FETAL PCB AND MEHG EXPOSURE: EFFECTS ON VISUAL P300 IN 5-YEAR-OLD INUIT CHILDREN

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ABSTRACT

Inuit communities living in Nunavik (Northern Québec, Canada) are highly exposed to polychlorinated biphenyls (PCBs) and methylmercury (MeHg) from marine food intake. A birth-cohort study was initiated within this population to evaluate the effects of perinatal exposure to contaminants on child development. Neurophysiological indices of cognitive functioning, the N1, P2 and P3 waves of event-related potentials (ERP), were measured in 5-year-old children. The main hypothesis was that prenatal exposure to contaminants would be associated with increased ERP latencies, suggesting delayed cognitive processing. Altogether, 111 children (mean age = 5.4 years) underwent neurophysiological testing. A visual Oddball paradigm was used. Latencies and amplitudes of N1, P2 and P3 at Cz electrode location were measured for deviant stimuli. Assessments from only 20 children satisfied the measurement criteria and were retained for statistical analyses. Hierarchical multiple regressions revealed longer P3 latency as a function of cord blood Hg ($p \leq .05$), while child's blood Hg levels at examination were related to shorter N1 latency ($p \leq .05$). No significant association was found for PCB exposure. The present results suggest that prenatal MeHg exposure delays the cognitive processing of information, and are in accordance with what was found in the Faroe Islands using neurobehavioral measures of attention. The absence of a significant relation between PCB exposure and ERP components contrasts with previously reported results from Rotterdam and Taiwan cohorts among older children, and may be related to the small number of children with reliable data in the present study. These results emphasize the need to reassess those children at a later age with similar methodology in a larger sample.

INTRODUCTION

The Inuit population living in Nunavik (Northern Québec, Canada) is likely to be exposed to neurotoxic levels of MeHg and PCBs due to their geographic situation and their high fish and marine mammal consumption. Multiple cohort studies suggest that exposure to such contaminants appears to be particularly harmful to the developing brain. Prenatal exposure to MeHg and PCBs has been related to different impairments on cognitive functioning, notably on attentional resources. As neurobehavioral measures are usually used to assess the subtle effects of exposure to moderate levels of these contaminants on attentional functions, neurophysiological measures are gaining popularity in the study field because of higher sensitivity and independence from cultural bias. Up to now, two studies were published that used a neurophysiological measure of cognition, the P300 component of the event-related potentials (ERPs), to assess the effects of prenatal PCB exposure in school-age children. In Taiwan, 7- to-12-year-old children prenatally exposed to PCBs showed longer P3 wave latencies and weaker amplitudes on an auditory oddball task, suggesting a delay in information processing and decreased attentional allocation[10]. In the Netherlands, 9-year-old children exposed to PCBs also showed longer latencies of the P3 wave on a similar auditory oddball task[19]. Although the developmental toxicity of MeHg has not been studied with cognitive ERPs yet in birth-cohort studies, the results of neurobehavioral assessments in the Faroese cohort suggest impairments of processing speed and short-term memory[16], which could be reflected by the P3 wave[16]. The present study used a visual Oddball paradigm to study information processing in preschool Inuit children from Nunavik exposed to both PCB and MeHg before and after birth.

METHOD

Participants

The study sample included 111 Nunavik Inuit children aged between 5 and 6 years who were originally involved in monitoring program aimed to document prenatal exposure to the environmental contaminants present in the food web between 1993 and 1996. The Nunavik region is located north of the 55th parallel, about 2,000 km from the Great Lakes in the United States (See Fig. 1).

Visual P300 and EEG recordings

A visual P300 protocol with oddball paradigm was used to measure cognitive information processing. The child sits still in front of a computer screen and is told to press a button as quickly as possible when the deviant stimulus (blue circle) appears, whereas no answer must be given when the standard stimulus (8 cm² red squares) is shown. Inter-stimulus interval was set at 1.2 second. This protocol included 3 trials of 125 stimuli presentation, and deviant stimuli were presented in 20% of the trials.

Data acquisition was performed with InstEP Systems™. ERPs were recorded with Ag-AgCl electrodes at frontal (Fz), central (Cz), parietal (Pz), occipital (Oz) and mastoid (M2) locations. Reference and ground electrodes were located on nose and forehead, respectively. High and low pass filters were set at 0.16 and 30. A pre-stimulus interval of 100 ms served as baseline, and an artefact rejection of 75 µV was used. N1, P2 and P3 waves were hand-peaked by an experimented neurophysiologist. Latency for each component was determined from the stimulus onset to the maximal peak whereas amplitude was calculated peak-to-peak. The grand averages of the retained participants are shown on Fig. 2.

N1 : Negative reflection occurring between 70 and 130 ms. Reflects the allocation of a channel for information processing out of the primary cortex.

P2 : Positive reflection occurring between 130 and 220 ms. Represents the inhibition of other channels of information competing for attention and further processing.

