CONFERENCE PAPER

PERSISTENT, BIOACCUMULATIVE, AND TOXIC COMPOUNDS IN THE CENTRAL AND EASTERN EUROPEAN COUNTRIES – THE-STATE-OF-THE-ART REPORT – HUMAN EXPOSURE

IVAN HOLOUBEK, ANTON KOČAN, IRENA HOLOUBKOVA, KLARA HILSCHEROVA, JIRI KOHOUTEK, JERZY FALANDYSZ, AND OTT ROOTS

Recetox-Tocoen & Associates, Brno, Czech Republic

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This review describes problems with persistent and bioaccumulative organic substances which posses toxic characteristics likely to cause adverse human health or environmental effects in countries of Central and Eastern Europe as far as human exposure is concerned. This paper is a part of a more detailed report on the subject.

Key words: exposure, persistent organic pollutants

Organic substances that are persistent, bioaccumulative, and posses toxic characteristics likely to cause adverse human health or environmental effects are called PBTs (persistent, bioaccumulative, and toxic substances). In this context, »substance« means a single chemical species, or a number of chemical species, which form a specific group by virtue of (a) having similar properties and being emitted together into the environment or (b) forming a mixture normally marketed as a single product. Depending on their mobility in the environment, PBTs could be of local, regional or global concern (1).

The region of the Central and Eastern Europe is in this Report located in the area from the Baltic Sea to the south of Adriatic Sea and from the Czech Republic to Baltic

countries. All other countries from former Soviet Union are not included in the report for the lack of information.

In general, there is little data on levels of PBT compounds in many countries of the Central and Eastern Europe (CEE). The situation is better with data on the industrial and pesticide chemicals from Croatia, Czech Republic, Poland, and Slovakia. Other countries such as Bulgaria, Hungary, and Slovenia produce satisfactory information about pesticide contamination. The rest of the CEE countries is able to produce only limited data on PBT sources and levels (2).

Elevated levels of organochlorines compared to other European countries indicated high exposure of young women in the former Czechoslovakia. The first survey in the field of PBT compounds carried out in the former Czechoslovakia in the early 60s was initiated by concerns over serious contamination of humans caused by the widespread use of DDT. Besides the measurement of DDTs in the human adipose tissue, later studies also paid attention to hexachlorobenzene (HCB), hexachlorocyclohexanes isomers (HCHs), and polychlorinated biphenyls (PCBs) in the 80s. A gradual decrease in concentrations of DDT, HCHs, and HCB in human adipose tissue was observed upon their ban.

However, considerable differences in residue levels determined in the human fat from different part of the Czech Republic have been frequently found. Moreover, in some regions, the otherwise general decrease is not so pronounced due to serious contamination of the locally produced and distributed foodstuffs. A very similar situation has been found in other CEE countries.

This region has very specific problems related to environmental pollution, which are the results of the recent wars. The destruction of industrial facilities and spilling of chemicals have had the worst effect for the environment in Bosnia and Herzegovina, Croatia, Serbia, and Montenegro. A potential result of food, water, and environment pollution is the dramatic increase in the digestive system carcinoma, particularly of the large intestine which has been observed in Bosnia and Herzegovina for last two years. Below, we will describe levels of various PBTs in human samples from CEE countries.

HUMAN EXPOSURE TO CHLORINATED PESTICIDES AND POLYCHLORINATED BIPHENYLS

Former Czechoslovakia, Czech Republic

Serious contamination of food chains by PCBs occurred in the past in the Czech and Slovak Republics. The main source of local dietary exposure were meat, milk, dairy products, and eggs produced by farms, silos, sheds, and other agricultural facilities using paint with the PCB content. The composition of a commercial technical mixture Delor 106 that was contained in these paints corresponds to Aroclor 1254 (3).

The monitoring programme in the former Czechoslovakia established in mid 80s revealed remarkably high levels of PCBs in human milk. However, the content of contaminants used to be expressed at that time as »total PCBs«. It should be noted that the comparability of generated data was rather poor, reflecting different »quantification strategies« applied by individual laboratories. The implementation of the con-

gener-specific method at the beginning of the 90s made it possible to get more information from various programmes about the PCB patterns (4).

Kočan and co-workers published a comparison between levels of PCBs and HCB found in human adipose tissue and blood samples from the former Czechoslovakia and other countries (5, 6). Unfortunately, the comparison between the present analytical results and older data is questionable because of different analytical performance, such as total PCB v. congener-specific analysis, packed column v. capillary column separation, and lower v. higher precision and accuracy. The importance of PCB analysis has increased after reports of dioxin-like toxicity of planar PCB congeners.

