**Association of *In Utero* Persistent Organic Pollutant Exposure with Placental Thyroid Hormones**

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**Keywords**

Polybrominated diphenyl ether; [Polychlorinated biphenyl](http://www.baidu.com/link?url=TSyFXVZuWRqT8eJR3vjUJLdKKa8eKLf4DPd2MLCcKyPKZJNKAfrxEkPnQ89bcUmsBNSkwNqOloVxLV0jpFGXywXDsxfWP-AKZwxSKOkFGRwF1ypoLXtWAElP_ZxIE_xF); Polychlorinated dibenzo-*p*-dioxin/furan; Organotin chemical; Organochlorine pesticide; Thyroid hormone; Placenta

**Abstract**

***Background***: *In utero* exposure to persistent organic pollutants (POPs) can result in thyroid function disorder, leading to concerns about their impact on fetal and neonatal development. The present study was performed to investigate the associations between placental levels of various POPs and thyroid hormones (THs).

***Methods***: In a prospective Danish study initially established for assessing congenital cryptorchidism, 58 placenta samples were collected from mothers of boys born with (28) and without (30) cryptorchidism. The concentrations of polybrominated diphenyl ethers (PBDEs), [polychlorinated biphenyl](http://www.baidu.com/link?url=TSyFXVZuWRqT8eJR3vjUJLdKKa8eKLf4DPd2MLCcKyPKZJNKAfrxEkPnQ89bcUmsBNSkwNqOloVxLV0jpFGXywXDsxfWP-AKZwxSKOkFGRwF1ypoLXtWAElP_ZxIE_xF)s (PCBs), polychlorinated dibenzo-*p*-dioxins/furans (PCDD/Fs), organotin chemicals (OTCs), organochlorine pesticides (OCPs), thyroxine (T4), 3,3’,5-triiodothyronine (T3), and 3,3’,5’-triiodothyronine (rT3) were measured. The associations between placental THs and various POPs were analyzed using multiple linear regression.

***Results***: Five PBDEs, 35 PCBs, 14 PCDD/Fs, 3 OTCs, 25 OCPs, T4, T3, and rT3 were measured in placenta samples. No correlation between THs and the odds of cryptorchidism was found. Several POPs were significantly associated with THs: a) T4 was inversely associated with BDEs 99, 100, ΣPBDE, and 2378-TeCDD, and positively associated with 1234678-HpCDF; b) T3 was positively associated with 2378-TeCDF and 12378-PeCDF; c) rT3 was positively associated with PCB 81, 12378-PeCDF and 234678-HxCDF, and inversely associated with tributyltin (TBT), ΣOTC, and methoxychlor (MOC).

***Conclusions***: Background exposures to several POPs were associated with modifications of the TH homeostasis in placenta, a possible mechanism for the impacts of POP exposures on children’s growth and development. This study provides new insight into the complexity of thyroid-disrupting properties of POPs. More research is needed to elucidate the effects of POPs on the transplacental passage of THs, as well as the biological consequences of exposure.

**Introduction**

Thyroid hormones (THs) are a group of tyrosine based hormones that act on cells of almost all tissues and therefore are involved in important physiological processes during a life span (1, 2). The human thyroid system is susceptible to disruption by endogenous (e.g., autoantibodies) or exogenous (e.g., iodine) factors, by interfering with the sodium-iodide symporter, TH metabolism, receptors, and TH transport (3). Consequently, there are concerns regarding the chronic background exposure to persistent organic pollutants (POPs), which have been shown or are suspected to have thyroid-disrupting properties.

POPs have been used in a wide variety of commercial and industrial applications over the past few decades. They are also released from traffic and incineration. Common POPs include polybrominated diphenyl ethers (PBDEs), polychlorinated biphenyls (PCBs), polychlorinated dibenzo-*p*-dioxins and furans (PCDD/Fs), organotin chemicals (OTCs), and organochlorine pesticides (OCPs), among others. Contamination of food is suspected to be the major exposure pathway to these chemicals for humans (4). Although the international agreements intended to reduce or cease their production, POPs are still widely detectable in the biotic and abiotic environment due to their high lipophilicity and resistance to degradation (5). Recently, POPs have been detected in human samples such as blood and breast milk from all over the world (6).

