The effects of exposure to environmental chemicals on child development

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PART 2

Exposure to environmental chemicals and neurological functioning from birth up to and including adolescence

CHAPTER 3

Prenatal exposure to polychlorinated biphenyls and their hydroxylated metabolites is associated with motor development of three-month-old infants

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ABSTRACT

Background: Polychlorinated biphenyls (PCBs) are ubiquitous environmental pollutants that are potentially toxic to the developing brain. Hydroxylated metabolites of PCBs (OH-PCBs) are suggested to be even more toxic. Little is known about their short-term effects on human health.

Objectives: To determine whether prenatal background exposure to PCBs and OH-PCBs was associated with the motor development of three-month-old infants.

Methods: Ninety-seven mother-infant pairs participated in this Dutch, observational cohort study. We determined the concentrations of PCBs and OH-PCBs in maternal blood samples during the third trimester of pregnancy. When the infants were three months old we evaluated their motor development by assessing the presence and performance of spontaneous movement patterns from video recordings. We calculated a Motor Optimality Score (MOS). The score could range from low (5) to high (28) optimality. We explored the correlations between PCB and OH-PCB levels and MOS. Subsequently, we tested whether the levels differed between infants with a low (<26) or high (≥26) MOS and whether the levels associated with detailed aspects of their motor repertoires.

Results: We found several associations between PCB and OH-PCB levels and MOS, including detailed aspects of the early motor development. High 4-OH-PCB-107 levels were associated with a low MOS (P=.013). High PCB-187 levels were associated with reduced midline arm and leg movements (P=.047 and P=.043, respectively). High 4'-OH-PCB-172 levels were associated with more manipulation (P=.033).

Conclusions: Prenatal exposure to high background levels of most PCBs and 4-OH-PCB-107 seems to impair early motor development, whereas only 4'-OH-PCB-172 showed the opposite.

INTRODUCTION

Polychlorinated biphenyls (PCBs), a significantly toxic group of industrial chemical compounds, were widely used commercially in, for example, hydraulic fluids, adhesives, inks and lubricants. Although the production and use of PCBs has been banned by law since 1985 in the Netherlands, humans and animals alike continue to be exposed to the contaminating effects of PCBs still remaining in the environment. Studies of populations with high exposures to PCBs caused by consuming contaminated fish, show neurobehavioral alterations in newborn infants.[1] Some studies in humans reported that prenatal exposure to PCBs might have an impact on mental and motor development, while others did not (for review see Majidi et al.[2]).

During the last decade, techniques have become available to detect the hydroxylated metabolites of PCBs (OH-PCBs) in human serum. Previously, we reported a correlation between the PCB and OH-PCB levels in maternal and umbilical cord plasma, indicating considerable placental transfer of these compounds.[3] The OH-PCB levels in umbilical cord plasma were approximately 50% of maternal levels, whereas the PCB levels were approximately 30% of maternal levels. Regarding the effects of OH-PCBs on child development studies are sparse. Exposure to one OH-PCB compound was negatively associated with neurodevelopment at the age of sixteen months.[4] In a study of children at school age, prenatal exposure to background levels of OH-PCBs correlates with worse fine manipulative abilities, better attention, and better visual perception.[5] Still, little is known about the toxicological impact of PCBs and especially of OH-PCBs on humans and very young infants in particular.

A reliable, non-invasive method to evaluate the brain function of young infants is to assess the quality of their spontaneous motor repertoire.[6] At the age of three months, the infant's motor repertoire is characterized by so-called fidgety movements (FMs). These are spontaneous movements of small amplitude, moderate speed, and variable acceleration in all directions. FMs are highly predictive of subsequent neurological outcome: most infants (96%) with normal FMs have normal neurological outcomes, while most infants (95%) in whom FMs were absent during this specific period develop cerebral palsy.[6] Several studies indicate that detailed aspects of FMs and the concurrent motor repertoire, as expressed in the Motor Optimality Score (MOS), are also valuable to assess neurological function in young infants[7] and predictive for functional outcome at school age.[8] As the period around 3 months after term is among the earliest when to assess development reliably, associations found between development and prenatal exposure to compounds may be relevant and important. Because of the limited time frame between exposure and assessment, effects of postnatal factors on development may be minimal.

