Environ Health Prev Med (2013) 18:205–214
DOI 10.1007/s12199-012-0306-6

REGULAR ARTICLE

Serum levels of hydroxylated PCBs, PCBs and thyroid hormone measures of Japanese pregnant women

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Received: 20 June 2012 / Accepted: 4 September 2012 / Published online: 30 September 2012

Abstract
Objectives The purpose of this study was to investigate the associations between serum concentrations of hydroxylated PCBs (OH-PCBs) and PCBs and measures of thyroid hormone status of Japanese pregnant women.
Methods The concentrations of free thyroxine (fT4), thyroid stimulating hormone (TSH), and thyroxine binding globulin (TBG) as well as 16 OH-PCB isomers and 29 PCB isomers were analyzed in the serum of 129 women sampled in the first trimester of gestation. Dietary and lifestyle information of the subjects was obtained by self-administered questionnaire. Multiple regression analysis was performed using measures of thyroid hormones as the dependent variable and serum levels of OH-PCBs/PCBs, urinary iodine concentration, and other potential covariates (age, BMI, smoking, etc.) as independent variables.
Results Geometric mean (GM) concentration of the sum of 16 isomers of OH-PCBs was 120 pg/g wet wt. and that of 29 isomers of PCBs was 68 ng/g lipid wt., respectively, in the serum of the subjects. Iodine nutrition was considered adequate to high from urinary iodine level (GM, 370 μg/g creatinine). The mean concentration of TSH, fT4 and TBG was 1.34 ± 1.37 μIU/mL, 1.22 ± 0.16 ng/dL and 33.0 ± 6.4 μg/mL, respectively, with a small number of subjects who were outside the reference range. Multiple regression analysis revealed that serum concentrations of OH-PCBs/PCBs were not significantly associated with any of the measures of thyroid hormone status.
Conclusions Exposure/body burden of OH-PCBs and PCBs at environmental levels does not have a measurable effect on thyroid hormones.

Keywords Hydroxylated polychlorinated biphenyls · Polychlorinated biphenyls · Thyroid hormone · Urinary iodine · Pregnant woman

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Introduction

Thyroid hormone is essential for normal development of fetal brain [1]. Congenital hypothyroidism, cretinism, causes mental retardation. Maternal thyroid deficiency can also result in abnormal development of newborns. Moreover, Haddow et al. [2] reported that even a mild maternal hypothyroidism adversely affects IQ of her offspring. The first trimester of pregnancy is a critical period for fetal development because the fetus is dependent on maternal thyroid hormone supply during this period [3, 4].

Previous studies indicated that prenatal exposure to polychlorinated biphenyls (PCBs) adversely affects children's intelligence and neurobehavioral functions [5]. One of the possible mechanisms for this effect was deteriorated thyroid hormone status caused by the toxic actions of some

congeners of PCBs [6]. A number of previous in vitro and in vivo studies on the effect of PCBs on thyroid hormone status suggested several mechanisms of action [7]. There are also a number of human studies which were carried out to see if association was found between thyroid hormone status and exposure to PCBs, however, the results were inconsistent across the studies [8, 9].

There were also indications that hydroxylated metabolites of PCBs, OH-PCBs, can impair thyroid hormone function; it is reported that OH-PCBs have stronger affinity to thyroid hormone transporter protein than the natural ligand (T4) and PCBs [10], and that OH-PCBs can inhibit sulfotransferase activity for the conjugation of thyroid hormone [11]. However, the number of human studies on the association between OH-PCBs exposure and thyroid hormone status is much more limited than that on PCBs–thyroid relationship [12–17]. It is, therefore, the aim of this study to see if exposure to OH-PCB affects thyroid hormone status of pregnant women taking its relevance to developmental effect of offspring into consideration.