POPs could be transferred to the fetus across the placenta (7, 8), which was traditionally considered to be a barrier and protect the fetus against toxicants circulating in maternal blood. Passive diffusion and/or active uptake are involved in this process. The placenta can also act as a repository for these lipophilic chemicals. Thus, there is a risk for disruption of the fetal development because POPs in placenta may affect the amount of THs delivered to the fetus. This is particularly important during early pregnancy when the fetus depends solely on maternally-derived THs (9). Even subtle changes in maternal TH transfer can lead to various detrimental outcomes such as miscarriage, intra-uterine growth retardation, hypertensive disorders, preterm delivery, and a decreased child IQ (9, 10).

Disruption of TH homeostasis following POP exposure has been observed *in vitro* and in animal studies (11, 12). Human epidemiological studies have also found associations between POP exposures and TH disruption in the general population (13, 14), i.e. in e-waste recycling workers (15, 16), pregnant women (17-20) and infants (20). Recent studies suggested that prenatal exposure to POPs could interfere with the neurological development of the children (21), as well as their motor, cognitive and behavioral performance (22, 23). However, all these studies were conducted with serum, plasma, or breast milk samples. Only one study was performed in placenta on the associations of THs with PBDEs (24), while no research is available for PCBs, PCDD/Fs, OTCs, and OCPs. It is therefore worthwhile to conduct a comprehensive analysis by including as many POPs as possible to have a complete overview of the influence of *in utero* POP exposures on THs.

The potential thyroid-disrupting effects of POPs in background exposed populations are of interest because THs act at extremely low serum concentrations, while POPs can mimic or inhibit the response of the hormones at low doses (25, 26). The purpose of this study was to investigate the possible effect of background exposures of POPs (PBDEs, PCBs, PCDD/Fs, OTCs, and OCPs) on placental levels of THs (thyroxine (T4), 3,3’,5-triiodothyronine (T3), and 3,3’,5’-triiodothyronine (rT3)) in a Danish population that gave birth to boys with and without cryptorchidism.

**Materials and Methods**

*Study Population*

The placenta samples were obtained from a joint prospective, longitudinal birth cohort study performed from 1997 to 2001 at the National University Hospital (Rigshospitalet, Hvidovre Hospital), Copenhagen, Denmark. The standardized recruitment strategy, inclusion criteria, participation rate and clinical examination techniques have been reported earlier with details (27, 28). Placentas were collected at birth by the midwives and kept frozen in polyethylene bags at -20 °C. Upon analysis, placentas were defrosted, mechanically homogenized and aliquoted into 20-mL glass tubes. Fifty-eight placenta samples were derived from mothers of healthy boys (n = 30) and mothers of boys with cryptorchidism (n = 28) at birth in a nested case-control design. The number of included placenta samples was determined by funding. The demographical variables of both groups are shown in **Table S1** (**Supplementary Material**).

This study followed the Helsinki II declaration (World Medical Association 2004). The Danish ethics committee (KF01-030/97) and the Danish Data Protection Agency (1997-1200-074) approved the study. Informed written consent was obtained from the parents of each boy.

*Data Collection*

We determined the total concentrations of T4, T3 and rT3 in placenta samples. Detailed information about the hormones, sample preparation, extraction procedure, analytical method, reagents, and instrumentation have been described earlier (29). Briefly, a 100-mg placenta sample was extracted using solid/liquid extraction, liquid/liquid extraction, and weak cation-exchangers (Bond Elut Plexa PCX) SPE cleanup. Finally, the compounds were quantified by liquid chromatography quadrupole time-of-flight mass spectrometry (LC-Q-TOF-MS). The method quantification limits (MQL) of T4, T3, and rT3 were 0.7, 0.3 and 0.2 ng g-1, respectively. The procedural recoveries for the hormones were 81.0 – 105%, with a coefficient of variation (CV) of 0.5 – 6.2%. The intra-day CVs and inter-day CVs were 0.5 – 4.19% and 1.19 – 6.38%, respectively (29).