The aim of this study was to determine whether prenatal background exposure to OH-PCBs and PCBs was associated with the motor development of three-month-old infants.

Because previous human and animal studies showed impaired developmental outcomes after prenatal exposure to OH-PCBs and PCBs, we hypothesized that exposure to higher levels of these compounds would be associated with poorer quality of the early motor development.

METHODS

Cohort

Ninety-seven mother-infant pairs were included in this observational study between September 1998 and December 2000. They were members of a cohort participating in prospective studies on exposure to PCBs and OH-PCBs and their potential effects on the development of the newborn infant.[3] Women from the northern part of the Netherlands were invited to participate by their midwife or obstetrician. Only women of Western European origin, who spoke Dutch as their native language, were included. Women whose pregnancy or delivery had involved serious illness or complications were excluded. Only infants born at term (between 37 to 42 weeks' gestation), who had no congenital anomalies or diseases, were included. Infants who had been admitted to a hospital more than one day after birth were not included. All parents had given their informed consent before videotaping commenced. The study was approved by the medical ethics committee of the University of Groningen.

Measurement of PCBs and OH-PCBs

Maternal blood samples were taken during the third trimester of pregnancy. A detailed description of the analysis of the blood samples is provided by Soechitram et al.[3] For this study the following ten PCBs were measured: PCB-105, PCB-118, PCB-138, PCB-153, PCB-170, PCB-180, PCB-187, PCB-146, PCB-183, and PCB-156. The following six OH-PCBs were measured: 4-OH-PCB-107, 3-OH-PCB-153, 4-OH-PCB-146, 3'-OH-PCB-138, 4-OH-PCB-187 and 4'-OH-PCB-172. The parent PCB compounds of the measured OH-PCBs are shown in Table 2a in the results section.[3] We used the same extraction and clean-up procedure as described by Hovander et al.[9] The samples were analyzed at the analytical laboratory participating in the study. We numbered the PCBs according to Ballschmiter et al.[10] and we numbered the OH-PCB congeners according to Letcher et al.[11] Concentrations of PCBs are given in ng/g lipid in maternal blood. Because they are hydrophilic, concentrations of OH-PCBs are expressed per gram fresh weight.

Assessment of the motor development

Video recordings were made at approximately three months of age during an outpatient visit to the clinic. A ten-minute optimal recording is sufficient to reliably assess the quality of an infant's motor repertoire.[6] All recordings were made with the infants lying supine and dressed lightly and comfortably. The infants' arms and legs were bare and they could move their limbs and trunks freely. The recordings were made during a period of active wakefulness. Interferences were avoided during recording. We also took care that the infants were not fussing, crying or sucking on a pacifier, as it is impossible to assess the quality of the motor repertoire under such circumstances.

The motor development of the three-month-old infants was assessed off-line by authors SAB and MMH according to the method described by Einspieler and Prechtl[6] and Bruggink et al.[8] MMH is certified by the GM Trust as an advanced scorer. SAB was trained by AFB and MMH. Author AFB is a licensed tutor and co-founder of the General Movements Trust. In case of doubt on the presence of normal or abnormal FMs or about the assessment of the MOS, the infants' recordings were re-evaluated with AFB to make the definitive judgment.

First, we assessed the quality of the FMs. Normally, FMs first occur at six to nine weeks post-term, and remain present until fifteen to twenty weeks, at which age intentional and antigravity movements appear and start to dominate the repertoire. We classified FMs as normal (N) or abnormal (A). Normal FMs are movements of small amplitude, moderate speed, and variable acceleration and involve movements of the neck, trunk, and limbs, in all directions. They are continually present, except during fussing and crying. Abnormal FMs resemble normal FMs but are exaggerated regard to amplitude, speed, and jerkiness. If FMs were not observed we noted this as 'absence of fidgety movements' (classified as abnormal FMs).