In many papers published in the last 15 years levels of lipophilic analytes in human blood samples are reported on a whole blood or a blood serum basis, but not on a lipid basis. This may be due, in part, to problems encountered in the quantitative isolation of blood serum lipids. However, many published procedures for the analysis of halogenated aromatics giving the results on a blood basis are actually also based on lipid isolation, but the isolated lipids are not weighed. Unfortunately, this approach makes the comparison between the levels of lipids from adipose tissue, blood, and milk impossible, although a high correlation has been observed between them.

PCBs and other organochlorinated compounds (OCCs) are most often monitored in human fat and human adipose tissue, but human serum or other tissues (placenta, liver, and heart) can also be analysed (7). The results concerning human biomonitoring of PCBs in the former Czechoslovakia and in the Czech and Slovak Republics published over the last two decades were summarised and reviewed by Černá and Bencko (7).

PCBs levels in human milk have been analysed extensively. The first data concerning the analysis of PCBs in body fluids and tissues of Czech population appeared in literature in 1985. The mean concentration of sum PCBs (related to the standard commercial mixture Delor 106) in 63 human milk specimens collected in Northern Bohemia was 2.83 mg/kg fat (i.e. 0.105 mg/kg milk with substantial inter-individual differences) (8).

In the 90s, such presentation of the PCB results was replaced by the analysis of individual congeners. In a WHO/EURO study of the region, the Czech and Slovak Republics showed higher levels of PCBs in districts producing technical mixtures of PCBs (district Michalovce, Eastern Slovakia) or using them intensively (district Uherské Hradiště, Eastern Czech Republic) (Table 1).

Table 1 The m	ean concentration of indicator c	congeners 138, 153,	and 180 in human milk of the	Э
Czech and Sl	lovak populations (μg/kg fat); 1 ε	sample pooled from	11 (Czech) and 10 (Slovak)	
	individual sa	amples (48, 49)		

Locality	Year	PCB 138	PCB 153	PCB 180	ΣPCBs ¹
Uherské Hradiště	1991–3	341.5	424.8	294.0	1,802.5 ¹
Kladno		171.9	215	137.4	897.4 ¹
Michalovce	1993–4	279.2	434.9	284.8	1,698 ¹
Nitra		139.8	207.7	137.9	825.2 ¹

¹ ΣPCBs = (PCB 138+PCB 153+PCB 180) x 1.70

1.70 = correction factor

Since 1994, the levels of indicator congeners in human milk have been systematically monitored within the System of Monitoring the Environmental Impact on Population Health of the Czech Republic. The results obtained in years 1994–1996 confirm the tendency toward a decrease (see Table 2) (9).

Year	Ν	PCB 138	PCB 153	PCB 180	ΣPCBs ¹
1994	282	236 (190)	398 (352)	308 (255)	1,601 (1,355)
1995	395	212 (178)	350 (323)	272 (248)	1,418 (1,273)
1996	285	184 (163)	230 (201)	150 (125)	959 (831)

Table 2The mean and median concentrations of indicator PCB congeners 138, 153, and 180 in
human milk of the Czech populations ($\mu g/kg$ fat) (9)

¹ ΣPCBs = (PCB 138+PCB 153+PCB 180) x 1.70

1.70 = correction factor

Hajšlová and co-workers determined the concentrations of PCBs and OCCs in human milk samples from three regions in the Czech Republic (4). The results were generated by the congener-specific analyses and compared with similar studies from other European countries.

Table 3 compares the levels of indicator PCBs in human milk between the Czech Republic (Prague subset) and similar studies from Norway, UK, the Netherlands, and Germany (4).

The sum of indicator PCBs was higher in Czech samples than in those of foreign origin while the levels of lower chlorinated PCBs (congeners no. 28 and 52) and pentachlorobiphenyl no. 101 were below the quantification limit of 5 ng/g fat. This fact may be attributed to different contamination pattern of the Czech diet and, consequently, different dietary exposure. Technical mixtures with the prevailing content of hexachloro- and heptachlorobiphenyls were mostly the primary sources of environmental pollution in the former Czechoslovakia. PCB 153 was the dominant congener in all the samples.