P3 : Positive reflection occurring between 250 and 750 ms. Reflects processes involved in the updating of working memory.

Biological measures

A 30-ml blood sample was obtained from the umbilical cord after it was severed, and a 10-ml blood sample was obtained from each participating child at time of testing. These samples allowed the assessment of mercury (Hg), PCB congeners and docosahexaenoic acid (DHA) concentrations. PCB congeners were measured in purified extracts with high-resolution gas chromatography. Total Hg concentrations were determined by cold vapour atomic absorption spectrometry. Analyses were performed at the laboratory of the Centre de Toxicologie du Québec. The fatty acid composition of plasma phospholipids was determined by the Lipid Analytical Laboratory at the University of Guelph. Phospholipids were isolated from the lipid extract by thin-layer chromatography. After transmethylation, the fatty acid profile was determined by capillary gas-liquid chromatography.

RESULTS

Analyses involving environmental contaminants were conducted with natural log-transformed values. Outlying data (>3 standard deviations below or over mean) were corrected following Winer's procedure[18]. A broad range of potential confounding variables pertaining to demographic and maternal characteristics as much as biological variables were documented. The relations between contaminant exposure and ERP components at Cz were analyzed with hierarchical multiple regressions. Co-variables related ($p<.20$) to both independent and dependent variables were included in the first step of the regression.

Of the 111 children taking part to the study, only 20 satisfied the criteria for the analyses of the ERPs. Main reasons for exclusion were lack of cooperation, misunderstanding of the task, computer crash, hearing problem and artefacts in the EEG signal resulting in insufficient accepted trials. The characteristics of both the whole sample and the retained participants are presented on Table 1. Hg exposure and DHA concentrations were similar between groups whereas PCB exposure was higher in the ERP-retained sample.

Intercorrelations between Hg, PCB 153 and DHA concentrations are presented on Table 2. Significant correlations were found between cord and child Hg, cord and child PCB 153, cord Hg and cord PCB 153, and between child Hg and child DHA.

Results of the regression analyses are presented on Table 3. After controlling for con-founders, cord Hg concentrations were significantly associated with longer P3 latency, and blood Hg levels at time of testing were significantly related to shorter N1 latency. DHA levels in cord blood and in child blood at testing tended to be related to shorter P3 latency and greater N1 amplitude, respectively. Cord and child PCB were not related to the ERP components documented.

DISCUSSION

This is the first study to assess the developmental neurotoxicity of MeHg with cognitive ERPs in a non-clinical population. The results suggest that prenatal MeHg exposure is related to slower information processing which could affect working memory efficiency, independently of sensory or motor impairments. This is in accordance with what was found in the Faroese cohort in seven-year-old children assessed with a neurobehavioral test battery. Interestingly, cord DHA levels tended to have the opposite effect, and control for this variable accentuated the statistical relation between cord Hg and P3 latency. These results outline the relevance of assessing polyunsaturated fatty acids when studying the effects of environmental contaminants in fish-eating populations.

Child's blood Hg level at testing was related to faster processing at the primary stages of information processing, which is also in accordance with what was found with visual ERP's in the same cohort[16], although this result is hard to interpret.

In this study, PCB exposure was not related to any of the ERP component studied. Although this contrasts with results obtained in two other cohorts of older children, this is in accordance with the findings of more adverse effects of MeHg over PCB in seafood eaters reported in the Faroe Islands[7]. It is possible that a lack of statistical power is responsible for these results, or that effects would appear at a later age.

To our knowledge, this is the first study for which such ERP protocols were used with so young children (4-6 years). For technical reasons, many subjects were excluded from the analyses, which limits the interpretations that can be put forward. The same children and 200 others from the same population are currently being assessed with auditory P300 protocol at age 10, which will provide the opportunity to improve our understanding of the present results.

Furthermore, since the number of subjects in the retained sample was very low, results from the present study should be interpreted with caution. Replication of the results in other cohort studies is necessary to conclude of such effects.