Second, we performed a more detailed analysis of the motor development by combining the quality of FMs with the concurrent motor repertoire. We used the Motor Optimality List compiled by Bruggink et al.[8] to determine the infant's MOS at three months post-term. The list distinguishes three aspects of the motor development: movement patterns, postural patterns, and movement character. The Motor Optimality List consists of five subcategories covering these three aspects:

1) Fidgety movements (FMs)
2) Repertoire of co-existent other movements
3) Presence and normality of individual movement patterns
4) Presence and normality of individual postural patterns
5) Quality of the concurrent motor repertoire

Each subcategory, except FMs, is assigned a score of either 4, 2, or 1, in descending order of normality. FMs are assigned a score of either 12 (normal), 4 (abnormal), or 1 (absent). The sum of the scores of these subcategories is the Motor Optimality Score. For three-month-old

infants MOS can range from low optimality with a minimum of 5 points to high optimality with a maximum of 28 points. In addition to MOS, we also dichotomized the score into a low (<26) or high (≥26) MOS. A low MOS is characterized by an abnormal score on more than one subcategory of the Motor Optimality List.

Finally, we determined which subcategories explained the lower MOS, i.e. were non-optimal. Next we analyzed those subcategories that were non-optimal in more than one fifth of the group of infants in more detail, to determine which movement and/or postural patterns, or which qualitative aspect of movement character, were responsible for the lower scores.

Inter-observer reliability

Because the qualitative assessment of the movements is based on pattern recognition, it is important that the inter-observer agreement is adequately high, so the method can be considered as objective. Einspieler et al.[12] found an inter-observer agreement with Cohen's kappa between 0.78 and 0.98 and Fjørtoft et al.[13] reported an inter-observer reliability in the assessment of FMs with Cohen's kappa of 0.75-0.91. Regarding presence of movement and postural patterns disagreement was reported between observers ranging from 1.2% for movement patterns and 2.6% of postural patterns.[8]

Statistical analyses

For statistical analyses we used the Spearman rank correlation test and the Mann-Whitney-U test to investigate correlations and associations between prenatal exposure to PCBs and OH-PCBs and several outcome variables. We used the following outcome variables:

1) Quality of FMs;

2) Scores on the Motor Optimality List, including MOS with subdivision in high MOS (≥26) and low MOS (<26);

3) Aspects of the subcategories of the Motor Optimality List, including movement clusters and the character of the movements, if more than one fifth (20 percent) of the group of infants had non-optimal scores regarding this particular subcategory.

We used logistic regression models to investigate the ORs. To determine whether the relation between PCBs and OH-PCBs and the quality of the motor development was confounded by other characteristics (gender, gestational age, birth weight, age at recording, maternal education level, and maternal smoking and alcohol consumption during pregnancy), we performed univariate logistic regression analyses. These characteristics were considered as possible confounders if they had a *P* value of less than .2 in the univariate analysis model. We performed multivariate logistic regression analyses (method: enter) to assess whether these confounders had influenced our results. A *P* value of less than .05 we considered statistically significant and a *P* value of more than .05 but less than .10, marginally significant. We used

the Statistical Package for the Social Sciences, version 18.0.3 (SPSS Inc, Chicago, Ill.) for the statistical analyses.

RESULTS

Of the 104 infants initially included in the study, seven were later excluded for various reasons: for three infants no PCB or OH-PCB values were available, two infants had not been videotaped at three months due to logistic reasons, and two infants were excluded because their motor development could not be evaluated from videotape because the recordings were too short. The final study group consisted of 97 mother-infant pairs. In Table 1 we present the characteristics of the mothers and infants who participated. The median age of the infants at the time of videotaping was 14 weeks (range 11 to 17 weeks). The concentrations of all measured PCBs and their respective OH-PCBs are given in Table 2a.

