

## THE BIOLOGICAL MONITORING PROGRAM OF PERSISTENT ORGANIC POLLUTANTS IN JAPAN: 1. CONCENTRATIONS OF ORGANOCHLORINE PESTICIDES IN BREAST MILK, CORD BLOOD AND MATERNAL BLOOD

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### Abstract

Persistent organic pollutants (POPs) are ubiquitous environmental contaminants that accumulate in lipid-rich body tissues. Although POPs are thought to be hazardous to health, the overall epidemiological data are as yet insufficient to draw any conclusions. Thus, exposure monitoring and epidemiological examination of the Japanese population are of importance to determine the health risks due to POPs exposure. The Ministry of the Environment of Japan (MOE) has been conducting systematic monitoring of POPs according to the Stockholm Convention. We provided some biological samples for the POPs biological monitoring project, and reanalyzed the report of the MOE. In this presentation, we summarize the data of organochlorine pesticides in human pair samples of breast milk, cord blood and maternal blood. We also analyzed the associations of pesticide concentrations with TSH and thyroid hormones in maternal and cord blood, since disruption of the hypothalamus-pituitary-thyroid axis is a hypothetical mechanism for POPs-induced adverse effects.

### Introduction

Persistent organic pollutants (POPs) are ubiquitous environmental contaminants that accumulate in lipid-rich body tissues. Their lipophilicity and intrinsic resistance to biological degradation processes are responsible for bioaccumulation and biomagnification in the food chain, and consequently they can be found in humans at considerable concentrations. Although these concentrations are usually decreasing and almost at the background level, longer term exposure may cause potential risks to human health.

In humans, some POPs have been claimed to possess endocrine-disrupting potency. DDE exposure is related to TSH and estradiol levels among middle-aged and elderly men.<sup>1</sup> There is a significant negative association between the serum HCB concentration and total T4 in cord blood.<sup>2</sup> These findings suggest that exposure to POPs may affect the hypothalamus-pituitary-thyroid and the hypothalamus-pituitary-gonadal axes. Reproductive defects may be associated in part with exposure to hormonally active environmental chemicals during fetal and childhood development.<sup>3</sup> A growing number of reports have demonstrated the association between adverse effects in children and exposure to POPs at low doses over a longer period. Human perinatal exposure to PCBs has been also shown to be associated with several adverse effects.<sup>4</sup> However, little information is available regarding the delayed neurobehavioral development in infants following exposure to DDE.<sup>5,6</sup> Perinatal exposure to HCB was also shown to be associated with poor social competence in children.<sup>7</sup> Although the overall epidemiological data are not yet sufficient to allow us to draw firm conclusions, exposure monitoring and epidemiological examination of the Japanese population are important for risk assessment.

In Japan, the Ministry of the Environment (MOE) has been conducting systematic monitoring of chemicals over a 30-year period. The MOE initiated refined environmental monitoring including POPs in FY2002 according to the Stockholm Convention.<sup>8,9</sup> Recently, the MOE also added biological monitoring of human samples. Since information on blood levels of POPs in Japan is very limited, this monitoring of all the POPs covered by the convention will contribute to the future effectiveness evaluation. We have been collaborating with the MOE's POPs monitoring project by providing biological samples from our prospective birth cohort study, The Tohoku Study of Child Development (TSCD). We reanalyzed the results from the MOE's monitoring project, and summarize the data of organochlorine pesticides in human pair samples of breast milk, cord blood and maternal blood in this presentation.<sup>10</sup> We also analyzed the associations of pesticide concentrations with TSH and thyroid hormones in maternal and cord blood, since the disruption of the hypothalamus-pituitary-

thyroid axis is a hypothetical mechanism for POPs-induced adverse effects.

### Materials and Methods

Biological samples analyzed were randomly selected from the participants in the TSCD, and provided anonymously to the MOE. The TSCD study protocol was previously reported.<sup>3</sup> Briefly, maternal peripheral blood was collected using heparin as the anticoagulant agent in the morning when the pregnancy was at 28 weeks. The cord blood was collected immediately after delivery. These whole blood samples were frozen at  $-80^{\circ}\text{C}$  until the chemical analysis. Breast milk was collected one month after delivery, and then frozen similarly.