Iodine, the essential component of thyroid hormone, is indispensable for neurodevelopment [18], and Dallaire et al. [16, 19] pointed out that iodine intake could be one of the possible modifying factors of PCBs–thyroid hormone relationship. However, there are only two studies that examined thyroid effect of PCBs exposure while taking iodine nutritional status into consideration [20, 21]. The Japanese are generally considered iodine-adequate, or even excessive, due to habitual consumption of iodine rich foods, such as seaweed and fish. Not only iodine deficiency but also iodine excess can affect thyroid function [22]. Thus, in this study we assessed iodine status of subjects as a potential covariate of the association between OH-PCBs and PCBs exposure and thyroid hormone status.

Subjects and methods

Participants

One hundred and twenty-nine pregnant women of 10–12 gestational weeks were recruited at a university hospital in Tokyo during the period 2009–2011. The criteria for eligibility was that the woman was of reproductive age (20–50), lived in Tokyo Metropolitan Area, and had no known diseases that could affect normal thyroid function. The hospital Ethics Committee approved the research and all mothers gave written informed consent prior their inclusion.

Sampling

Blood and urine were sampled from the subject at one of the regular health checks during the first trimester. Blood was taken without anticoagulant and serum was recovered into a polypropylene (PP) tube immediately after sampling. The urine sample was taken by the subject herself in a paper cup and an aliquot was dispensed into several PP tubes. The PP tube was washed with ultrapure water and methanol (for HPLC use, Kanto Chemical Co. Ltd., Tokyo) prior to use. Serum and urine samples were stored at −20 °C. Maternal obstetrical information was obtained from medical records. Dietary and lifestyle information (district of residence, frequency of alcohol drinking and dining out, frequency of seaweed, fish, meat, milk, egg, bean, vegetable and fruits intake) was collected by a self-administered questionnaire.

Thyroid hormone analyses

Thyroid stimulating hormone (TSH) and free thyroxine (fT4) concentrations in serum was determined by electrochemiluminescence immunoassay (ECLIA) with a commercial kit (ECLusys TSH, fT4, Roche, Japan) and thyroxine binding globulin (TBG) concentration was determined by radioimmunoassay (RIA) with a commercial kit (RIA-gnost TBG, CIS Bio International, France) at a commercial laboratory (SRL Co. Inc., Tokyo, Japan). Reproducibility of the measurement of fT4, TSH and TBG in this laboratory was 3.5, 2.0 and 4.4 %, respectively. Reference range for fT4, TSH and TBG in this laboratory is 0.90–1.70 ng/dL, 0.500–5.00 μIU/mL and 12–30 mg/dL, respectively.

PCBs and OH-PCBs analyses

Serum concentrations of OH-PCBs and PCBs were determined according to Takasuga et al. [23]. In brief, a serum sample (1 mL) was extracted in dichloromethane under acidic conditions after the addition of ^{13}C-labelled OH-PCBs and ^{13}C-labelled PCBs as internal standard. The dichloromethane was passed through a silicagel column and PCBs and OH-PCBs fractions were eluted with hexane and acetone, respectively. The acetone fraction was treated with diethyl sulfate and ethylated OH-PCBs were extracted into hexane. The two hexane extracts was individually cleaned up by Florisil column chromatography, which was packed with 22 % H_2SO_4–silicagel and 44 % H_2SO_4–silicagel. Twenty-nine isomers of PCB (IUPAC number: CB-28, 74, 66, 101, 99, 118, 105, 146, 153, 137, 164/163, 138, 167, 156, 178, 182/187,183, 177, 172, 180, 193, 170, 190, 202, 199, 203, 194, 206 and 209) and penta, hexa and hepta OH-PCB isomers [OH-PeCB isomer A (unknown isomer), 4′-OH-CB120, 3-OH-CB118, 4-OH-CB107, OH-PeCB isomers B, OH-HxCB isomer A, 4′-OH-CB165/3-OH-CB153, 4-OH-CB146, 3′-OH-CB138, 4′-OH-CB130, 4′-OH-CB159, OH-HxCB isomers B, OH-HpCB isomer A,