Table 1. Characteristics of the study group (N=97 mother-infant pairs)

Characteristic	Value
Gender, male/female	51/46
Gestational age (weeks)	40 ± 1
Apgar score 3 min	10 (6-10)
Birth weight (grams)	3607 ± 494
Maternal education level	
Below average (≤11 years education)	13
Average (12-13 years education)	40
Above average (≥14 years education)	44
Maternal smoking during pregnancy yes/no	22/75
Maternal alcohol consumption during pregnancy yes/no	24/73
Age at recording (weeks)	14 ± 1
Motor Optimality Score	26 (18-28)
1) Fidgety movements	96/1
2) Repertoire of co-existent other movements	77/20
3) Presence and normality of individual movement patterns	95/2
4) Presence and normality of individual postural patterns	96/1
5) Quality of the concurrent motor repertoire	50/47

Data are given as frequencies (n/n), median (min-max), or mean ± SD. The subcategories of the Motor Optimality List are displayed as number of infants with normal/reduced scores.

Table 2a. Concentrations of PCBs and their respective OH-PCBs in maternal serum during pregnancy

Compound	Concentration (ng/g lipid weight)	Compound	Concentration (ng/g fresh weight)
PCB-105	4.0 (2.1-11.0)	4-OH-PCB-107	0.067 (0.039-0.102)
PCB-118	20.9 (14.6-32.8)		
PCB-138	67.9 (47.3-86.3)	4-OH-PCB-146	0.070 (0.053-0.102)
		3'-OH-PCB-138	0.046 (0.033-0.066)
PCB-153	89.8 (63.1-124.0)	3-OH-PCB-153	0.038 (0.025-0.055)
		4-OH-PCB-146	
PCB-170	19.0 (13.1-25.6)	4'-OH-PCB-172	0.017 (0.010-0.022)
PCB-180	44.9 (31.2-58.7)		
PCB-187	11.8 (8.5-17.4)	4-OH-PCB-187	0.137 (0.105-0.174)
PCB-146	8.0 (4.6-13.2)		
PCB-183	8.1 (5.5-10.4)		
PCB-156	11.1 (7.6-14.5)		
Sum PCBs	290.0 (215.7-391.4)	Sum OH-PCBs	0.389 (0.288-0.535)

Data are given as median (interquartile range)

Motor development

Out of the 97 infants, 96 showed normal FMs. One infant had abnormal FMs. MOS in our study group ranged from 18 to 28. Out of the 97 infants, seventeen infants had a low MOS (<26) and eighty infants had a high MOS (≥26). Table 1 illustrates that a relatively large proportion of the infants had abnormal scores on the second and fifth subcategories. We examined these subcategories in more detail. The second subcategory on the Motor Optimality List assesses the age-adequacy of the repertoire of co-existent other movements. Twenty infants had abnormal scores on this subcategory. As shown in Table 2b, we clustered several movement patterns into one of four categories to provide more detail. Most infants showed midline leg movements, manipulation movements, and antigravity movements, while approximately a third of the infants showed no midline arm movements. Manipulation includes the following movement patterns: hand-hand manipulation, foot-foot manipulation and fiddling at cloths or blanket. The fifth subcategory on the Motor Optimality List scores the character of the concurrent movements and represents a global score of movement character. Fifty infants showed smooth and fluent movement character, whereas 47 infants showed abnormal movement character. The frequencies of abnormal movement characters are given in Table 2b. Several infants scored more than one abnormal movement character, e.g. monotonous and cramped.

Table 2b. Aspects of the second and fifth subcategory of the Motor Optimality List (N=97)

Movement clusters	**Present**	**Absent**
Midline arm movements	61	36
Midline leg movements	91	6
Manipulation	86	11
Antigravity movements	94	3
Character (global score)	**Present**	**Absent**
Jerky	13	84
Monotonous	25	72
Cramped	10	87
Cramped-synchronized	1	96
Tremulous	15	82
Stiff	18	79

PCB and OH-PCB levels and motor development

We found no associations between PCB and OH-PCB levels and the quality of FMs. We also found no correlations between PCB and OH-PCB levels and MOS. After dichotomizing MOS into a low and high MOS, higher levels of OH-PCB-107 were found to relate to low MOS (*P*=.04). No other compound was associated with a low MOS (see supplementary material). Exposure to higher OH-PCB-107 was associated with a reduced repertoire of co-existent other movements (*P* =.08).