Chemical determination was performed with high resolution gas chromatography-high resolution mass spectrometry (HRGC-HRMS) by IDEA Consultants, Inc. (Tokyo, Japan) as part of the MOE project as described in another report in this book. TSH, total T4 and total T3 were measured from the plasma of cord and maternal blood by SRL, Inc. (Tokyo, Japan). The statistical analyses were performed using JMP ver. 5.1.2. When the levels of data were not normally distributed, the data were log-transformed for statistical analysis.

The TSCD was approved by the Medical Ethics Committee of the Tohoku University Graduate School of Medicine, and all mothers provided signed informed consent.

Table 1 Pesticide concentrations in breast milk, cord blood and maternal blood (pg/g-fat)

Chemicals	Breast milk Median (Min-Max)	Cord blood Median (Min-Max)	Maternal blood Median (Min-Max)
Aldrin	nd (nd)	nd (nd)	nd (nd)
<i>cis</i> -Chlordane	460 (200-3100)	440 (210-1460)	620 (220-2060)
<i>trans</i> -Chlordane	180 (80-1400)	330 (120-770)	190 (130-490)
Oxychlordane	11700 (2700-46800)	4940 (1280-17530)	5520 (1540-17270)
<i>cis</i> -Nonachlor	3400 (860-10570)	960 (280-2780)	1680 (470-4860)
<i>trans</i> -Nonachlor	22480 (6620-100950)	6690 (1690-26260)	12830 (3620-52370)
<i>o,p'</i> -DDD	nd (nd-510)	nd (nd-100)	nd (nd-100)
<i>p,p'</i> -DDD	300 (100-14510)	120 (nd-590)	240 (60-430)
<i>o,p</i> -DDE	380 (180-950)	250 (90-600)	340 (170-730)
<i>p,p'</i> -DDE	143300 (31700-331500)	68180 (12330-385690)	93270 (17280-271390)
<i>o,p</i> -DDT	1220 (550-4170)	450 (190-1420)	700 (200-2130)
<i>p,p'</i> -DDT	7620 (2310-19390)	2450 (560-7330)	3950 (1080-10070)
Dieldrin	4290 (2100-17480)	3140 (1370-13580)	3280 (1440-9810)
Endrin	nd (nd-490)	nd (nd)	nd (nd)
Heptachlor	nd (nd-370)	nd (nd-170)	nd (nd)
<i>trans</i> -Heptachlorepoxyde	nd (nd)	nd (nd)	nd (nd)
<i>cis</i> -Heptachlorepoxyde	4480 (1800-24140)	2490 (670-12720)	2680 (730-12520)
HCB	16380 (6870-37260)	16700 (6440-39980)	13810 (5560-39580)
$\alpha$ -HCH	290 (150-1570)	310 (130-1910)	220 (120-580)
$\beta$ -HCH	49010 (11500-213990)	29030 (4860-90490)	29350 (4750-196100)
$\gamma$ -HCH	220 (50-2340)	340 (150-5080)	220 (100-2180)
$\delta$ -HCH	nd (nd-310)	nd (nd-140)	nd (nd)
Mirex	740 (170-1880)	410 (120-1380)	1110 (280-2890)
Parlar-26	1880 (760-7040)	660 (230-3020)	960 (300-2550)
Parlar-40	20 (nd-100)	nd (nd-180)	nd (nd-70)
Parlar-41	230 (nd-560)	nd (nd-240)	110 (nd-220)
Parlar-44	230 (60-640)	nd (nd-380)	70 (nd-200)
Parlar-50	3150 (1280-12490)	850 (280-4140)	1440 (480-4220)
Parlar-62	230 (nd-820)	nd (nd-510)	40 (nd-360)

n=68 for breast milk and cord blood, n=49 for maternal blood.