4-OH-CB187, OH-HpCB isomers B, 4′-OH-CB172] were analyzed using a gas chromatograph (HP 6890, Agilent Technologies, CA, USA) coupled with a high resolution mass spectrometer (Micromass Autospec Ultima, Waters, Milford, MA, USA). A capillary column, DB-5MS [60 m × 0.32 mm I.D. (0.25 μm), J&W, Agilent Technologies, CA, USA] was used for separation of OH-PCBs and HT8-PCB (60 m × 0.25 mm I.D., Kanto Chemical Co., Inc., Tokyo, Japan) was for PCBs. The detection limit in serum was 0.6 pg/g wet wt. for OH-PCBs and PCBs. Recovery of internal standards was 83 ± 10 and 70 ± 19 % for PCBs and OH-PCBs, respectively. PCBs concentrations were corrected by serum concentration of total lipid and expressed as ng PCB/g lipid. Total lipid concentration in serum was calculated using the summation method by Akins et al. [24].

Iodine analysis

Iodine concentration in urine was measured using inductively coupled plasma mass spectrometry (ICP-MS) according to a previously published method [25]. The urine sample (200 μL) was diluted with 9.7 mL of 2.5 % tetramethylammonium hydroxide (TMAH) (TAMAPURE-AA TMAH, Tama Chemicals Co., Ltd., Kawasaki, Japan) and 0.1 mL of 100 ng/mL tellurium for internal standardization. The sample was filtered through a syringe filter (pore size 0.45 μm) (Minisart; Sartorius Stedim Biotech, Goettingen, Germany) prior to measurement. The iodine and tellurium ions were monitored at $m/z = 127$ and $m/z = 130$, respectively, by ICP-MS (Agilent 7500ce, Agilent Technologies Japan, Ltd., Tokyo, Japan). The concentration of iodine was corrected by creatinine concentration, which was determined by a commercial kit (LabAssay^{TM} Creatinine, Wako Pure Chemical Industries, Ltd., Osaka, Japan) based on the Jaffe method. The detection limit of iodine was 3 μg/L. Accuracy of iodine analysis was checked by using urine reference material (RM) (Seronorm Urine Blank, SERO AS, Billingstad, Norway). The mean and standard deviation of triplicate measurement was 144 ± 3 μg/L, which was in agreement with the assigned range of the RM (131–147 μg/L).

Statistical analyses

Statistical analyses were performed using SPSS for Windows version 12.0 (SPSS Japan Inc., Tokyo, Japan). Normality of distribution was assessed by the Shapiro–Wilkes test. Serum fT4, TSH, OH-PCBs and PCBs concentrations and urinary iodine concentration was log-transformed for statistical analysis.

The association between the concentrations of thyroid hormones measures and OH-PCBs or PCBs was assessed by a multiple regression analysis. One of the measures of thyroid functions (fT4, TSH or TBG) was used as the dependent variable and the serum concentration of OH-PCBs or PCBs was used as an independent variable. The concentration of individual isomer or sum of the concentrations of congeners ("Σ29 PCB isomers" and "Σ16 OH-PCBs isomers") was included in different models. Maternal age, maternal pre-pregnancy weight before pregnancy, parity and maternal smoking before pregnancy was included as covariates. Urinary iodine concentration was also included as an independent variable. Dietary and lifestyle factors collected by questionnaires were not included in the model because of lack of significant association between dependent variables ($p < 0.1$, one-way analysis of variance or Pearson's correlation analysis).

Results

Study population

Table 1 lists the characteristics of the subjects of this study. Although the number of subjects recruited in this study was 129, part of the information was missing for a few subjects, therefore, the number of subjects is variable depending on the item.

