of ΣHO-PCBs in umbilical plasma were highest in the Lower North Shore samples. More than 30 compounds were identified as HO-PCBs, of which 11 were positively identified with authentic standards. Three more HO-PCBs found in humans (16) were tentatively identified but could not be confirmed because no authentic standards were available. The main metabolite in 27 of the 30 samples was 4-HO-CB187. This compound was also the dominant metabolite in fish eaters from Sweden, Black-footed and Laysan albatross, and polar bear (13,43,44). Two possible parent PCBs can form 4-HO-CB187 through two different hydroxylation mechanisms. The first involves the direct insertion (45) of a hydroxyl group onto the para position of CB187. Direct insertion has been demonstrated to occur in in vitro metabolism studies of halobenzenes (46) and CB52 (47). CB187 is an abundant congener found in biota and accounted for a mean of 3.4% of the ΣPCBs in all the umbilical cord plasma samples. It is found as a small percentage (0.54%) in the Aroclor 1254 mixture, but is more abundant in Aroclor 1260 (5.4%) (48). The second mechanism of oxidation is the formation of a 3,4 (meta-para)-epoxide in CB183 followed by a 3,4 shift of chlorine to the meta position similar to the National Institutes of Health (NIH) shift of 1H first described by Guroff et al. (49). Epoxide formation in the metabolism of PCBs has been demonstrated in in vitro studies (50) as well as in vivo studies (17) using CB77 as substrate. CB183 composed a mean of 0.8% of the ΣPCBs in the umbilical cord plasma and constitutes approximately 0.2% and 2.4% of Aroclor 1254 and 1260 mixtures, respectively (48). Interestingly, the major PCB metabolite in umbilical cord plasma, 4-HO-CB187, is formed from PCBs that make up a small percentage of the ΣPCBs in the samples.

The second most abundant metabolite in umbilical cord plasma was 4-HO-CB146. This metabolite can be formed by direct insertion onto CB146 or by NIH shift of

Figure 4. Relationship between log-transformed concentrations of precursor PCBs (CB146, 153, and 138) and the second most abundant metabolite in umbilical cord plasma, 4-HO-CB146. CB138 coelutes with CB163 and they were quantitated as a single peak.

Figure 5. Relationship between log-transformed concentrations of free T₄ and sum of all chlorinated phenolic compounds (sum PCP and ΣHO-PCBs).
chlorine in the metabolism of CB138 or CB153. These three parent PCBs compose a large percentage of the ΣPCBs (between 11 and 47%) quantitated in all the samples. CB153 (mean 15% of ΣPCBs) and CB138 (mean 8.3% of ΣPCBs) are the two most abundant PCBs determined in the plasma samples and are major components in Aroclor mixtures (48). All three potential parent PCBs were significantly (p < 0.001) correlated with 4-HO-CB146 (Figure 4).

The third most abundant metabolite was 4-HO-CB107, which can be formed from CB107 (direct insertion), CB105 (NIH-Cl shift), or CB118 (NIH-Cl shift). Both CB105 and CB118 are major congeners in Aroclor 1254, composing 5.2% and 10.5% of the total (48). CB107 is a minor congener in Aroclor 1254 (0.6%), and it is rarely found in environmental samples, including these umbilical cord plasma samples. Concentrations of the potential parent PCBs CB105 (mean 1.3% of ΣPCBs) and CB118 (mean 4.8% of ΣPCBs) were significantly correlated (r = 0.69, r = 0.81, respectively; p < 0.001) with 4-HO-CB107 concentrations. In contrast to our study results, 4-HO-CB107 was previously found to be the main metabolite in adult Inuit whole blood, Latvian fish consumers, Baltic seals, white-tailed eagles, and rats dosed with Aroclor 1254 (12,13,16,30).

The relationship between metabolites and their potential precursor PCBs could not be resolved further using multiple-step regression analysis (forward or backward). Concentrations of major metabolites were highly correlated with all PCBs, even unrelated congeners. Therefore, it is not possible from the present data to determine which congeners are the precursors of the metabolites—that is, the relative importance of NIH chlorine shift to direct insertion.

Hydroxylated PCB patterns vary among individuals (12,13). This variation can be caused by selective retention or selective formation of metabolites or by differences in PCB exposure. The retention of specific HO-PCBs is probably similar for all humans. The main structural requirement for retention is the capability to bind to TTR (9). This requirement is thought to occur because of the para position of the biphenyl ring, but not exclusively, because meta-substituted metabolites are also found in plasma. Humans have varying concentrations of TTR in plasma, and some genetic abnormalities are known (50). Generally, concentrations are in excess of the metabolite to HO-PCBs (12). Thus, the main determinant of the pattern of HO-PCBs in blood is likely the formation of metabolites from the parent PCB congeners.

It was interesting to note that the geometric mean ratio of 2HO-PCBs to ΣPCBs concentrations was similar (~ 0.2) among regions. The ratio was twice that found in a previous study involving whole blood of Canadian Inuit (0.11) (12). Because the relationship of the log-transformed concentrations in umbilical cord plasma had a slope of between 0.5 and 0.6 (Figures 2 and 4), the ratio of metabolites to PCBs decreased with increasing PCB concentrations. The range was from approximately 0.4 at low PCB concentrations (500 pg/g; Figure 2) to approximately 0.1 at high PCB concentrations (5,000 pg/g; Figure 2), similar to that found in adult whole blood. There was no apparent effect of PCB concentration on the ratio of metabolites to PCBs in adult whole blood (15). The generally higher ratio in umbilical cord plasma samples may reflect the difference in composition of fetal and adult blood. For example, umbilical cord plasma has approximately half the lipid content and less transthyretin than adult plasma (52). It has been shown previously that PCBs are most concentrated in plasma lipoproteins (53). Another possible explanation for the differences in the metabolite/PCB ratio in adult and fetal blood could involve enhanced placental transfer of HO-PCBs from the mother. The transfer of PCB metabolites from dosed mice to the fetus was tested by Sinjari et al. (20). They showed that 4'-HO-CB79 concentrations in fetal plasma were twice that of the maternal plasma, 24 hr after exposure, indicating enhanced transport of the metabolite, which likely occurs through binding to TTR.

When the individual chemical residual data were compared to thyroid hormone markers, only PCP concentration was significantly correlated with all PCBs, even unre- lated metabolites. This may be partly responsible for this decrease.

TTR has been shown to be important in T4 transport in cerebral spinal fluid (7). If chlorinated phenolic compounds can significantly alter plasma levels of TTR-bound T4, this may lead to brain thyroid hormone deficiencies in utero, possibly affecting brain develop- ment (57). TTR is also important in thyroid hormone transport across the placental barrier (58). Maternal sources of thyroid hormones are thought to influence fetal brain development (59). The binding of metabolites to TTR may also improve transport of halogenated phenolic compounds across the placenta, as has been shown in mice (19). Thus, phenolic compounds may be able to disrupt maternal sources of thyroid hormones, pene- trate into fetal circulation, and disrupt local thyroid hormone supply in the developing fetus. The potential of PCP and HO-PCBs to disrupt thyroid hormone homeostasis in the developing fetus warrants further investigation to confirm the effects observed in the present study. A study is currently underway that will examine the relationship between halogenated phenolic compounds, thyroid hormones, and retinol concentrations in newborns from a larger cohort.

**References and Notes**

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