Next, we investigated whether exposure to PCBs and OH-PCBs was associated with the movement clusters within the repertoire of co-existent other movements (Table 3). High levels of PCB-187 were associated with reduced midline arm movements and reduced midline leg movements, *P*=.04 and *P*=.06, respectively. High levels of PCB-183 were associated with reduced midline leg movements and reduced manipulation, *P*=.01 and *P*=.05, respectively. High levels of 4'-OH-CB172 were associated with more midline leg movements and more manipulation, *P*=.04 and *P*=.02, respectively. We also found an association between exposure to some PCB levels and the presence of antigravity movements. High levels of the following compounds were associated with reduced antigravity movements: PCB-118, PCB-146, PCB-105, PCB-138 (*P*<.05). The associations between high levels of PCB-153, PCB-156, and the sum of the PCBs measured, were nearly significant with reduced antigravity movements (*P*<.10). We then determined whether exposure to PCBs and OH-PCBs was associated with movement character. Regarding the scores on the fifth subcategory, quality of the concurrent motor repertoire, we found that high levels of PCB-118 and PCB-138 were associated more frequently with a cramped movement character (*P*=.05 and *P*=.09, respectively). The sum of the PCB levels measured was also more frequently associated with a cramped movement character (*P*=.09).

Table 3. Prenatal exposure to PCBs and OH-PCBs related to movement patterns and movement character at the age of three months.[a]

Compound	Clusters of movement patterns				Cramped movement character
	Absent midline arm movements	Absent midline leg movements	Absent manipulation	Absent antigravity movements	
PCB-118				++	++
PCB-146				++	
PCB-153				+	
PCB-105		+		++	
PCB-138				++	+
PCB-187	++	+			
PCB-183		++	+		
PCB-156				+	
PCB-180					
PCB-170	(+)				
Sum PCBs				+	+
4-OH-PCB-107					
3-OH-PCB-153					
4-OH-PCB-146			-		
3'-OH-PCB-138					
4-OH-PCB-187			-		
4'-OH-PCB-172		- -	- -		
Sum OH-PCBs					

[a] Calculated by the Mann-Whitney-U-test; ++ $P<.05$ and + $P<.10$, (+) $P=.101$, indicating that exposure to higher levels is associated with absent movements or a cramped character; - - $P<.05$ and - $P<.10$, indicating that exposure to higher levels is associated with presence of the movements.

Multivariate analyses

We determined whether other factors might have confounded our findings. Univariate logistic regression analyses revealed that only maternal smoking during pregnancy, a higher gestational age, and female gender were associated with a low MOS at $P<.20$ (Table 4). Next, we performed univariate logistic regression analyses to determine the odds ratios (ORs) for the effect of exposure to PCBs and OH-PCB compounds on the motor development. In Table 5 we present those ORs that almost reached significance at $P<.10$. Finally, we repeated the logistic regression analyses adjusting for these potential confounders and provide the adjusted ORs in Table 5 as well. Although the ORs did not change much after adjustment, some significant associations disappeared. Others remained significant. These included the associations between PCB-118, PCB-187, 4-OH-PCB-107, 4'-OH-CB172 and several aspects of the motor development.

Table 4. Unifactorial analyses of factors associated with MOS <26

Variable	OR (95% CI)	*P*-value
Female gender	2.36 (0.79-7.00)	0.123 *
Gestational age >40 weeks	2.53 (0.76-8.43)	0.131 *
Birth weight >3660 grams	0.49 (0.17-1.46)	0.203
Recording <30 min after feeding	0.67 (0.04-10.25)	0.771
Maternal smoking during pregnancy	3.03 (0.99-9.27)	0.052 *
Maternal alcohol use during pregnancy	1.34 (0.42-4.28)	0.624
Age at recording >14 weeks	0.63 (0.20-1.95)	0.417

MOS= Motor Optimality Score; **P*<.20.