Thyroid hormone status

Table 2 shows concentrations of fT4, TSH and TBG in serum and that of iodine in urine taken in the first trimester of pregnancy. Although the subjects were apparently free of clinical symptoms of thyroid diseases, serum concentration of fT4, TSH and TBG for some subjects was outside the reference range. The geometric mean (GM) of urinary

Table 1 Characteristics of subjects of this study

	n	Mean ± SD
Age (years)	127	33.8 ± 5.0
Pre-pregnancy body weight (kg)	128	52.0 ± 6.6
Height (cm)	128	159.2 ± 4.9
Parity (number of subjects)	128	
0	62	
≥1	66	
Smoking during pregnancy (number of subjects)	127	
No	124	
Yes	3	
Smoked until becoming pregnant (number of subjects)	124	
No	108	
Yes	16	

SD standard deviation

Table 2 Concentrations of fT4, TSH, TBG in serum and I in urine of the present subjects

	n	Mean (SD)	Geometric mean (GSD)	Range
TSH (μIU/mL)	129	1.34 (1.37)	0.837 (3.15)	0.014–10
fT4 (ng/dL)	129	1.22 (0.16)	1.21 (1.14)	0.90–1.79
TBG (μg/mL)	129	33.0 (6.4)	32.4 (1.2)	17.7–44.7
Urinary iodine (μg/g creatinine)	122	920 (2700)	370 (3.0)	81–27000

SD standard deviation, *GSD* geometric standard deviation

Table 3 Serum concentrations of ΣOH-PCBs (pg/g wet wt.) and each isomer of the present subject ($n = 128$)

	Percent detected (%)	GM (GSD)	Median	Range	
				Min	Max
ΣOH-PCBs_16 isomers (Pe-Hp)	–	120 (1.7)	120	37	330
OH-PeCB isomer A	100	10 (1.8)	11	1.9	38
4′-OH-CB120	96	1.6 (1.8)	1.7	<LOD	5.1
3-OH-CB118	98	2.5 (2.1)	2.8	<LOD	11
4-OH-CB107	100	24 (1.7)	26	4.8	95
OH-PeCB isomer B	100	3.1 (1.7)	3.3	0.8	8.4
OH-HxCB isomer A	100	4.3 (1.9)	4.3	0.6	21
4′-OH-CB165 (3-OH-CB153)	100	4.3 (1.9)	4.2	0.7	15
4-OH-CB146	100	19 (1.9)	21	4.2	91
3′-OH-CB138	100	5.6 (1.9)	6.1	0.9	19
4′-OH-CB130	48	0.47 (1.7)	0.3	<LOD	1.9
4′-OH-CB159	88	1.1 (2.0)	1.3	<LOD	4.4
OH-HxCB isomer B	100	3.4 (1.7)	3.4	0.8	14
OH-HpCB isomer A	100	4.7 (1.7)	4.6	1.1	18
4-OH-CB187	100	29 (1.7)	31	8.7	97
OH-HpCB isomer B	90	1.3 (2.1)	1.4	<LOD	5.9
4′-OH-CB172	100	3.8 (1.8)	3.8	0.9	16

LOD limit of detection, *GM* geometric mean, *GSD* geometric standard deviation

GM and median values were calculated by substituting one-half of LOD for undetectable samples. One subject with low recovery of internal standard without known reason was excluded from this table

iodine concentration was 370 μg/g creatinine with a range 81–27000 μg/g creatinine.

Serum concentrations of OH-PCBs and PCBs

Table 3 shows the concentrations of ΣOH-PCBs (16 isomers) and individual isomers of OH-PCB. The GM of ΣOH-PCBs was 120 pg/g wet wt. (range 37–330 pg/g wet wt.). ΣOH-PCBs concentration increased with maternal age ($r = 0.341$, $p < 0.001$), while it decreased with maternal pre-pregnancy weight ($r = -0.230$, $p = 0.009$). Mean serum ΣOH-PCBs concentration was higher in primiparous [GM (GSD): 160 (1.5) pg/g wet wt.] than multiparous subjects [100 (1.6) pg/g wet wt.] (*t* test, $p < 0.001$) (Table 4). This significant difference was also found for the concentrations of individual isomers (data not shown). It has to be noted that there was no difference in mean age between primiparous and multiparous subjects (*t* test, $p = 0.368$). Figure 1 shows the relationship between maternal age and ΣOH-PCBs concentration divided by parity.

