Levels of PCBs and Selected Organochlorine Pesticides in Humans from Selected Areas of the Slovak Republic. Part III. Milk

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1. Introduction

Polychlorinated biphenyls (PCBs) were produced from 1959 to 1984 in the Michalovce District in eastern Slovakia. They were used intensively in Slovakia as heat exchanger liquids and also as ingredients in paint. PCB congeners are being found in significant amounts in all environmental compartments of this country, including food and human tissues[1-6]. Although relatively strict PCB limits for food use were not normally overstepped, PCB levels in humans have increased due to their uninterrupted intake and low biodegradation. A similar situation has occurred with organochlorine pesticides such as DDT and lindane, both of which were banned in Slovakia in the 70's. Surprisingly high concentrations of hexachlorobenzene (HCB) found in human samples from Slovakia are probably caused by its use in agriculture and its formation during industrial manufacture of some chlorinated solvents.
As already done in the cases of human adipose tissue and blood samples, human milk samples were collected in the same five model areas (potential sources are shown in parentheses); i.e., the *Michalovce* District (PCB production at the Chemko Strážské), the *Veľký Krtíš* District (increased contamination of dairy products due to PCB content in paint used for coatings of silage silos), the town of *Bratislava* (chemical industry, dense automobile traffic, municipal, industrial and hospital waste incineration), the *Nitra* District (a prevailingly agricultural region), the *Myjava* region (a rural highland region characterized by home-made food, including cow's milk, butter, eggs, pork, etc.).
At the DIOXIN '93 Symposium in Vienna and DIOXIN '94 Symposium in Kyoto as a part I and II of this study[5,6], levels of PCB congeners, HCB, p,p'-DDE and p,p'-DDT were reported in blood and adipose tissue samples from the human population living in the above-mentioned selected areas .

2. Experimental

According to written detailed instructions based on the WHO protocol, we collected from breast-feeding mothers (primiparae, milk sampling 2 weeks-2 months after delivery, unchanged residence at least 5 years before pregnancy) collected over the course of several

days about 100 ml of milk into a glass vial with a PTFE cap. These were collected by manual expression or by means of a manual breast pump. The vials were kept in a freezer at -30°C until analysis. Individual questionnaires were completed in the presence of a physician or a nurse to obtain information on age, body height and weight both of the mother and her newborn, dietary habits, previous and current occupation, smoking habits, and domicile. Selected questionnaire data summarized for the individual areas are given in Table 1.

Table 1. Some data on human breast milk donors

Area		Bratislava	Myjava	Nitra	Michalovce	V.Krtíš
No. of Donors		13	5	9	12	11
Age [years]	Mean	24.6	21.8	24.1	20.7	21.5
	Range	20-28	19-29	21-30	17-26	18-25
Number of	Smokers	2	0	0	2	2
	Ex-smokers	1	1	2	2	0
	Nonsmokers	10	4	7	8	9
Diet	Mixed	13	5	8	11	11
	Vegetarian	0	0	1	1	0
Domicile	Urban	12	2	5	8	8
	Rural	1	3	4	3	3
BMI [$kg.m^{-2}$]	Beginning of Pregnancy	19.5	22.3	21.2	19.6	19.8
	Just Before Parturition	23.8	27.3	26.8	24.1	24.7
Mean Fat Content in Milk [%]		2.94	3.33	2.32	3.56	3.67

Milk samples were homogenized ultrasonically, treated with a sodium oxalate solution, and extracted several times with an n-hexane:diethylether mixture (1:1). After evaporating the solvents, the lipids were obtained gravimetrically. A 50 mg portion was cleaned-up by a Florisil-H_2SO_4/silica-Florisil column[5]. Nine PCB congeners (IUPAC Nos. 28, 52, 101, 138, 153, 180, 105, 118, and 156) and three organochlorine pesticides (HCB, p,p'-DDE and p,p'-DDT) were determined using HRGC/ECD. A DB-5 column (60 m x 0.25 mm, 0.25 µm film thickness) was used for the separation of the PCB congeners and pesticides. Together with the unknown milk lipid samples, a blank, a recovery sample (plant oil spiked with PCBs and organochlorine pesticides) and CRM's sample were analyzed. HRGC/ECD chromatograms were evaluated by calibration curves of standard mixtures of the analytes in n-heptane. The results were corrected for recovery. The analyzing laboratory works in compliance with the GLP principles and successfully participated in proficiency testing aimed at organochlorine pesticides and PCBs (GEMS/Food-EURO, UK; Deutsche Gesellschaft für Arbeitsmedizin und Umweltmedizin, F.R.G.)