Table 2. Correlation of pesticide concentrations among breast milk, cord blood and maternal blood

	Breast milk						
	<i>c</i> -CHL	<i>t</i> -CHL	OxyCHL	<i>c</i> -Nonachlor	<i>t</i> -Nonachlor	<i>p,p'</i> -DDE	<i>p,p'</i> -DDT
Cord blood	0.543**	0.191	0.831**	0.836**	0.871**	0.837**	0.796**
Maternal blood	0.729**	0.291	0.943**	0.954**	0.959**	0.920**	0.878**

  

	Breast milk						
	Dieldrin	<i>c</i> -HCE	HCB	$\beta$ -HCH	Mirex	Parlar-26	Parlar-50
Cord blood	0.821**	0.800**	0.879**	0.800**	0.673**	0.789**	0.778**
Maternal blood	0.819**	0.928**	0.921**	0.844**	0.894**	0.904**	0.917**

Pearson's *r* after log-transformed. *n*=68 for breast milk and cord blood, and *n*=49 for breast milk and maternal blood.

\*\* *p*<0.001

### Results and Discussion

Concentrations of pesticides in breast milk, cord blood and maternal blood are shown in Table 1. The highest values were observed for *p,p'*-DDE in all three materials. Since the use of DDT was prohibited in 1971 in Japan, this finding indicates the nature of intrinsic resistance to biological degradation of DDT/DDE. Mirex and toxaphene were measurable in most samples. Since neither chemical has ever been used in Japan, the route and the source of contamination are not fully understood.

High correlations of most chemicals among the three materials were observed as shown in Table 2. These findings indicated the usefulness of breast milk as a monitoring material for human exposure. Breast milk is rich in fat and therefore lipophilic chemicals such as POPs are accumulated, and it can be easily obtained from lactating women. In breast milk, most chemicals also correlate with each other (data not shown).

Since the working hypothesis on POPs-induced adverse effects is the disruption of the hypothalamus-pituitary-thyroid axis, associations of pesticides with TSH, T4 and T3 in maternal blood and cord blood were analyzed as shown in Table 3. It was confirmed that dioxins, PCBs and DDT/DDE were associated with TSH and thyroid hormones. In addition, other minor pesticides such as HCB, nonachlor, and toxaphenes were also associated with TSH, T4 and T3. Since there were clear multicollinearities among the chemicals, the causal relationships between the pesticide exposures and the levels of TSH and thyroid hormone remain to be clarified.

POPs exhibit bioaccumulation and biomagnification in the food chain, and therefore human exposure is thought to be mainly through the consumption of fish. However, maternal fish intake did not correlate with the concentrations of any of the chemicals (data not shown).

Further monitoring assessments and epidemiological examinations will make it possible to understand the exposure characteristics and biological effects of POPs on humans.

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Table 3. Relationship of POPs with maternal age, gestational age and concentrations of TSH and thyroid hormones