Table 5 shows the concentrations of serum ΣPCBs (29 isomers) and that of each isomer. The GM of ΣPCBs was 68 ng/g lipid wt. (range 20–210 ng/g lipid wt.). ΣPCBs concentration increased with maternal age ($r = 0.399$, $p < 0.001$), but it was not correlated with maternal pre-pregnancy weight. Mean serum PCBs concentrations were higher in the primiparous [GM (GSD): 87 (1.4) ng/g lipid wt.] than in multiparous subjects [55 (1.6) ng/g lipid wt.] (*t* test, $p < 0.001$) and this difference was also significant when the individual isomer was tested except for two (CB-206, CB-209) (Table 4).

The positive correlation between the concentrations of OH-PCB isomer and the parent PCB isomer was significant

Table 4 Comparison of serum concentration of OH-PCBs, PCBs and age between primiparous and multiparous subjects

	Primiparous (*n*)		Multiparous (*n*)		*p**
Age (years)	61	33.4 (5.6)	66	34.2 (4.4)	0.368
ΣOH-PCBs_16 isomers (pg/g wet wt.)	61	160 (1.5)	66	100 (1.6)	<0.001
ΣPCBs_29 isomers (ng/g lipid wt.)	59	87 (1.4)	66	55 (1.6)	<0.001

Arithmetic mean and arithmetic SD for age, and geometric mean and GSD in the parenthesis for OH-PCBs and PCBs concentration

* *t* test. Log-transformed concentration of PCB and OH-PCB was used for comparison

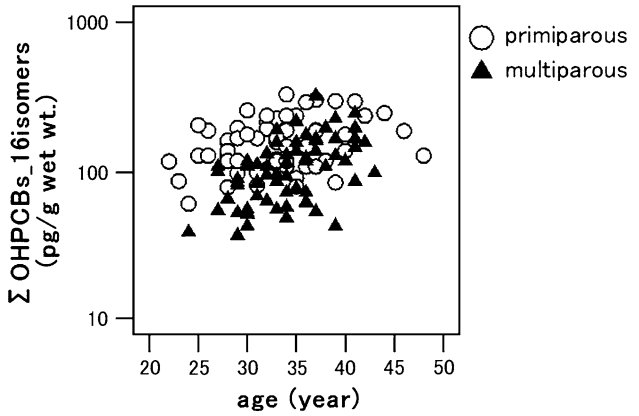

Fig. 1 Scatter plot of subject's age and serum OH-PCBs concentration (sum of 16 isomers). Correlation between age and OH-PCBs for primiparous subjects was 0.343 ($p = 0.007$) and that for multiparous subjects was 0.517 ($p < 0.001$)

for all of the pairs (Table 6). Pairing of OH-PCB isomer and corresponding parent PCB isomer was determined according to Hovander et al. [26].

Association between OH-PCBs and PCBs and thyroid functions

The multiple regression analyses demonstrated that none of the independent variables included in the model (serum concentration of OH-PCBs or PCBs with other covariates including urinary iodine concentration) significantly explain the variation of thyroid hormone status measures, i.e., serum concentration of fT4, TSH and TBG. In Table 7 is shown one such example.

Discussion

Serum concentration of OH-PCBs and PCBs and their variation

The serum or plasma concentrations of OH-PCBs in the Japanese have been reported in a limited number of publications [17, 27–29]. Enomoto et al. [27] reported concentrations of six isomers of OH-PCBs in the whole blood of Japanese pregnant women (e.g., 4-OH-CB146, median 11 pg/g wet wt.). Median 4-OH-CB146 concentration in the present study (21 pg/g wet wt.) was in agreement with that of Enomoto et al. [27] (whole blood) if 40 % hematocrit was assumed. On the other hand, the concentration was lower than that in serum of Japanese women diagnosed with breast cancer [17]. The reason for this difference may be due to the higher age of subjects (51.1 ± 9.8 years) in that study than that in the present study (33.8 ± 5.0 years).