Regarding the composition of the market basket of countries listed in Table 3, fish and fish products are now consumed in higher rates. The annual consumption statistics in Norway, UK, the Netherlands, and the former Czechoslovakia in the end 80s were 41.1 kg, 19.9 kg, 9.2 kg, and 6.8 kg of fish and fish products per capita, respectively (10). A relatively high content of PCBs was often reported in fish from the Baltic and the North Sea. As the biodegradation of PCBs in fish is very limited, the higher intake of lower chlorinated PCBs (that are typically accumulated in these biota) via this commodity may be reflected in elevated levels in human milk (4).

The comparison of PCB levels in human milk between three Czech regions – Prague standing for industrial urban agglomeration, Kladno, a small industrial city, and Uherské Hradiště, a locality characterised by high environmental burden of PCBs from paint industry and a wide use of these paints in local agricultural facilities –

Country	Norwa N=28	ay ¹ B	UI N=	K² 32	The Nethe N=1	rlands³ 9	Germ 198 N=	any⁴ 4/5 69	Germa 1990/ N=6	any⁴ ′91 8	Czech Re 1993 N=1	epublic⁵ 3/4 I7
Congener PCB No.:	Average I (range)	Median	Average (range)	Median	Average (range)	Median	Average (range)	Median	Average (range)	Median	Average (range)	Median
28	7.8 (nd–24.2)	22.1	31.5 (nd–188)	17.0	12.1 (0.2–188.6)	5.8	0.5 (0.5–40)	5	17 (9–46)	18	nq ⁷	-
52	nq ⁶ (nd–154)	-	26.2 (nd–32.7)	6.4	2.6 (0.5–60)	1.5	0.5 (6–44)	2	13	15	nq ⁷	-
101	1.1 (nd–4.7)	nd	15.0 (nd–82)	4.9	1.5 (0.2–10.0)	1.1	15 (0.5–19)	21	14 (0.5–55)	17	nq ⁷	-
118	26.2 (9.6–56.7)	23.7	28.6 (5–197)	19.4	35.5 (9.7–94.0)	32.7	na ⁷ (7–59)	-	na ⁷	-	28.5	25
138	86.8 (74.6–185.9)	91.0	68.1 (18–261)	56.4	129.9 (43.8–314.3)	124.2	250 (60–660)	254	168 (65–669)	184	289 ⁷ (100–558)	260
153	114.4 (49.6–259.4)	9.9	85.9 (27–275)	71.8	186.3 (59.9–475.7)	174.7	325 (70–750)	324	240 (108–968)	264	379 (142–702)	331
180	50.6 (9.7–108.3)	46.0	74.9 (1–210)	70.1	76.9 (2.5–418.8)	71.3	160 (40–400)	162	173 (75–1,023)	194	240 (91–447)	226
ΣPCBs	286.9	-	330.2	-	444.8	-	751	-	625	-	937	-

Table 3 Comparison of levels of indicator PCBs in human milk between the Czech Republic (Prague subset) and Norway, UK, the Netherlands, and Germany (ng/g in fat) (4)

¹ (50) ² (51)

³ (52)

4 (53)

⁵ (4)

⁶ not quantified due to the interference with an unknown peak

⁷ below the limit of quantification, <5 ng/g in fat
 ⁸ PCB 138 was coeluted with PCB 163 (expressed as PCB 138)

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clearly showed that the levels of PCBs were the highest in the latter locality (Table 4). This region is more contaminated than other regions and therefore the PCB levels in crops grown there are higher, resulting in higher body burden as evident from the human milk analysis. The levels of PCBs in Prague and Kladno were lower and did not distinctly differ from one another. As to the chlorinated pesticides, the higher content of DDT (and its metabolite DDE) found in Uherské Hradiště may be attributed to its wide agricultural use in the past. Levels of HCB, which may be formed in various combustion processes, were slightly higher in the industrial area such as Prague and Kladno. The contents of b-HCH was similar in all sets of examined samples (4).

 Table 4 Content of PCBs and OCCs in human milk – interregional comparison in the Czech Republic (ng/g in fat) (4)

Region	ΣPCBs	β-нсн	DDTs	HCB	Number of samples
Prague	1,096	71 79	998 832	639 570	17 17
Uherské Hradiště	1,529	80	1,283	482	12

 Σ PCBs = sum of congeners nos.: 28, 66, 70, 74, 105, 118, 138, 153, 156, 170, and 180. DDT = p,p'-DDE+p,p'-DDT

A newer study (11) was focused on distribution of PCBs and OCCs in various human tissue samples from selected regions of the Czech and Slovak Republics and on comparison with the results from other foreign and local studies.