3. Conclusions

- As observed in the human adipose tissue[6] and blood serum[5] samples, HCB and p,p'-DDE were present in the milk samples at higher concentrations than individual PCB congeners and p,p'-DDT (Fig. 1 and 2). HCB levels are noteworthy as they are one or two orders of magnitude higher than reported from other countries[7,8,10-24].

- PCB, DDE, DDT and HCB levels determined in the Slovak human milk are compared with reported foreign data in Table 2.

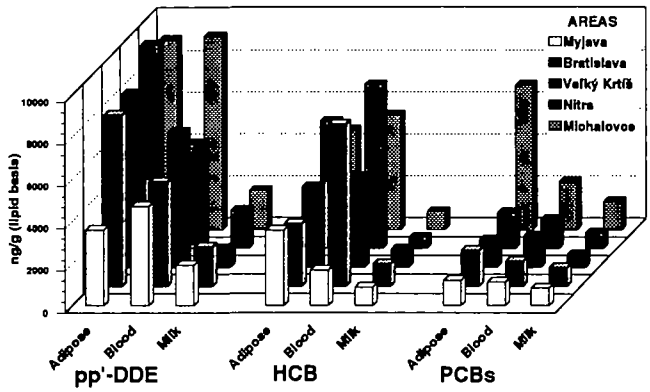

Fig. 1. Comparison of p,p'-DDE, HCB and summed PCB congener (IUPAC Nos. 28, 52, 101, 138, 153, 180, 105, 118, and 156) levels in human adipose tissue[6], blood serum[5] and milk samples from the human general population of the five model areas of Slovakia

**) this study*

- Levels of all the pollutants determined in the human milk lipids were substantially lower than those in the blood and adipose tissue samples collected from the same areas (Fig. 1). The differences in PCB levels in the milk samples from the Michalovce District compared to the other districts were not as large as the adipose tissue and blood serum differences (Fig. 2). This could be caused by the lower age of breast-feeding mothers in comparison with the mean ages of the adipose tissue and blood donors and by decreasing organochlorine pesticide and PCB contamination of the environment after their use had been banned.

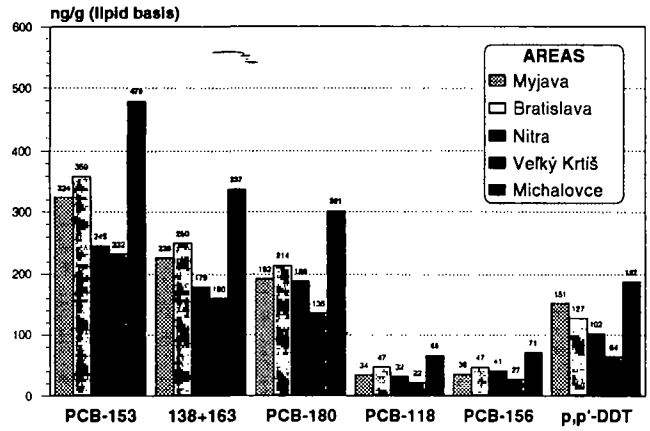

Fig. 2. Average levels of some PCB congeners and p,p'-DDT milk lipids from the human general population of the five model areas of Slovakia