PBDEs, PCBs, PCDD/Fs, OTCs and OCPs were targeted for analysis in this study. The methods for the sample cleanup and quantification of the compounds have been described in detail previously (28, 30-32). A brief description of the methods is given in the **Supplementary Material**. In addition, we included the World Health Organization toxic equivalent (WHO-TEQ) values of PCDD/Fs and PCBs. This parameter was developed to measure the combined toxicity of dioxin and dioxin-like compounds through the toxic equivalency factors (TEFs), which assesses the toxicity of each congener relative to 2378-TeCDD (33).

*Statistics*

We determined the potential effects of POPs on thyroid status for congeners with a detection frequency > 50%, except for dibutyltin (DBT), tributyltin (TBT), and triphenyltin (TPhT), which were detected in 38%, 41% and 34% of the samples, respectively. We included OTCs regardless of detection frequency because of the growing concerns about environmental effects as well as a lack of studies on the TH effects of these chemicals. Concentrations below LOQs were replaced by the LOQ divided by the square root of 2. POP concentrations were heavily right-skewed and were log10-transformed to reduce the influence of outliers. T4, T3 and rT3 were normally distributed and not transformed. Normality was confirmed using Kolmogorov-Smirnov test. We used Spearman’s rank correlations to evaluate the interrelationship of POP congeners and analysis of variance (ANOVA) to examine associations between demographic characteristics and POP concentrations. Adjusted and unadjusted logistic regressions were used to compare placental TH concentrations in cases with cryptorchidism and controls. As no differences were observed, cases and controls were pooled for analysis of the association between POPs and THs by multiple linear regression for individual POP congeners and the sums.

Potential confounders considered for inclusion in the models were parameters known to influence THs, i.e., maternal age, gestational age, parity (nulliparous vs. one or more live birth), maternal pre-pregnant body mass index (BMI), smoking during pregnancy (yes vs. no), mode of delivery (three categories: vaginal delivery, vacuum extraction, and cesarean section), birth weight, and birth length. Final models included variables that were loosely associated with the THs (*p* < 0.20) in bivariate analyses.

All statistical analyses were conducted using SAS (version 9.4; SAS Institute Inc., Cary, NC) and R (version 3.4.2; R Foundation for Statistical Computing, Vienna, Austria). A *p*-value < 0.05 was considered significant, and *p* < 0.10 was considered as a tendency of association.

**Results**

**Table 1** summarizes the characteristics of all the participating women and newborns. The mean (±SD) age of women was 30.4 ± 3.5 years. Among them, 19 (32.8%) were > 30 years of age; forty-seven (81%) had a BMI value of < 25; thirty-nine (67.2%) did not smoke; forty-nine (84.5%) had a normal delivery; thirty-five (60.3%) were nullipara; fifty-four (93.1%) babies were delivered at over 37 weeks of gestation (full term). The demographical parameters did not differ significantly between the cryptorchid and control groups except for gestational age (*p* = 0.06) (**Table S1, Supplementary Material**).

As shown in **Table 1**, T4 was measured in the highest concentration with a mean of 37.6 ng g-1 fresh weight (fw) (range: 16.3 – 68.3 ng g-1 fw), followed by rT3 with a mean of 4.57 ng g-1 fw (range: 1.82 – 9.03 ng g-1 fw). T3 was measured in the lowest concentration with a mean of 0.85 ng g-1 fw (range: 0 – 2.34 ng g-1 fw). TH levels were generally similar across age, BMI, parity, mode of delivery, and birth length of the infants. T4 was correlated with gestational age (*p* = 0.01), birth weight (*p* = 0.06), and smoking (*p* = 0.03). However, T3 and rT3 were not correlated with any of the studied characteristics. Placental concentrations of THs did not significantly differ between cryptorchid and control boys (**Table 2).**