The analysis included sets of samples consisting of various human tissues originating from three regions, two representing very high environmental contamination (Uherské Hradiště, Czech Republic and Michalovce, Slovakia) and one (Prague) representing industrial urban agglomeration with intensive anthropogenic activities and many potential sources of pollution. The results were used to assess PCB and OCC body burden of the Czech and Slovak population. The biotic samples were collected during post-mortem of people killed in car accidents (Michalovce and Prague) or who died of natural causes (Uherské Hradiště). The analysis included abdominal adipose tissue (from subcutaneous area), liver, kidney, heart muscle (in the case of region Uherské Hradiště only), and the adipose tissue from the mesenteric area (only in the sets from Prague and Michalovce).

These measurements (11) confirmed again that organochlorine residues in the tissues of Czech and Slovak origin were often higher (especially for PCBs and DDTs) than levels in humans from other countries. The sum of indicator PCBs in samples from Michalovce and Uherské Hradiště was substantially higher than for example in Finland, UK, or the Netherlands.

With exception of the Slovak samples, in all other studies tri- and tetrachlorobiphenyls (PCB 28 and 52) were present in trace concentrations (up to 10 μ g/kg). Surprisingly high amounts of these congeners were found in the specimens collected in Michalovce. The elevated levels of lower chlorinated congeners in human tissues indicate actual exposure to PCB technical mixtures with the low chlorine content (12). On the other hand, the high levels of PCB 28 and 74 only (the most persistent congeners among low chlorinated PCBs) suggest long-term occupational exposure to such technical mixtures (11). The occurrence of both types of exposures in the locality Michalovce are highly probable, but the data on the profession of donors were not available. The study showed that vegetables played an important part in the intake of lower chlorinated PCBs due to the ambient air contamination in the area. However, relatively high concentrations of PCB 28 found in Michalovce seem unlikely to be associated with the high consumption of vegetables.

Biological monitoring of different human tissues indicated that concentrations of PCBs in blood, adipose, and muscle tissues were about the same as when calculated on a lipid basis. In this study (11) higher PCB levels were found in tissue richer in fat such as adipose. The distribution patterns of PCBs in other analysed tissues revealed similarity to those found for the DDT group. HCH isomers predominantly accumulated in the liver and kidney, reaching concentrations several times higher (2.5–5 and 2–3 times more, respectively) than in the adipose tissue. Similarly, HCB tended to accumulate in the soft tissue.

The sample subset from the region Uherské Hradiště was analysed for 38 individual chlorobiphenyl congeners. The contribution of individual chlorobiphenyls to adipose tissue slightly differed from contribution to other tissues (liver, kidney and heart muscle) typically containing higher amounts of more polar lipids (phospholipids). The PCB pattern was almost equal in the latter matrices. The levels of relatively more polar lower chlorinated congeners were higher in these tissues than in the adipose tissue. Nevertheless, highly chlorinated PCBs, particularly those with 2,4,5-chlorines in one ring and at least one chlorine in 4 position in the second one (congeners nos. 138, 153, 170, and 180) prevailed in all analysed samples. This finding indicates long-term environmental exposure in the analysed region. The contribution of highly chlorinated PCBs to the total amount of PCBs exceeded 70%.

For general population, food is the main route of environmental exposure to polychlorinated biphenyls, dibenzo-p-dioxins, and dibenzofurans (PCBs/PCDDs/Fs) (12, 13). According to the System of Monitoring the Environmental Impact on Population Health of the Czech Republic, the intake of PCBs (expressed as the sum of 7 indicator PCB congeners) for the average Czech population in 1996 was 86 ng/kg body weight. The population exposure to 11 toxic PCB congeners in 1998 was estimated to 12.3 pg of international toxic equivalent of 2,3,7,8-tetrachlorodibenzo-p-dioxins (I-TEQ) per kg body weight. The levels of indicator PCB congeners in human milk show a declining trend (7, 13). The levels of PCDDs/PCDFs in the pooled human milk samples collected in 1998 were about 10 pg TEQ/g fat. In the recent years, the monitoring system also included the analysis of PCDDs/PCDFs in the human adipose tissue to determine the background exposure to these compounds.

In the years 1996–1999, adipose tissue samples were collected during postmortem according to a specific protocol in four districts of the Czech Republic. Two were industrial (Plzeň and Ústí nad Labem) and two rural and recreational (Beroun and Ždár nad Sázavou). The basic characteristics of sampled persons are summarised in Table 5.