Table 5. Associations between PCB and OH-PCB levels and aspects of the motor repertoire

Compound	Observed movement patterns	OR (95% CI)	OR adjusted for confounders [a] (95% CI)
PCB-118	Absent antigravity movements	1.41 (0.98-2.04) [b] *	1.84 (1.02-3.32) [b] **
PCB-146	Absent antigravity movements	4.09 (0.81-20.64) [b] *	3.80 (0.65-22.26) [b]
PCB-138	Absent antigravity movements	1.27 (0.96-1.68) [b] *	1.21 (0.90-1.62) [b]
PCB-187	Absent midline arm movements	1.96 (1.01-3.79) [b] **	1.81 (0.89-3.67) [b]
	Absent midline leg movements	2.80 (1.03-7.57) [b] **	2.74 (0.96-7.87) [b] *
PCB-156	Absent antigravity movements	11.06 (0.73-168.85) [b] *	8.47 (0.48-148.96) [b]
PCB-170	Absent midline arm movements	1.57 (0.94-2.64) [b] *	1.46 (0.85-2.53) [b]
Sum PCBs	Absent antigravity movements	1.07 (0.99-1.15) [b] *	1.06 (0.98-1.15) [b]
4-OH-PCB-107	MOS < 26	3.56 (1.30-9.73) [c] **	2.79 (0.94-8.32) [c] *
	Reduced repertoire of co-existent other movements	3.32 (1.26-8.74) [c] **	2.79 (0.99-7.83) [c] *
4'-OH-PCB-172	Absent midline leg movements	0.17 (0.02-1.38) [d] *	0.17 (0.02-1.42) [d]
	Absent manipulation	0.24 (0.06-0.89) [d] **	0.19 (0.05-0.79) [d] **

[a] Confounders were gender, gestational age and maternal smoking during pregnancy; [b] per 10 ng/g lipid weight; [c] per 0.10 ng/g fresh weight; [d] per 0.01 ng/g fresh weight; ***P*<.05; **P*<.10; MOS= Motor Optimality Score.

DISCUSSION

This observational cohort study indicated that prenatal background exposure to PCBs and OH-PCBs may be associated with the motor development of three-month-old infants. We found no associations between PCB and OH-PCB levels and the quality of FMs, but associations did exist between several PCBs and OH-PCB levels and aspects of the infants' motor development. Firstly, prenatal exposure to higher levels of 4-OH-PCB-107 was associated with a poorer quality of the motor development as reflected by a lower MOS. Secondly, some PCBs and OH-PCBs were associated with the acquisition of developmental milestones, such as the presence of midline movements, manipulation, and antigravity movements. Most associations might indicate delayed development of age-adequate movements after exposure to higher levels of the compounds, except for some associations where we found the opposite. Thirdly, exposure to higher levels of PCB-118 was associated more frequently with a cramped movement character. The results partially confirmed our hypothesis that prenatal exposure to higher PCB and OH-PCB levels was associated with a poorer quality of the early motor development.

Our most important finding was that exposure to high 4-OH-PCB-107 levels was associated with a low MOS (< 26), although the strength of the association weakened after adjustment for confounders. No other PCBs or OH-PCBs correlated with MOS. Higher levels of 4-OH-PCB-107 were associated almost significantly with a reduced repertoire of co-existent other movements. That 4-OH-PCB-107 might be more toxic compared to other OH-PCB compounds is consistent with findings in earlier studies in humans and animals. Regarding a study in rats, this metabolite was found to induce long-term adverse effects on behaviour and neurodevelopment in rats.[14] In human studies, Park et al. found that 4-OH-PCB-107 is the only metabolite that associates significantly with a decrease in the Mental Development Index of sixteen-month-old infants.[4]