The serum concentrations of OH-PCBs in this study were generally lower than those reported in other countries [15, 30–38]. For example, median concentration of 4-OH-CB146 in the serum of Faroe Islanders (790 pg/g wet wt.) [31] was around 40 times that of this study. Elevated plasma concentrations of OH-PCBs were also reported for Inuits [15]. The higher serum OH-PCBs concentration of Inuit and Faroe Islanders should be due to their heavy dependence on marine resources, such as sea mammals and fatty fish, which could be the source of both OH-PCBs and parent PCBs.

The concentrations of PCBs in serum were similar to those of Japanese females reported so far [40–44]. The concentrations of major PCB isomers were lower than those of studies of people who were expected to be heavily exposed for PCBs (e.g., fisherman and habitual sea mammal eaters) [15, 16, 45, 46].

Japanese people are a fish eating population, however, the serum OH-PCBs concentration was lower than those reported in the studies in Netherlands [33] or Slovakia [34] and also PCBs concentration in this study was lower than that in populations who seemingly were not heavily dependent on marine resources [47, 48]. The reason is not clear, but exposure to PCBs/OH-PCBs via the sources other than food may have to be considered in other countries [39].

A positive correlation between age and serum OH-PCBs/PCBs concentrations, as observed in the present subjects (Fig. 1), has already been reported in previous studies [17, 49, 50], which has been attributed to the long biological half-life of PCBs. The positive correlation found between serum concentrations of OH-PCBs and maternal age may be due to the result of metabolism of increasing body burden of parent PCBs with age. It is also well

Table 5 Serum concentrations of ΣPCBs and each isomer (ng/g lipid wt.) of the present subject ($n = 126$)

	Percent detected (%)	Geometric mean (GSD)	Median	Range	
				Min	Max
ΣPCBs_29 isomers	–	68 (1.6)	69	20	210
CB-28	100	0.68 (1.6)	0.63	0.24	6.7
CB-74	100	1.8 (1.8)	1.8	0.26	7.5
CB-66	100	0.66 (1.7)	0.66	0.17	4.0
CB-101	100	0.48 (1.6)	0.50	0.14	2.0
CB-99	100	2.4 (1.7)	2.6	0.62	6.5
CB-118	100	3.9 (1.8)	3.9	0.74	13
CB-105	100	0.80 (1.7)	0.84	0.17	2.4
CB-146	100	2.0 (1.6)	2.1	0.63	5.8
CB-153	100	16 (1.7)	16	4.2	54
CB-137	98	0.40 (1.7)	0.42	<LOD	1.3
CB-164/163	100	3.2 (1.7)	3.4	0.90	9.8
CB-138	100	8.2 (1.7)	8.9	2.2	28
CB-167	99	0.56 (1.8)	0.54	<LOD	2.0
CB-156	100	1.3 (1.7)	1.3	0.31	4.6
CB-178	100	0.84 (1.6)	0.88	0.26	2.8
CB-182/187	100	4.6 (1.6)	4.8	1.6	13
CB-183	100	1.1 (1.6)	1.1	0.38	3.4
CB-177	100	0.86 (1.6)	0.86	0.28	2.2
CB-172	100	0.46 (1.6)	0.46	0.14	1.5
CB-180	100	8.6 (1.6)	8.6	2.7	29
CB-193	100	0.70 (1.7)	0.72	0.19	2.2
CB-170	100	3.0 (1.6)	3.0	0.91	9.3
CB-190	100	0.61 (1.6)	0.61	0.19	1.8
OB-202	100	0.45 (1.6)	0.44	0.13	1.7
OB-199	100	1.1 (1.6)	1.1	0.34	3.6
OB-203	100	0.70 (1.6)	0.71	0.21	2.2
OB-194	100	1.2 (1.6)	1.2	0.34	4.4
CB-206	99	0.40 (1.5)	0.39	<LOD	1.2
CB-209	98	0.33 (1.6)	0.32	<LOD	1.2