Regions	with lower pollution	Regions with	0	
Beroun	Žď ár nad Sázavou	Plzeň town	Ústí nad Labem	Overall
12.0	14.0	12.0	23.0	61.0
4.0	12.0	7.0	8.0	31.0
8.0	2.0	5.0	15.0	30.0
52.8	45.4	60.1	62.0	56.2
40–62	27–60	45–74	42-84	27–84
Average age – men Average age – women				51.3 61.0*
	Regions Beroun 12.0 4.0 8.0 52.8 40-62 age - men age - wome	Arr Point Arr Point Beroun Žďár nad Sázavou 12.0 14.0 4.0 12.0 8.0 2.0 52.8 45.4 40-62 27-60	Regions with lower pollution Regions with Beroun Žďár nad Sázavou Plzeň town 12.0 14.0 12.0 4.0 12.0 7.0 8.0 2.0 5.0 52.8 45.4 60.1 40-62 27-60 45-74	Regions with lower pollution Regions with higher pollution Beroun Žďár nad Sázavou Plzeň town Ústí nad Labem 12.0 14.0 12.0 23.0 4.0 12.0 7.0 8.0 8.0 2.0 5.0 15.0 52.8 45.4 60.1 62.0 40-62 27-60 45-74 42-84

Table 5	Characteristics of anal	vsed samples by sex	age and region (13	()
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* P<0.01

Polychlorinated dibenzo-p-dioxin (PCDD) and polychlorinated dibenzofuran (PCDF) levels, selected PCB congeners with dioxin toxicity (77, 126, 169, 123, 14, 105, 167, 156, 157, and 189), as well as the indicator PCB congeners (28, 52, 101, 118, 138, 153, 180) were determined in the fat melted off from homogenised samples of human adipose tissue. The toxicity equivalency factors (TEFs) proposed by *Van den Berg and co-workers* (14) were used to calculate the dioxin toxic equivalency (TEQ). Table 6 shows the overall mean total TEQ values as well as their distribution between sexes and age groups. Because of the log-normal distribution, median values were used in descriptive statistics. The sum of all PCDDs was about twice as high as the sum of all PCDFs. The concentration of PCDDs/Fs/PCBs correlated with age (r=0.47, P<0.01). Differences between sexes in concentrations and TEQ values, though women showed slightly higher values, were not significant despite the fact that the mean

Group	Ν		2,3,7,8-TCDD	PCDDs*	PCDFs	PCBs	Total
Overall study	61	Median Ranges	2.4 0.7–6.7	9.4 2.3–29.0	16.9 4.4–142	69.2 20.8–309	98.0 28.0–480
Men	31	Median Ranges	1.9 0.7–5.5	8.1 2.3–16.6	16.3 4.4–41.1	66.0 20.5–163	92.0 28.0–192
Women	30	Median Ranges	2.7 1.2–6.7	10.6 4.3–28.7	19.6 8.6–142	72.1 36.3–301	100.0 54.0–480
Overall, age >50	39	Median Ranges	2.6 0.7–6.7	10.4 3.3–29.0	19.1 6.7–142	80.4 30.0–309	102.0 40.0–480
Overall, age <50	22	Median Ranges	2.0 0.9–4.9	8.4 2.3–17.7	16.3 4.0–41.1	61.0 21.0–164	86.0 28.0–188

Table 6 Concentrations of PCDDs/Fs/PCBs in human adipose tissue expressed asTEQ (pg/g fat) (13)

* PCDDs including 2,3,7,8 TCDD

age of women was almost 10 years greater than that of men and that the difference between the age groups was significant (P < 0.01).

Table 7 shows the distribution of the total dioxin toxicity (TEQ) between PCDD, PCDF, and PCB groups. PCDDs (median value) contributed with about 9.4 pg TEQ/ g fat, PCDFs with 16.9, and PCBs with 69.2 pg/g fat to the total TEQ value. Compared with levels in human milk, these levels were distinctly higher (13).

Table 7	Dioxin toxic equivalents (TEQ) and the percentage of contribution to total TEQ (pg/g fat)
	in human adipose tissue (13)

Group	PCDDs (%)	PCDFs (%)	PCBs (%)
Overall	9.8	17.7	69.2
Men	9.0	18.0	73.0
Women	10.4	19.2	70.5

In agreement with the results published by *Schechter and co-workers* (54), 2,3,4,7,8-PCDF was the dominant congener among the PCDD/F compounds in our group. This compound contributed to more than 80% of the dioxin toxic equivalents of PCDF congeners. OCDD as the congener found in the largest amount (its median concentration was 114 pg/g fat), participated with about 53% of the total PCDDs/Fs concentration, but its contribution to the dioxin toxicity was negligible. PCBs 156 and 126 accounted for 84% in men and 95% in women of TEQ portion derived from the PCB congeners with dioxin toxic activity.