The concentrations of a total of 82 POPs and their sums, as well as the WHO-TEQ values of PCDD/Fs and PCBs in placenta are shown in **Table S2 (Supplementary Material).** The limit of quantification (LOQ) values were 0.004 – 0.14 ng g-1 lipid for PBDEs (30), 0.17 pg g-1 lipid for non-*ortho*-PCBs, 0.01 ng g-1 lipid for mono- and di-*ortho*-PCBs, 0.12 – 0.25 pg g-1 lipid for tetra to hepta chlorinated PCDD/Fs, 1.2 pg g-1 lipid for octa chlorinated PCDD/Fs (34), 0.1 ng g-1 fw for DBT, 0.02 ng g-1 fw for TBT and TPhT (31). The limit of detection (LOD) values of the OCPs ranged from 0.01 ng g-1 lipid to 1.24 ng g-1 lipid (28).

As shown in **Table 3** and **Table S3** **(Supplementary Material).** T4 was inversely significantly associated with the sum of 5 PBDEs (β = -19.0; 95% CI: -35.7, -2.37; *p* < 0.026) as well as with BDE 99 (β = -20.2; 95% CI: -35.2, -5.29; *p* < 0.009), BDE 100 (β = -13.5; 95% CI: -26.8, -0.22; *p* < 0.047) with a tendency for BDE 47 (β = -14.1; 95% CI: -28.5, 0.24; *p* < 0.054). T3 and rT3 were not significantly associated with any of the PBDE congeners.

T4 showed negative tendencies with PCB 99 (β = -15.9; 95% CI: -33.4, 1.65; *p* < 0.075), PCB 118 (β = -13.4; 95% CI: -29.0, 2.27; *p* < 0.092) and PCB 167 (β = -11.7; 95% CI: -25.7, 2.29; *p* < 0.099). No statistically significant associations between T3 and PCB congeners was observed. rT3 was positively significantly associated with PCB 81 (β = 2.55; 95% CI: 0.37, 4.72; *p* < 0.023) with a positive tendency for PCB 101 (β = 0.80; 95% CI: -0.06, 1.65; *p* < 0.067) and PCB 183 (β = 2.08; 95% CI: -0.35, 4.51; *p* < 0.091).

T4 was inversely significantly associated with 2378-TeCDD (β = -18.4; 95% CI: -34.6, -2.25; *p* < 0.026) with a tendency for 12378-PeCDD (β = -13.4; 95% CI: -28.8, 2.01; *p* < 0.087), while positively and significantly associated with 1234678-HpCDF (β = 14.1; 95% CI: 4.37, 22.8; *p* < 0.005) with a positive tendency for OCDD (β = 10.8; 95% CI: -1.94, 23.5; *p* < 0.095). T3 was positively significantly associated with 2378-TeCDF (β = 1.08; 95% CI: 0.33, 1.83; *p* < 0.006) and 12378-PeCDF (β = 0.73; 95% CI: 0.07, 1.40; *p* < 0.032). rT3 was positively significantly associated with 12378-PeCDF (β = 2.53; 95% CI: 0.41, 4.64; *p* < 0.020) and 234678-HxCDF (β = 3.11; 95% CI: 0.94, 5.27; *p* < 0.006), and although not statistically significant, showed a positive tendency with 123478-HxCDF (β = 2.68; 95% CI: -0.47, 5.82; *p* < 0.093), 123678-HxCDF (β = 2.66; 95% CI: -0.41, 5.73; *p* < 0.088) and 1234678-HpCDF (β = 1.43; 95% CI: -0.24, 3.10; *p* < 0.092).

rT3 was inversely significantly associated with TBT (β = -2.65; 95% CI: -3.79, -1.51; *p* < 0.001) and the sum of OTCs (β = -4.96; 95% CI: -7.78, -2.14; *p* < 0.001), while none of the OTCs showed a significant association with T4 or T3.