LOD limit of detection, *GM* geometric mean, *GSD* geometric standard deviation

GM and median values were calculated by substituting one-half of LOD for undetectable samples. Three subjects with low recovery of internal standard were excluded from this table

Table 6 Correlation between the concentrations of OH-PCB isomer and the parent PCB isomer

	Parent compound	r*	p
4-OH-CB107	CB-118 + CB-105	0.632	<0.001
4-OH-CB146	CB-153 + CB-138	0.735	<0.001
4′-OH-CB172	CB-170 + CB-180	0.711	<0.001
3′-OH-CB138	CB-138	0.607	<0.001
ΣOH-PCBs_16isomers	ΣPCBs_29isomers	0.755	<0.001

* Pearson correlation coefficient. Log-transformed PCB and OH-PCB concentration was used in all analyses

documented that the body burden of PCBs of woman decreased after pregnancy and lactation [49, 51], which was also the case with the present subjects (Table 4). This has generally been attributed to "excretion" of body burden via the lactation [52]. The significant difference in serum concentration of OH-PCBs between primiparous and multiparous subjects (Table 4) may also be due to a similar trend of parent PCBs. It must be noted that age dose not play a significant role in this difference because there was no age difference between primiparous and multiparous subjects (Table 4).

Since the biological half-time of OH-PCBs is short (3.8 days for 4-OH-CB107 and 15 days for 4-OH-CB187 in rats) [53], the observed age-dependent elevation, as well

Table 7 An example of the result of multiple regression analysis

Dependent variable	fT4		
	Standardized β	95 % CI	p
ΣOH-PCBs_16 isomers	0.059	−0.046, 0.078	0.613
Parity	−0.104	−0.085, 0.030	0.346
Urinary iodine	−0.082	−0.033, 0.014	0.406
Age	−0.051	−0.007, 0.004	0.406
Smoking before pregnancy	−0.019	−0.082, 0.067	0.841
Maternal weight before pregnancy	−0.004	−0.210, 0.202	0.968
Coefficient of determination R^2	0.030		

Models adjusted for maternal age, maternal weight before pregnancy, urinary I, smoking before pregnancy and parity

OH-PCBs, PCBs, fT4, TSH, urinary iodine and maternal weight were log-transformed for analysis

as a significant decrease after pregnancy and lactation, in serum concentration suggested that a major portion of circulating OH-PCBs is derived from metabolism of the parent PCB body burden rather than OH-PCBs exposure.

Association between serum concentrations of thyroid measures and OH-PCBs

The association between serum or plasma concentrations of OH-PCBs and thyroid hormone in adults has been reported in some previous studies [12, 15–17]. In none of these studies, significant association between the concentrations of OH-PCBs and fT4 in serum/plasma has been found and this was consistent with the present result. Dallaire et al. [15] reported a negative association between the concentrations of total T3 and 7 OH-PCB isomers in Inuit adults whereas they found a positive association between the sum of the concentrations of 11 OH-PCBs and T3 in another Inuit population composed of pregnant women [16]. Since T3 was not measured in this study, we do not have any idea if the serum T3 level had association with OH-PCBs in our subjects. It must be noted that in the two Dallaire et al., studies, the direction of association was inverse without known reasons. It must also be noted that the association between TBG and OH-PCBs concentration was not consistent within the two Dallaire et al., studies.