Regional differences were observed in concentrations of individual congeners as well as in TEQ values. Samples from industrialised districts (N=35) showed slightly higher levels in the fat tissue than rural districts (N=26). However, the differences were not significant, probably because the mean age of persons in industrialised areas was significantly higher that in the rural areas. No significant regional or sex differences es were observed in the contribution of individual congeners to the total TEQ.

Among the indicator PCB congeners, PCB 138, 153, and 180 contributed substantially to the total PCB level in human adipose tissue. The results are given in Table 8 as the sum of those congeners. The level of PCBs correlated significantly with age (r=0.3,

Group	Median	Ranges
Overall	1,753	786–8,167
Men	1,822	786–3,536
Women	1,695	836-8,167
Age >50	2,082*	1,147–8,178
Age <50	1,611	786–3,362

 Table 8
 PCB levels expressed as the sum of indicator PCB congeners 138, 153, and 180 (median) (pg/g fat) (13)

* P<0.05

P<0.01). Significantly higher values were obtained in the age group above 50 years (P<0.05) than in the group below 50 years of age. No significant sex or regional differences in concentration levels were found.

To conclude, the presented values bring the first human data characterising the background exposure of the Czech population. It is evident that the Czech population ran a higher exposure risk in the past. PCBs contributed more dioxin-like toxicity in human tissues than did dioxins and dibenzofurans. However, data concerning the level of PCDDs/Fs have been insufficient to delineate the time-related trends of the body burden of the Czech population, which stresses the importance of further monitoring activities.

Slovakia

The incidence of carcinogenic diseases in Slovakia is among the highest in the world. It is possible that increasing environmental pollution by toxic chemicals, including persistent chloroaromatic compounds, may contribute to this undesirable trend (5, 6). The potential exposure of human population in Slovakia has been studied systematically for many years. The researchers from the Institute of Preventive and Clinical Medicine in Bratislava have published many research papers and reports in the last decade.

The growing attention to the study of contamination of total diet, mother's milk and adapted cow's milk, dairy products, and other food commodities (15). The contamination levels of organochlorine insecticides and indicator congeners of PCBs in milk and butter from Bratislava markets were studied in 1994. The obtained results for organochlorine insecticides showed that the highest levels were measured for p,p'-DDE and p,p'-DDT. Those findings may be attributed to the use of non-standard imported feed in the farms. This was particularly true in the months of early spring when the domestic feed for animals was lacking. In spite of measures taken in 1973, lindane (γ -HCH) preparations (e. g. Lindan[®], Hermal[®], and Sanigran[®]) were applied. The findings of total DDT in Slovakia (0.059 mg/kg on fat basis) can be compared with those in butter in the Czech Republic, for example by *Strnad* (16), with mean of 0.053 mg/kg on fat basis and *Bartonicek and Rob* (17) with 0.033 mg/kg on fat basis. In Poland, the total mean DDT level in butter was 0.096 mg/kg on fat basis and the maximum 0.272 mg/kg on fat basis (v. the Slovak maximum of 0.208) (18).

Although the mean levels of HCB were relatively low, the maximum levels could be influenced by environmental contamination with this compound. Agriculture in the 90s was not the main source of HCB environmental inputs in the CEE countries. Many HCB residues came from the deposits of chemical wastes. A case in point are the outskirts of the Slovak capital Bratislava, considered a »hot spot« of HCB contamination (18). The HCB levels found (mean value 0.004 mg/kg on fat basis) were in a good correlation with those in the butter samples from the area of Schwerin (Germany) in 1990 with the mean content of 0.007 mg/kg on fat basis (17) and with the 1991 Czech report by *Strnad* (18) with a mean level of 0.014 mg/kg on fat basis. One year earlier (1990), Bartonicek (16) found the mean HCB level in butter at 0.044 mg/kg on fat basis.

AS regards the conditions of the CEE milk farming at that time, one sample of butter represents a mean obtained from the milk coming from a number of farms. From this point of view, the contamination levels measured in the butter can be

regarded as the measure of environmental contamination with persistent organochlorine compounds (17).