- The average ratios of p,p'-DDE to p,p'-DDT in the milk samples from Bratislava, Myjava, Nitra, Michalovce and Veľký Krtíš were 14.7, 12.6, 17.5, 9.8 and 13.9, respectively. This is also less than observed in the adipose tissue and blood serum samples. Congeners IUPAC No. 153, 180 and 138 were the most abundant PCB congeners found in all of the samples (Fig. 2).
- Considering a 5 kg infant sucking daily 800 g of mother's milk in the Michalovce District, the average daily intake of the sum of the six PCB congeners was 7.5 $\mu g.kg_{b.w.}^{-1}.day^{-1}$. The

Table 2. PCB, HCB, p,p'-DDE and p,p'-DDT levels found in human milk lipids from various countries

Year of Sample Collection	No. of Samples	Country	Average Concentration [μg.kg⁻¹, fat basis]					Ref.
			6-PCB[1]	PCB-118	HCB	pp'-DDE	pp'-DDT	
1986	412	Canada[2]	1042	20	-	-	-	10
1991	16	Canada, Quebec[2]	109	17	-	-	-	17
1993	107	Canada, Quebec, Inuits	706	-	136	1212	-	12
1991	32	United Kingdom	302	29	-	-	-	16
1990/1991	57	United Kingdom, Wales	232	18	-	-	-	13
1991	28	Norway, Oslo[2]	261	26	-	-	-	8
1990/1992	195	Netherlands, Rotterdam	439	35.5	-	-	-	23
1993	24	Jordan, Amman	351	-	-	-	-	15
1990/1991	68	Germany, Middle-Hesse	692	-	-	-	-	22
1991	113	Germany, N.Rhine Westphalia[2]	581	-	177	504	27	24
1989/1990	59	Jordan, Amman	-	-	290	2040	450	14
1991	51	Spain, Madrid	-	-	0.8	604	12.5	11
1985	100	Israel, Jeruzalem	-	-	80	2440	290	18
1979/1980	54	USA, Hawaii	-	-	46	1989	162	19
1981/1982	50	Finland	-	-	64	850	36	21
1987	64	Italy	271	41	192	2050	154	7
1990	25	Czech Rep., Jihlava	-	-	1644	-	-	20
1993/1994	50	Slovakia[3]	785	39	829	1667	126	

[1] The sum of IUPAC Nos. 28, 52, 101, 138, 153 and 180

- not reported

[2] Not all 6 congeners were measured (mostly 138, 153 and 180)

[3] Present study (arithmetic mean values calculated from mean levels determined in the blood samples collected in the five model areas)

range could be from 2.8 to 4.4 $\mu g.kg_{b.w.}^{-1}.day^{-1}$ in the other four model areas, which is substantially higher than the acceptable or tolerable daily intake of 1 $\mu g.kg_{b.w.}^{-1}.day^{-1}$ (involves all PCB congeners) established in some countries[25]. Concerning HCB and p,p'-DDE+p,p'-DDT, the daily would range in all the 5 areas from 2.0 to 5.1 $\mu g.kg_{b.w.}^{-1}.day^{-1}$ and from 5.6 to 11.5 $\mu g.kg_{b.w.}^{-1}.day^{-1}$, respectively. In the case of DDE+DDT, these intakes are lower than the ADI established by WHO (20 $\mu g.kg_{b.w.}^{-1}.day^{-1}$), but in the case of HCB these intakes are two orders of magnitude higher than the provisional tolerable daily intake of 0.08 $\mu g.kg_{b.w.}^{-1}.day^{-1}$ according to the Health Protection Branch of Health and Welfare Canada[9].

- The presented results from the pilot study involve a relatively small number of specimens. Therefore, it is questionable to evaluate any statistical parameters. The goal of the study is to obtain introductory information on the occurrence of some persistent organochlorines in the Slovak general human population. Based on the pilot data, a case study aimed at areas with increased levels of the organochlorine compounds in the Slovak human population should start in the near future.

This publication is partly based on work sponsored by the U.S. - Slovak Science and Technology Joint Fund in cooperation with the Ministry of Health of the Slovak Republic and the U.S. Department of Health and Human Services under Project Number 94023.

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