Figure 1

North America and Nunavik

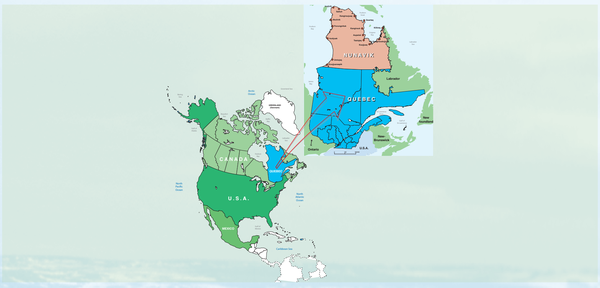

Figure 2

P300 Grand Average for the retained participants

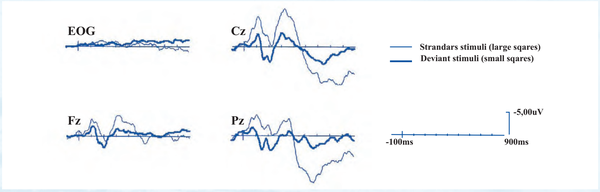

Table 1

Sample characteristics

	Total sample			P300 retained sample			t-test
	n	Mean	Range	n	Mean	Range	
Child characteristics							
Gender (% female)	111	56%		20	75%		**
Age at testing (yrs)	111	5.43	4.78 - 6.17	20	5.50	4.96 - 6.06	n.s.
Breastfeeding duration (weeks)	109	57.47	0 - 258	20	73.880	- 255.7	n.s.
Maternal characteristics							
Age	110	24.47	15 - 39	20	23.35	15 - 32	n.s.
Nb years of schooling	111	9.0	5 - 16	20	8.57	5 - 16	n.s.
Socioeconomic status (SES)1	108	28.46	8 - 57	20	25.93	8 - 53.5	n.s.
Parity	110	4.25	1 - 10	20	4.40	2 - 8	n.s
Crowding	109	1.00	0.5 - 2.0	20	1.02	0.57 - 1.40	n.s.
Depressive symptoms	96	9.07	5 - 15	19	8.79	6 - 13	n.s.
Biological variables							
Cord Hg (nmol/L)	111	111.8	9 - 520	20	107.25	11 - 310	n.s.
Child Hg (nmol/L)	111	47.50	1 - 191	20	42.30	2 - 174	n.s.
Cord PCB 153 (µg/kg of lipids)	111	122.5	22 - 654	20	130.88	23 - 490	t
Child PCB 153 (µg/kg of lipids)	110	159.9	7.5 - 1467	20	204.174	11.2 - 1467	t
Cord blood lead (µmol/L)	111	0.24	0.04 - 1.31	20	0.21	0.07 - 0.50	n.s.
Child blood lead (µmol/L)	111	0.26	0.05 - 1.79	20	0.15	0.05 - 0.57	n.s.
Cord DHA (% phospholipids)	102	3.36	1.12 - 6.35	17	3.69	2.14 - 5.95	n.s.
Child DHA (% phospholipids)	110	2.42	0.57 - 4.86	20	2.34	1.175 - 4.17	n.s.
Alcohol during pregnancy (%)	106	39.6%		20	40%		n.s
Tobacco during pregnancy (%)	108	77.8%		20	80%		n.s.
Marijuana during pregnancy (%)	106	23.6%		19	15.8%		*

[1]Hollingshead index for the mother and her partner or, if she was not self-supporting, for her primary source of support (Hollingshead, 1975).
† $p \leq .10$; * $p \leq .05$; ** $p \leq .01$

Table 2

Intercorrelations between plasma PCB 153, blood Hg and blood DHA concentrations

	Mercury		PCB 153		DHA	
	Cord	Child	Cord	Child	Cord	Child
Mercury						
Cord		.63**	.59**	.36	.15	.13
Child			.51*	.33	.29	.51*
PCB-153						
Cord				.60**	.17	.15
Child					-.20	.13
DHA						
Cord						.22
Child						

Note. n=20 except for cord DHA for which n=17. Analyses were performed with transformed log-values.
*p≤.05; **p≤.01

Table 3

Regression coefficients for Hg, PCB 153 and DHA after adjustment for confounding variables (n=20)

	ERP LATENCY (deviant stimuli, Cz)					
	N1*		P2		P3	
Variables	βstd	Model $R^2_{adjusted}$	βstd	Model $R^2_{adjusted}$	βstd	Model $R^2_{adjusted}$
Cord Hg	-.57†	.25†	-.36	.08	.47*	.25*
Cord DHAb	-.14				-.44§	
Child Hg	-.50*	.33*§	-.38§	.10*	-.39	.01§2
Cord PCB-153	-.24	.12 3	-.27	.02	.24	.01
Child PCB-153	-.22	-.02	-.18	.13 4	.27	.06‡

	ERP LATENCY (deviant stimuli, Cz)					
	N1*		P2		P3	
Variables	βstd	Model $R^2_{adjusted}$	βstd	Model $R^2_{adjusted}$	βstd	Model $R^2_{adjusted}$
Cord Hg	-.08	-.05	.18	.03 2	.003	.10 1
Child Hg	-.08	.11	-.18	.34* &7	.09	-.05
Child DHA	.49 †					
Cord PCB-153	-.35	.09 5	-.05	.30* 6	.00	.07 5
Child PCB-153	-.46	.14 5	.08	.06 &7	.21	.02 5

[1]Model controls for crowding; [2]Model controls for SES; [3]Model controls for alcohol consumption during pregnancy (yes/no); [4]Model controls for child age; [5]Model controls for maternal depression; [6]Model controls for parity; [7] Model controls for child lead levels. *N1 components were only reliable for 17 subjects; [&]Cord DHA concentrations were available for 17 subjects
† $p \leq .10$; * $p \leq .05$.

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