The results concerning PCBs (15) in fifty samples of mother's milk and 20 samples of adapted cow's milk for bottle feeding supplied from the lactarium of the Pediatric Hospital in Bratislava were examined for the presence of 6 indicator congeners of PCBs. Similar to the Czech study by *Schoula and co-workers* Congeners nos. 138, 153, and 180 were predominant in all samples (4, 11). The highest mean as well as the maximum level was found for PCB 138 for both types of samples.

The average daily intake based on the sum of indicator congeners was 2.56 μ g/kg bw for mother milk, and 0.85 μ g/kg bw for adapted cow's milk. The calculated average daily intake from mother milk exceeded the value of Acceptable Daily Intake which is 1 μ g/kg bw, but this was also observed in other countries. From this point of view bottle feeding would seem more advantageous, since the PCB levels are significantly lower in adapted cow's milk. Furthermore, the content of all congeners found in adapted cow's milk was substantially below the permissible level (0.04 mg/kg for 28, 52, 101, 180 and 0.05 mg/kg for 138 and 153 in fat).

Samples of subcutaneous abdominal adipose tissues were obtained from the Slovak population through post-mortem and analysed for the level of PCBs (20). The concentrations found in Bratislava (men, N=32, women, N=14), were in the ranges 0.8 to 10.4 and 0.6 to 7.7 mg/kg fat, in Trenčín (men, N=18, women, N=11) 2.1 to 5.5 and 0.4 to 6.1 and in Martin (men, N=17, women, N=13) 0.4 to 4.7 and 0.4 to 4.8, respectively.

Fifty samples of human blood collected in 1992 from the general human population living in five selected areas of the Slovak Republic (the Michalovce, Velký Krtiš and Nitra Districts, Myjava area, and Bratislava) were analysed for 18 PCB congeners and some organochlorine pesticides (HCB, lindane, p,p'-DDE and p,p'-DDT). Table 9 shows the levels of these pollutants in serum lipids averaged for all the samples analysed.

Pollutant	Mean	Median	Range
PCBs (18 congeners)	1.79	1.33	-0.53- 9.20
HCB	5.38	4.27	-0.16- 23.20
γ-HCH	0.012		<0.01-0.18
p,p'-DDE	6.05	4.39	-1.30- 34.80
p,p'-DDT	0.27	0.23	<0.01-0.79

Table 9 Levels in serum lipids averaged from five selected areas in Slovakia in 1992 (µg/kg) (20)

About three times higher levels of PCBs were found in the samples from the Michalovce District where PCB formulations had been produced. PCB levels in the Slovak population in 1992 were similar to earlier findings, but they substantially exceeded human PCB levels found in other countries. The difference was even more evident with HCB the levels of which were about hundred times higher than the corresponding levels in the USA, Japan, Finland, or Canada. The paper also described the arithmetic mean values of PCBs, p,p'-DDE, p,p'-DDT and HCB found in

the serum lipids of men and women. The mean levels of p,p'-DDT and PCBs were lower in women.

The data reviewed herein suggest that the levels of PCBs and related compounds in humans from the former Czechoslovakia are higher than reported by studies of other countries. Higher exposure was found in the vicinity of PCB mixture manufacturing or using. Since PCB production was banned in 1984, the total environmental PCB contamination and consequent body burden in the general population of the Czech and Slovak Republics have decreased. However, human exposure has not been excluded entirely (7).

The other part of this project was focused on the investigation of levels of PCBs and selected organochlorine pesticides in human milk in the same five model areas (21). Based on the WHO protocol, the human milk samples were collected from breast-feeding mothers (primiparae, milk sampling 2 weeks-2 months after delivery, unchanged residence at least 5 years before pregnancy).

HCB and p,p'-DDE were present in the human milk samples at higher concentrations than individual PCB congeners and p,p'-DDT. HCB levels are noteworthy, as they are one or two orders of magnitude higher than reported from other countries (see Table 10). Surprisingly high concentrations of HCB found in human samples from Slovakia were probably caused by its use in agriculture and its formation during industrial manufacture of some chlorinated solvents.