A second important finding of our study was that prenatal exposure to higher PCB and OH-PCB levels was associated with the development of age-adequate movement patterns. Exposure to higher levels of 4'-OH-PCB-172 was associated with more manipulation and more midline leg movements. We found a near-significant association of exposure to higher levels of 4-OH-PCB-146 and 4-OH-PCB-187 and more manipulation. The study by Roze et al. suggests negative effects of hydroxylated metabolites on manipulation in children at school age, while we found predominantly positive effects on manipulation in three-month-old infants.[5]

Unlike exposure to OH-PCBs, exposure to higher levels of PCBs was negatively associated with the development of age-adequate movements. Exposure to higher levels of PCB-105, PCB-187, PCB-183, and PCB-170 was associated with reduced midline movements of arms or legs or of arms and legs. Exposure to higher levels of PCB-183 was also associated with

reduced manipulation. Although only three infants had no antigravity movements, we would like to emphasize that we did find an association between exposure to higher levels of six out of ten PCBs and absent antigravity movements. Our findings may suggest that prenatal exposure to higher levels of PCBs has negative effects on motor development. Whether these negative effects persist during later life requires further study.

Our results of adverse effects of PCBs on motor development were also consistent with findings in animal studies. For a review see Faroon et al.[15] Two studies suggested that OH-PCBs might be even more neurotoxic than PCBs.[16, 17] We could confirm this for OH-PCB-107. In general, however, our findings in three-month-old infants suggested that exposure to OH-PCBs might be less neurotoxic compared to exposure to PCBs. Interestingly we even found positive associations between motor development and several OH-PCBs. It is difficult to determine what the implications of these findings are for functioning in later life. A possible explanation for the difference in neurotoxicity between PCBs and OH-PCBs might be the difference in exposure levels: the exposure to PCBs was higher compared to the exposure to OH-PCBs. This observation requires more detailed study.

Our last finding was that exposure to higher levels of PCB-118 was associated more frequently with cramped movements. The association of exposure to PCB-138 and the sum of the PCBs measured nearly reached significance with cramped movements. Exposure to PCB and OH-PCB levels was not associated with other abnormal movement characters. Further study is required to evaluate the relation between prenatal exposure to environmental toxins and motor development, including movement character, in later life.

Our findings can be explained in several ways. Although there are many theories on the way PCBs and their hydroxylated metabolites may affect neurodevelopment, the main hypothesis involves disruption of thyroid hormone homeostasis.[18] Thyroid hormones regulate neuronal proliferation and cell migration and differentiation, as well as the timing of these processes. Studies in rats showed that the transport of thyroid hormone to the brain requires thyroxine (T4) to pass through the blood-brain barrier by binding to the thyroid hormone transport protein transthyretin (TTR).[19] A study on rats found that the metabolite 4-OH-PCB-107 binds to TTR in maternal and fetal plasma, suggesting that the binding of a compound to TTR *in vivo* could lead to facilitated maternal to fetal transfer, decreased maternal and fetal plasma T4 levels, and decreased fetal brain T4 levels.[20] Hydroxylated metabolites of PCBs show *in vitro* binding affinities as high as twelve times the binding affinity of the natural ligand T4.[21] According to these findings the binding of OH-PCBs to TTR may lead to brain thyroid hormone deficiencies *in utero,* possibly affecting brain development.

The strength of our study is that we examined in great detail the influence of prenatal background exposure to PCBs and OH-PCBs on several aspects of motor development at a very young age. There are, however, several limitation that need to be pointed out.