β-hexachlorocyclohexane (β-HCH) showed negative tendencies with T4 (β = -13.9; 95% CI: -28.7, 0.90; *p* < 0.065), T3 (β = -0.68; 95% CI: -1.44, 0.09; *p* < 0.081) and rT3 (β = -2.06; 95% CI: -4.50, 0.38; *p* < 0.097), although none of them were statistically significant. Methoxychlor (MOC) was inversely significantly associated with rT3 (β = -2.30; 95% CI: -3.88, -0.72; *p* < 0.005).

**Discussion**

The primary goal of this study was to determine placental concentrations of THs and to examine their associations with various POPs. Background exposures of POPs could interfere with the thyroid homeostasis in placenta, and thereby influence the THs delivered to the fetus, which are critical for both the fetal and neonatal development. Although the effect of each chemical seems scarce, the added effects may cause inappropriate consequences.

We measured T4, T3 and rT3 by an UPLC-Q-TOF-MS method, which offers better sensitivity and specificity than radio immunoassay (RIA)-based methods, as explained previously (35). The TH concentrations measured in our study corresponded well with previous findings (24, 36).

*In utero* exposure to POPs occurs during gestation as a result of placental transfer. Thus, placental concentrations of some contaminants may be representative of fetal exposures (37). The exact mechanisms involved in the transplacental transfer of POPs are not fully understood. Binding of POPs to transthyretin (TTR) and TH membrane transporters may affect this process. Our results revealed a widespread background contamination of POPs in human placenta. BDE 47 and BDE 153 are the major components compared with other BDE congeners, which is in accordance with a previous study (38). ΣPBDE concentration was lower than those measured in placental samples from South China (mean: 13.3 ng g-1 lipid; range: 4.32 – 42.0 ng g-1 lipid) (8) and the USA (mean: 14.6 ng g-1 lipid; range: 0.62 – 521.8 ng g-1 lipid) (24) due to the lower human exposure of PBDEs in Europe (8). Wang *et* al. reported lower PCB concentrations than our results, and PCB 118, 156 and PCB 105 had the highest levels (39). This difference may be due to the region-specific pollution, as well as the sampling time because the human exposure of PCBs continues to decrease since 1970s. The ΣPCDD/F concentrations were in the same range as in a previous study in placenta from cesarean section (40). The placental levels of OTCs were in the same range with our previous study (31).

We observed positive correlations between T4 and gestational age, maternal smoking, and birth weight, which were in accordance with previous studies (41-44). Therefore, these characteristics were included in the statistical analysis as cofounders. However, some other researchers have also found associations of THs with mode of delivery (41), alcohol consumption (45), and maternal BMI (46).

We investigated the possible relationship between THs and cryptorchidism because a previous review suggested that THs may play a role in the development and function of the testis (47). Multiple logistic regression revealed no difference of THs between the two groups, which could be due to the small number of cases. Bruker-Davis *et* al. also reported no difference of THs in cord blood from mothers that gave birth to boys with and without cryptorchidism (48). However, the results for analyzing the effect of THs may be very different when a different matrix (i.e., maternal serum, cord blood, infant serum or placenta) and different TH measurements (i.e., total concentrations or free concentrations) were used.

This study revealed that POP exposures in general result in lower levels of total T4 or increased levels of total T3 and total rT3. Previous studies have reported positive (18-20), negative (18, 20), and no associations (49) between certain POPs and TH concentrations in pregnant women and newborns. However, our results may not be directly comparable to these studies because the biological matrix for exposure measurements and TH determination is not the same. Only one study exists on THs and brominated flame retardants in placenta, in which a sex-specific manner of association was observed: placental BDE-99 and BDE-209 were negatively associated with rT3 levels in male infants, while BDE-99 and 2,4,6-tribromophenol (2,4,6-TBP) were positively associated with T3 concentrations in female infants (24).

