DIOXINS, PCBs AND ORGANOCHLORINE PESTICIDES IN HUMAN BREAST MILK FROM MALAYSIA

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Introduction

Contaminations by persistent organic pollutants (POPs) such as polychlorinated dibenzo-*p*-dioxins (PCDDs), dibenzofurans (PCDFs), polychlorinated biphenyls (PCBs) and organochlorine pesticides in the environment have been of great concern due to their endocrine disrupting effects on humans and wildlife. Chemically stable and lipophilic properties of these contaminants led to their high contamination in higher trophic biota, including human. Despite the intensive monitoring efforts and anticipated results of decreasing trends of POPs in developed countries as a consequence of their regulation on use and waste treatment, little information are available on their contamination status in developing countries even though these chemicals are still being used and unintentionally produced in several parts of these countries. To ensure the reliability of exposure data and to delineate contamination status, fate and behavior in tropical developing countries, during last few years, our research groups conducted monitoring studies using various environmental matrices including air, water, sediment, soil, biota and human from several Asian developing countries [1-6]. From these results, existing sources of OCs and formation of dioxins and related compounds could be predicted in this region [4-7]. However, there is very little information addressing the accumulation of OCs pollution in Malaysia. Particularly available data are only on marine biota [2]. To date no data are available on OCs contaminations in human milk samples from Malaysia. The present study aims at understanding recent contamination of POPs, including dioxins and related compounds, PCBs and OCs pesticides in human breast milk from the general population of Malaysia.

Materials and Methods

Samples: Breast milk samples were collected from primiparas mothers from Penang and Kedah, Malaysia during 2003. Samples were placed in analytical-grade glass containers with Teflon-lined caps, frozen immediately and kept at –20^{o}C in the laboratory until chemical analysis. Nine samples were used for dioxins and related compounds analysis, and 17 samples for PCBs and OCs pesticides analysis. The donors completed informed consent form and an exposure assessment

questionnaire following WHO protocols. The range of mothers' age is 24-35 years with body mass index (BMI) between 19.0-22.2 kg/m^2. None of the mothers was a smoker.

Chemical analysis: PCDDs, PCDFs, PCBs and OCs pesticides were analyzed based on the method published elsewhere 3). Identification and quantification of PCDDs, PCDFs, non- and mono-ortho PCBs were performed using high resolution gas chromatography-high resolution mass spectrometry (HRGC-HRMS). PCBs, DDTs, HCHs, CHL compounds, and TCPMe were determined by gas chromatograph with an electron capture detector (GC-ECD) and mass spectrometry (GC-MS) (Fig. 4) 3). TEQs were estimated based on human/mammals-TEFs proposed by WHO 7).

Statistical analysis: Statistical analyses were carried out using Stat View program version 5 (SAS Inc., 1998). Spearman's rank correlation coefficient was used to measure the strength of the association between the mother's age, breast-feeding periods and OCs concentrations, and between concentrations of DDTs and TCPMe in human breast milk. A p value of <0.05 values were considered significant.

Results and discussion

Persistent organic pollutants (POPs) were detected in all the Malaysian milk analyzed in this study (Table 1). To our knowledge, this is the first report on the accumulation of dioxins, dioxin-related compounds, PCBs and OCs pesticides in human milk from Malaysia.

Table 1: Concentration of dioxins, dioxin-related compounds (pg/g lipid wt.), PCBs and OCs pesticides (ng/g lipid wt.) in breast milk from primiparas mothers in Malaysia.

	Age	Fat	Dioxins and Related Compounds (pg/g lipid wt.)*					PCBs and OCs Pesticides (ng/g lipid wt.)**					
	(Years)	(%)	PCDDs	PCDFs	non-*ortho* PCBs	mono-*ortho* PCBs	TEQs	PCBs	DDTs	HCHs	CHLs	HCB	TCPMe
Mean	30	1.7	130	12	62	11000	13	80	1600	230	23	11	8.1
Median	29	1.4	150	13	57	7500	14	60	740	140	21	5.2	5.3
Range	23-38	0.32-4.0	34-200	3.3-20	24-100	1200-41000	3.0-24	23-450	180-5700	27-1000	8.2-54	1.3-78	1.1-38