The first limitation of our study is the high likelihood of Type I error because of the large number of comparisons. Because we tested PCBs and OH-PCBs in relation to specific aspects of the quality of infants' motor development, some results could be due to chance. Nevertheless, we believe that exploring different PCBs and OH-PCBs was justified as part of a careful evaluation of a rich data set in the context of hypothesis-driven research.[22] We aimed to report our results transparently, therefore we did not adjust our threshold for significance for multiple comparisons. Being a hypothesis generating study, and considering the fact that we investigated background exposures of PCBs and OH-PCBs, we focused in our analyses in particular on associations, and also regarded those with significance P<.10 as potentially interesting. A second limitation is the small sample size. We included 104 mother-infant pairs in our study of which data from 97 mother-infant pairs could be analyzed. A third limitation is that we included the mother-infant pairs on a voluntary basis. Although we expected to have drawn a random sample of mothers from our region, a certain amount of bias on the part of the mothers cannot be ruled out. On the one hand, some mothers may have been aware of the concerns about these environmental pollutants and would perhaps have been selective about what they ate. On the other hand, other mothers may have been equally aware of the concerns but may have lacked the means of being selective about their food-intake and worried about what they had eaten as a consequence. Nevertheless, we believe that on the whole our sample is a valid representation of the population in our region.A fourth limitation is that because the threshold levels of the toxicity of PCBs and OH- PCBs are unknown, we did not statistically test low versus high PCB and OH-PCB levels in relation to the early motor development. A final limitation is that we cannot exclude the possibility of confounding by other persistent organic pollutants. Nevertheless, it seems rather unlikely that these pollutants associated with only one PCB metabolite, i.e. 4-OH-PCB-107, to the exclusion of other metabolites in our study.

CONCLUSIONS

Prenatal exposure to high background levels of most PCBs and 4-OH-PCB-107 seems to impair the motor development of three-month-old infants, whereas only 4'-OH-PCB-172 was associated with a more optimal quality of the infants' motor development. Our results suggested that prenatal exposure to background levels of PCBs and their metabolites may influence the motor development in three-month-old infants adversely and sometimes favorably. In general, the PCBs seem more neurotoxic than their metabolites. More research is needed to investigate whether prenatal exposure is also associated with functional developmental outcomes in later life.

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SUPPLEMENTARY MATERIAL

Supplementary Table 1. Associations between PCB- and OH-PCB-levels and MOS <26

Compound	OR (95% CI)	OR adjusted for confounders [a] (95% CI)
PCB-118	0.92 (0.66-1.28) [b]	0.92 (0.64-1.32) [b]
PCB-146	0.94 (0.39-2.26) [b]	0.80 (0.31-2.07) [b]
PCB-153	1.01 (0.90-1.14) [b]	0.97 (0.86-1.10) [b]
PCB-105	0.74 (0.35-1.55) [b]	0.73 (0.37-1.45) [b]
PCB-138	1.05 (0.90-1.23) [b]	1.01 (0.85-1.19) [b]
PCB-187	1.20 (0.56-2.58) [b]	0.93 (0.41-2.10) [b]
PCB-183	0.97 (0.81-1.16) [b]	0.94 (0.79-1.13) [b]
PCB-156	0.80 (0.27-2.38) [b]	0.64 (0.20-2.10) [b]
PCB-180	1.01 (0.78-1.31) [b]	0.96 (0.73-1.26) [b]
PCB-170	1.15 (0.61-2.14) [b]	0.98 (0.50-1.90) [b]
Sum PCBs	1.01 (0.97-1.05) [b]	1.00 (0.96-1.04) [b]
4-OH-PCB-107	3.56 (1.30-9.73) [c] **	2.79 (0.94-8.32) [c] *
3-OH-PCB-153	1.55 (0.26-9.05) [c]	1.08 (0.14-8.42) [c]
4-OH-PCB-146	1.39 (0.43-4.54) [c]	1.32 (0.37-4.66) [c]
3'-OH-PCB-138	2.18 (0.61-7.85) [c]	1.77 (0.41-7.55) [c]
4-OH-PCB-187	1.42 (0.56-3.60) [c]	1.39 (0.52-3.72) [c]
4'-OH-PCB-172	0.67 (0.30-1.53) [d]	0.65 (0.28-1.51) [d]
Sum OH-PCBs	1.24 (0.93-1.65) [c]	1.18 (0.86-1.62) [c]

[a] Adjusted for the confounders gender, gestational age and maternal smoking during pregnancy; [b] per 10 ng/g lipid weight; [c] per 0.10 ng/g fresh weight; [d] per 0.01 ng/g fresh weight; ***P*<.05; **P*<.10; MOS= Motor Optimality Score.