The mechanisms involved in the thyroid-disrupting process of POPs are diverse and complex: (i) POPs may disrupt the activity of the thyroid gland by interference with the thyroid-stimulating hormone (TSH)-receptor, sodium iodide symporter (NIS), thyroid peroxidase (TPO), as well as other receptors on the thyrocyte; (ii) POPs may competitively bind to TH binding proteins, i.e., thyroid binding globulin (TBG), transthyretin (TTR) and thyroid hormone receptors (TR); (iii) POPs may affect the peripheral TH metabolism and clearance by activation or inactivation of the enzymes, i.e., iodothyronine deiodinases and UDP-glucuronosyltransferases (UDPGTs) (50). In blood, certain POPs and their hydroxylated metabolites may competitively bind with TH-binding proteins and deiodinases, leading to an increase in the free T4 concentration. The feedback regulation via TSH may compensate for this change, resulting in a stable concentration of T4 in serum (50). Total T4 in placenta mainly originates from the transplacental passage of maternal free T4, while total T3 and total rT3 were derived entirely from placental and foetal metabolism of T4 (deiodinases D2 and D3) (51). Disrupting the TH-protein (i.e., TTR and TH membrane transporters) binding and metabolism in placenta may be of significance. TTR plays a crucial role in transferring free T4 across the placenta to the fetal compartment (50). Binding of certain POPs to TTR may facilitate the transport of these compounds, while reducing T4 delivery to the fetus. Different compounds exhibit different affinities to these proteins. PBDEs, PCBs, and especially their hydroxylated metabolites have a high degree of structural resemblance of T4 and are therefore able to competitively bind with TTR.

*Associations of Concentrations of T4 with POPs*

Our results revealed inverse associations of BDE 47, 99, 100 and ΣPBDE with T4, which were in accordance with a previous report (24). This could be due to the interference of these chemicals with the TH transport system because strong affinities between TTR and BDE 47, 99, and BDE 100 have been observed previously (52). PCBs were expected to show associations with T4 levels because they have a high degree of structural resemblance to T4. Boas *et* al. also suggested that perinatal exposure to PCBs decrease THs (50). However, only negative tendencies between T4 and PCB 99, 118 and PCB 167 were found in this study. This might be due to the differences in the applied matrix, as well as the differences in biological indicators of exposure between different studies. For example, Majidi *et* al. analyzed the TH effect of PCBs using standardized concentrations expressed in total PCB equivalent per kg of lipids in maternal plasma (μg PCBMPEQ kg-1 lipids). The results suggested little evidence for the impact of PCBs on thyroid function in pregnant women and newborns (53). In addition to TTR, PCDD/Fs are able to bind to and activate the aryl hydrocarbon receptor (AhR), inducing UDPGT as well as cytochrome P450 enzymes, which stimulate the glucuronidation of T4 and biliary excretion of this conjugate, resulting in lower levels of T4 (54). This may explain the inverse association of 2378-TeCDD with T4 observed here. However, we also observed positive associations between T4 and 12378-PeCDD, 1234678-HpCDF, and OCDD here, probably due to the inhibitory effect of dioxins on the activity of placental deiodinases. Human and animal studies revealed negative associations between T4 levels and pesticides such as dichlorodiphenyltrichloroethane (DDT) (and its metabolite DDE) and hexachlorobenzene (HCB). Interference with the thyroid peroxidase (TPO) activity and binding protein are possible mechanisms (50). β-HCH was negatively associated with T4 in this study. This might be explained by the similar bioconcentration factor of β-HCH and HCB, which may lead to a similar mechanism of action.