*9 samples; **17 samples

Dioxins and related compounds: Among dioxins and dioxins-like compounds, mono-*ortho* PCBs were predominantly detected in all the samples in an average of 11000 pg/g lipid wt., followed by PCDDs (130 pg/g lipid wt.), non-*ortho* PCBs (62 pg/g lipid wt.), PCDFs (12 pg/g lipid wt.) and TEQ levels (13 pg TEQs/g lipid wt.), respectively. The abundance of mono-*ortho* PCBs in this study may be due to exposure from technical PCBs [8]. In fact, one donor who accumulated the highest levels of dioxins had higher mono-*ortho* PCBs and total PCBs in breast milk, which supports the above contention. Other sources of dioxin could also be the combustion processes. Recently, our research group reported the formation of dioxins by combustion of waste in municipal dumping sites in Malaysia as exemplified by the extremely high levels of dioxins in the soil of this site [6], is similar to other dumping sites of developing countries [5]. In this study, PCDDs/Fs-TEQs contributed 66% to the total TEQs levels while the remaining was by coplanar-PCBs. Although, TEQs levels in human breast milk from Malaysia (13 pg TEQs/g lipid wt.) were

much lower than those of developed nations and former Soviet countries, and only slightly higher within developing countries (Fig. 1) [3], one donor mother accumulated relatively high level of TEQs (24 pg TEQs/g lipid wt.) comparable to the mean value of Japanese (22 pg TEQs/g lipid wt.). This implies that some of the residents Malaysia may be exposed to specific pollution source of dioxins and related compounds. Despite the decreasing trend of dioxin contamination in developed nations as a consequence of installation of highly efficient incinerators and strict regulation on chemical production and usage, formation of dioxins and related compounds in open dumping sites in Asian developing countries, including Malaysia may increase raising a considerable human health concern in future. In fact, large scale dumping of municipal waste in developing countries are quite common and the continuous burning under low temperature by spontaneous combustion or intentional incineration, of wastes may produce dioxins and related compounds [5, 6].

PCBs and OCs pesticides: With regard to other OCs, DDTs and HCHs were the predominant contaminants ranging up to 5700 ng/g lipid wt. and 1000 ng/g lipid wt., respectively (Table 1), whereas, PCBs, CHLs, HCB, and TCPMe were detected with concentrations one to two orders of magnitude less than DDTs and HCHs. The pattern of OCs in human breast milk observed in this study (DDTs>HCHs>PCBs>CHLs>HCB) was different from that in mussels, which showed higher levels of CHLs and DDTs compared to PCBs and HCHs (CHLs>DDTs>PCBs>HCHs>HCB) [2]. This is because of the distribution and fate of persistent OCs may be different in terrestrial and marine environment, resulting in differences in OCs contamination in both spatial and temporal terms [9]. Furthermore, a newly identified endocrine disrupting chemical, TCPMe was also detected in all the samples analyzed in this study, ranging from 1.1 - 38 ng/g lipid wt., indicating widespread contamination by this micro contaminant in human from Malaysia. A significant positive correlation was also observed between concentrations of TCPMe and DDTs ($p<0.05$, data not shown) similar to several other studies [3, 4], hence supporting the hypothesis that the sources of TCPMe could be technical DDTs [10].

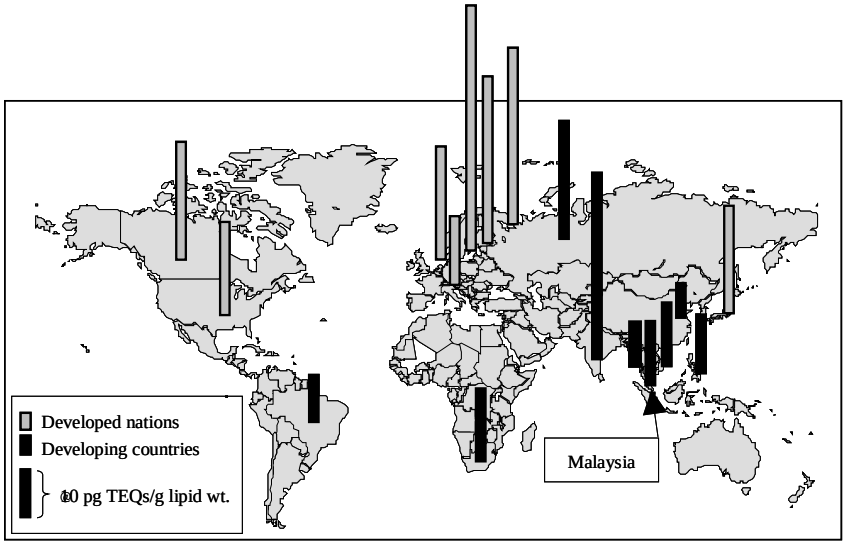

Figure 1: Comparison of TEQ levels in human breast milk all over the world.