The results of several previous in vitro studies have indicated that OH-PCBs have potency for thyroid effect. An isomer of OH-PCB with hydroxyl group in meta or para position in its structure (e.g., 3-OH-CB118), was demonstrated to be a potent inhibitor of enzymatic sulfation of thyroid hormone in an in vitro assay [11]. Other in vitro and in vivo studies demonstrated that OH-PCBs competitively bound to transthyretin (TTR) over thyroid hormone [10, 54, 55]. The present study, as well as some of the previous studies [12, 17], however, indicates that human exposure to OH-PCBs at environmental levels may not affect thyroid hormone levels in blood. It must be pointed out, however, that even if OH-PCBs affect circulating thyroid hormone by interfering binding of thyroid hormone to TTR, it may not affect the level of circulating thyroid hormone because of a minor abundance of TTR as thyroid hormone carrier protein in the blood. Therefore, although we did not see any effect on serum measures of thyroid hormones, it does not necessarily mean no effect of OH-PCBs for thyroid hormone actions.

Association of serum concentrations of thyroid measures and PCBs

It has been proposed that PCBs affect circulating thyroid hormones by (1) decreasing synthesis and secretion of T4 from the thyroid gland, (2) reducing biological half-time of T4 via the activation of UDP-glucuronosyltransferase, (3) competitively binding to carrier protein based on the findings of in vitro and in vivo studies [7, 8]. It was therefore expected that serum concentration of fT4 negatively, while that of TSH positively, correlate with the concentration of PCBs. However, we did not find any statistical associations between serum concentrations of PCBs and fT4 or TSH as in the case with OH-PCBs. Many human studies examined the association of serum measures of thyroid hormone status and PCBs and found inconsistent results. However, the majority of the previous studies found no association between the two [9] as has been found in the present study. The inconsistency across studies has generally been attributed to differences in exposure/body burden levels, measured isomers, or dietary habit and lifestyle of subject populations, across the studies. As mentioned earlier, even if serum levels of thyroid hormone do not change with PCBs exposure, it does not necessarily means that PCBs do not have any effect on thyroid actions.

Iodine as a covariate of thyroid functions

In this study we examined urinary iodine concentration as an independent variable in the multiple regression analysis. Iodine deficiency during pregnancy is associated with reduced thyroid hormone levels [56]. Meanwhile, excessive iodine intake also affects thyroid function [22, 57]. Japanese people are generally considered iodine adequate because of abundant seaweed and fish consumption: therefore, we included iodine status to see if higher (even excessive) iodine nutrition itself affects thyroid functions or it modifies the possible effect of OH-PCBs/PCBs on thyroid measures. As expected, urinary iodine levels indicated that our subjects are iodine adequate or even high

(Table 2), however, it did not contribute to variation in thyroid hormone measures. As pointed out by Dallaire et al. [16, 19], co-exposure to OH-PCBs/PCBs and iodine takes place via the marine products consumption in general populations, therefore, OH-PCBs and PCBs effects on thyroid functions have supposedly been investigated in iodine adequate populations as in the case with the present study. In must be noted that, except for a few studies [20, 21], iodine status has not been examined in the publications on PCBs and thyroid function. Effects of OH-PCBs/PCBs exposure on thyroid functions under iodine less than adequate conditions may be of importance because some of the suggested mechanisms of effect of PCBs on thyroid function involved enhanced uptake of iodine into the thyroid [58, 59].

Conclusion

In summary, we did not find associations between OH-PCBs/PCBs exposure and thyroid hormone measures in the serum of pregnant women at first trimester of gestation in this study. This result was consistent with that of the majority of the previous studies, though not all, indicating that OH-PCBs/PCBs exposure at environmental levels does not have measurable effect on circulating thyroid hormones. However, it does not exclude the potential effect of OH-PCBs/PCBs on thyroid hormone actions in target tissues and organs without change of circulation thyroid hormone levels. It is particularly of concern for fetal exposure because the fetus is generally more vulnerable to thyroid hormone disruption than is the adult.

Acknowledgments We sincerely appreciate the subjects who participated in this study and to the technical staff and clinical staff of Showa University Hospital.

Conflict of interest The authors declare that they have no conflict of interest.

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