	Maternal age	Gestational age	TSH and thyroid hormones					
			Maternal blood			Cord blood		
			TSH	T4	T3	TSH	T4	T3
<b>Breast milk</b> (n=68, except for n=67 for maternal TSH/T4/T3)								
TEQ	ns	ns	0.232 <sup>§</sup>	ns	0.225 <sup>§</sup>	ns	-0.224 <sup>§</sup>	ns
PCB	ns	ns	0.205 <sup>§</sup>	0.231 <sup>§</sup>	0.257*	ns	ns	ns
<i>trans</i> -Nonachlor	ns	ns	ns	ns	0.281*	ns	-0.238 <sup>§</sup>	ns
<i>p,p'</i> -DDE	ns	ns	ns	ns	ns	ns	ns	ns
<i>p,p'</i> -DDT	ns	ns	ns	ns	ns	ns	ns	ns
Dieldrin	0.229 <sup>§</sup>	-0.265*	ns	ns	0.235 <sup>§</sup>	ns	ns	ns
<i>cis</i> -Heptachlorepoxyde	ns	ns	ns	ns	ns	0.243*	-0.212 <sup>§</sup>	ns
HCB	ns	ns	ns	ns	ns	ns	-0.228 <sup>§</sup>	ns
$\beta$ -HCH	ns	ns	ns	ns	ns	ns	ns	ns
Mirex	0.422**	-0.232 <sup>§</sup>	0.234 <sup>§</sup>	ns	ns	0.233 <sup>§</sup>	ns	ns
Parlar-26	ns	ns	ns	0.291*	0.395**	ns	ns	ns
Parlar-50	ns	ns	ns	0.287*	0.381**	ns	ns	ns
<b>Maternal blood</b> (n=49, except for n=48 for maternal TSH/T4/T3)								
TEQ	ns	ns	0.264 <sup>§</sup>	0.272 <sup>§</sup>	0.375**	ns	ns	ns
PCB	ns	ns	0.277 <sup>§</sup>	0.287*	0.402**	ns	ns	ns
<i>trans</i> -Nonachlor	ns	ns	0.274 <sup>§</sup>	ns	0.429**	ns	-0.243 <sup>§</sup>	ns
<i>p,p'</i> -DDE	ns	ns	0.242 <sup>§</sup>	ns	0.252 <sup>§</sup>	ns	ns	-0.251 <sup>§</sup>
<i>p,p'</i> -DDT	ns	ns	ns	0.299*	0.426**	ns	-0.279 <sup>§</sup>	ns
Dieldrin	0.351*	ns	ns	ns	0.247 <sup>§</sup>	ns	ns	ns
<i>cis</i> -Heptachlorepoxyde	ns	ns	ns	ns	ns	ns	ns	ns
HCB	ns	ns	0.251 <sup>§</sup>	0.310*	0.391**	ns	ns	ns
$\beta$ -HCH	ns	ns	ns	ns	0.267 <sup>§</sup>	ns	ns	ns
Mirex	0.391**	-0.251 <sup>§</sup>	0.342*	ns	0.324*	0.284*	ns	ns
Parlar-26	ns	ns	ns	0.267 <sup>§</sup>	0.468**	0.261 <sup>§</sup>	ns	ns
Parlar-50	ns	ns	ns	0.296*	0.500**	0.269 <sup>§</sup>	ns	ns
<b>Cord blood</b> (n=68, except for n=67 for maternal TSH/T4/T3)								
TEQ	ns	ns	0.203 <sup>§</sup>	ns	ns	ns	-0.270*	ns
PCB	ns	ns	0.239 <sup>§</sup>	ns	0.240 <sup>§</sup>	ns	ns	ns
<i>trans</i> -Nonachlor	ns	ns	0.238 <sup>§</sup>	ns	0.244*	ns	-0.217 <sup>§</sup>	ns
<i>p,p'</i> -DDE	ns	ns	ns	ns	ns	ns	-0.208 <sup>§</sup>	ns
<i>p,p'</i> -DDT	ns	ns	0.206 <sup>§</sup>	ns	ns	ns	-0.238 <sup>§</sup>	ns
Dieldrin	ns	-0.237 <sup>§</sup>	ns	ns	ns	ns	-0.302*	-0.260*
<i>cis</i> -Heptachlorepoxyde	ns	-0.278*	ns	ns	ns	0.262*	-0.335**	-0.208 <sup>§</sup>
HCB	ns	ns	ns	ns	ns	0.202 <sup>§</sup>	-0.293*	ns
$\beta$ -HCH	0.206 <sup>§</sup>	ns	ns	ns	ns	ns	ns	ns
Mirex	0.294*	-0.239 <sup>§</sup>	0.234 <sup>§</sup>	ns	ns	ns	ns	ns
Parlar-26	ns	-0.237 <sup>§</sup>	ns	ns	0.300*	ns	ns	ns
Parlar-50	ns	-0.220 <sup>§</sup>	ns	ns	0.324**	ns	ns	ns

Pearson's r after log-transformed. <sup>§</sup> p<0.1, \* p<0.05, \*\* p<0.01