*Associations of Concentrations of rT3 with POPs*

rT3 is an inactive metabolite of T4 formed by deiodination. Inhibition of DIO activities by POPs may lead to reduction in T4 metabolism, causing lower levels of placental rT3 (55). For example, Leonetti *et* al. reported a negative association between rT3 and BDE 99 among males (24). However, we did not see a significant association between BDE 99 and rT3 here, probably because of the differences in study design and sampling methodology. Our study found that TBT, ΣOTC, β-HCH, and MOC were inversely associated with rT3. Besides, inhibition of the DIO activities can lead to an increase of T4 levels, which was not observed in our study. We also found positive associations between rT3 and PCB 81, 101, 183, 12378-PeCDF, 123478-HxCDF, 123678-HxCDF, 234678-HxCDF, and 1234678-HpCDF, which suggested an alternative mechanism of action. Interestingly, the association between rT3 and OTC placental exposure was assessed for the first time. OTCs are a group of chemicals used as biocides in antifouling paints. TBT can cross the placenta, inducing physiological and morphological changes, resulting in abnormal fetal and postnatal development (56). The present study suggested that *in utero* exposure to OTC, especially TBT, leads to lower level of rT3.

*Associations of Concentrations of T3 with POPs*

We found only few associations between POPs and the biologically active thyroid hormone, T3. Among the 82 POPs investigated, T3 only showed positive associations with 2378-TeCDF and 12378-PeCDF, and negative tendency with β-HCH, suggesting a low sensitivity of T3 homeostasis for POP exposures. Placental T3 is produced by deiodination in the placenta and foetus, whereas T4 is partly derived from the transplacental passage of maternal free T4. Therefore, the feedback regulation, which is induced by T3, may not take place in placenta, and thus maternal pituitary regulation might be out of regulatory function for placental T3.

We also estimated the associations of THs with the WHO-TEQ values of PCDD/Fs (PCDD/F\_WHO-TEQ) and PCBs (PCB\_WHO-TEQ). As shown in **Table S1** (**Supplementary material**), no significant associations were observed.

*Strengths and Limitations*

This study has several unique strengths: (a) This is the first study investigating thyroid-disrupting effect of as many as 82 POPs, as well as the WHO-TEQ values of PCDD/Fs and PCBs, which therefore provides an overview of the possible relationships between the ubiquitous POPs and THs in placenta; (b) UPLC-Q-TOF-MS was adopted for TH analysis. This approach provides better accuracy and reliability than RIA methods used in previous studies, producing more reliable results; (c) A wide variety of demographical characteristics were assessed and considered in the statistical analysis. Some of these variables could influence the THs during pregnancy. Taking various characteristics into consideration increases the robustness of the analysis. However, the study also has certain limitations. For example, the number of samples is limited (n=58), which may reduce the statistical power. Only placenta samples from mothers that gave birth to boys were included in this research, thus we could not reveal any sex-dependent effect. The hydroxylated metabolites of certain POPs (e.g., OH-PCBs and OH-PBDEs), which generally show higher affinities in binding with the TH-binding proteins than the mother compound, were not included in this study. The TH and POP concentrations in this study reflect the situation at delivery instead of the exposure during the entire pregnancy. Our previous study observed considerably higher concentrations of PBDEs per gram fat in breast milk than in placenta and suggested that milk analyses might be more reliable toward the lower end of concentrations (30). Additionally, free THs may be more important for analyzing the effects of POP exposure, while total TH concentrations were measured in this study.

Finally, with as many as 82 POP congeners, their sums and 3 THs, some significant associations may occur by chance. However, in particular the associations between PBDEs and T4 appear robust, as all associations showed the same direction and there is a well-known structure resemblance of PBDEs to T4.

**Conclusion**

In summary, our results suggest that background exposure to POPs can alter thyroid homeostasis in pregnant women, subsequently affecting the thyroid homeostasis in placenta. Our results highlight the challenges of assessing effects on thyroid function, especially during pregnancy, due to the complexity of contaminant mixtures and the sensitivity of the thyroid system of the pregnant woman and the fetus. Finally, the results of this study should be interpreted with caution due to the limited number of subjects included in the analysis. The findings should be confirmed with more placenta samples from both boys and girls, also including the DIO enzyme activities and the hydroxylated metabolites of PBDEs and PCBs.

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**Disclosure Statement**

The authors report no conflicts of interest in this work.

**Associated Content**

**Supplementary Material**

Supplementary information associated with this article is available free of charge at…

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