Table 2: Comparison of PCBs and OC pesticides levels (ng/g lipid wt) in human breast milk over the world.

Country	Year	*n*	PCBs	DDTs	CHLs	HCHs	HCB	Reference
Germany	1995-1997	246	550	240	-	40	80	See Minh *et al.* [5]
Sweden	1997	40	324	143	-	-	12	See Minh *et al.* [5]
UK	1997-1998	168	-	470	-	103	43	See Minh *et al.* [5]
Canada	1992	497	238	244	34	24	15	See Minh *et al.* [5]
Australia	1995	60	500	1200	-	350	-	See Minh *et al.* [5]
Japan	1998	49	200	290	85	210	14	See Minh *et al.* [5]
Russia	1996-1997	140	380	1040	37	280	91	See Minh *et al.* [5]
Ukraine	1993-1994	197	594	2700	38	730	168	See Minh *et al.* [5]
India	2000	8	30	420	0.9	650	1.0	See Minh *et al.* [5]
China	1999-2000	186	37	3200	-	1000	-	See Minh *et al.* [5]
Cambodia	2000	49	25	1500	1.6	5.2	1.7	See Minh *et al.* [5]
Philippines	2000	10	72	190	15	4.7	-	See Minh *et al.* [5]
Vietnam	2000-2001	48	76	2200	4.5	36	3.2	See Minh *et al.* [5]
Malaysia	2003	17	80	1600	23	230	11	This study

Residue levels of OCs, particularly DDTs in breast milk were higher in developing and former Soviet countries, including Malaysia than those in developed countries, indicating their usage until very recent years for both agricultural and human health purposes. Interestingly, during worldwide comparison, HCHs in human breast milk from Malaysia were in the higher range (Table 2). Further, chlordane compounds in this study were also highest among Asian developing and Soviet countries. This fact suggests existing sources of pollution by these persistent OC pesticides in the general population of Malaysia. Although, PCBs levels in breast milk from Malaysia were slightly

higher within developing countries, their residues were still lower than those in developed countries, suggesting less contamination by PCBs in general population of developing countries, including Malaysia. Considering all these facts, further research should be conducted to elucidate the temporal trend and potential risk to the humans.

Accumulation kinetics: No significant correlation was observed between OCs levels in breast milk and mother's age (Figure 2). This is in contrast to various primiparas study that levels of OCs in breast milk were positively correlated with mother's age [4, 11]. The possible reason for this may be limited number of sample for dioxins and related compounds (n=9), and for other OCs (n=17), narrow range of age of the donor mothers (23-38 years) and or external factors. Quinsey *et al.* [12] also found no significant correlation between OCs concentrations and age of primiparas mothers when the sample number was small. Furthermore, there is no correlation between OCs levels in breast milk and frequency of fish and meat consumption (data not shown). For examples, frequency of fish and meat consumption of the mother who had the highest level of dioxins and related compounds in this study was smaller than other mothers (data not shown). These results imply that other foodstuffs may also be responsible for exposure of the general population of this study. Further study using large number of samples and various foodstuffs are needed to clarify the variation of human exposure, accumulation levels and exposure sources of OCs in Malaysia.

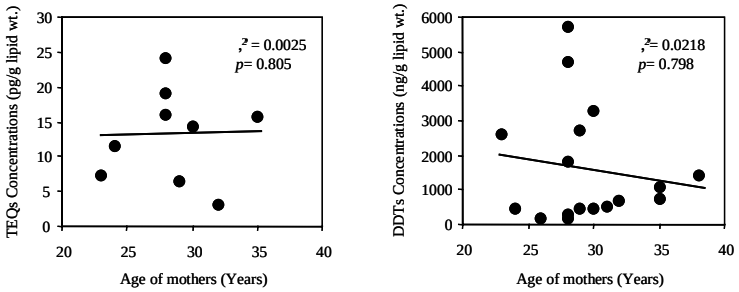

Figure 2: Relationships between concentrations of TEQs and DDTs in human breast milk and age of primiparas mothers in Malaysia.

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