Year of	No. of	Country	Average concentration (μ g/kg, fat basis)					
collection	samples	country	6 PCBs ¹	PCB-118	НСВ	p,p'-DDE	p,p'-DDT	
1986	412	Canada ²	1,042	20	_	_	_	
1991	16	Canada, Quebec ²	109	17	-	-	-	
1993	107	Canada,						
		Quebec, (Inuit)	706	-	136	1,212	-	
1991	32	UK	302	29	-	-	-	
1990–91	57	UK, Wales	232	18	-	-	-	
1991	28	Norway, Oslo ²	261	26	-	-	-	
1990–92	195	Netherlands, Rotterdam	439	35.5	-	-	-	
1993	24	Jordan, Amman	351	-	-	-	-	
1190–91	68	Germany, Middle Hesse	692	-	-	-	-	
1991	113	Germany,						
		N. Rhine Westphalia ²	581	-	177	504	27	
1989–90	59	Jordan, Amman	-	-	290	2,040	450	
1991	51	Spain, Madrid	-	-	0.8	604	12.5	
1985	100	Israel, Jerusalem	-	-	80	2,440	290	
1979–80	54	USA, Hawaii	-	-	46	1,989	162	
1981–82	50	Finland	-	-	64	850	36	
1987	64	Italy	271	41	192	2,050	154	
1990	25	Czech Rep., Jihlava	-	-	1,644	_	-	
1993–4	50	Slovakia ³	785	39	829	1,667	126	

Table 10	PCB and OCP levels found in human milk lipids from various countrie	s (21)
		- \ /

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

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

³ (25)

The levels of all pollutants determined in the human milk lipids were substantially lower than those in the blood and adipose tissue samples collected from the same areas. The differences in PCBs levels between milk samples from Michalovce District (location of a former PCB producer) and other districts were not as high as the differences in the adipose tissue and blood serum. This reason may be sought in the lower age of breast-feeding mothers than the mean ages of the adipose-tissue and blood donors and in the decreasing environmental OCP and PCB contamination after the ban.

The average ratios of p,p'-DDE to p,p'-DDT in the milk samples from Bratislava, Myjava, Nitra, Michalovce, and Velký Krtiš were 14.7, 12.6, 17.5, 9.8, and 13.9, respectively. This is less than was observed in the adipose tissue and blood serum samples (5, 6). PCB congeners nos. 153, 180, and 138 were dominant in all samples.

Having in mind that an average 5-kg infant suckling in the Michalovce District consumed 800 g of mother's milk, the calculated average intake of the sum of six PCB congeners was 7.5 μ g/kg bw per day. This intake ranged from 2.8 to 4.4 μ g/kg bw in the other four model areas, which is substantially higher than the acceptable or tolerable daily intake of 1 μ g/kg bw per day (involves all PCB congeners) established in some countries. The calculated daily intake of HCB and p,p'-DDE+p,p'-DDT, ranged in all areas from 2.0 to 5.1 μ g/kg bw and from 5.6 to 11.5 μ g/kg bw, respectively. The intake of DDE+DDT was lower than the acceptable daily intake (ADI) value established by WHO (20 μ g/kg bw per day), but the intake of HCB was two orders of magnitude higher than the tolerable daily intake of 0.08 μ g/kg bw issued by the Health Protection Branch of Health and Welfare Canada.

The presented results from the pilot study involved a relatively small number of specimens and it was questionable to evaluate any statistical parameters. However, a very unique study was based on these results in 1997–1998 and its results are presented below.

Kočan and co-workers also studied the PCDDs/Fs and coplanar PCBs levels in blood serum samples collected from the Slovak human general population (5 districts) and occupationally exposed workers (22). These pollutants were also determined in blood samples taken from a limited number of workers employed for a long time at a municipal waste incinerator or a PCB production plant.

Table 11 summarises the levels of PCDDs/Fs and of two of the most toxic coplanar PCBs (PCB-126 and PCB-169). The mean total lipids in the serum were 6.79 g/L. The mean TEQ_{PCDDs}, TEQ_{PCDFs}, and TEQ_{planPCBs} for each sampling location were similar, including the MWI samples, whereas TEQ_{PCDFs} and TEQ_{planPCBs} values were several times higher in samples from Chemko Strážské (former producer of PCBs mixtures). Increased TEQ levels in the Chemko samples were caused mainly by higher concentrations of 2,3,4,7,8-PeCDF and 3,3',4,4', 5-pentaCB (126). 2,3,4,7,8-PeCDF contributed 53% to the total mean TEQ found in the Slovak general population, 60% in the case of the MWI workers, and 80% in the case of the occupationally exposed workers in PCB production. There were no substantial differences between the PCDDs, PCDFs, and planPCBs levels in the general population samples from the sampling districts.

In spite of long-term exposure at the old-type waste incinerator, PCDDs/Fs and coplanar PCBs levels in the workers blood kept within the general population levels (Table 7). This is in agreement with published data, although some studies report a slight increase in the concentrations of higher chlorinated congeners.

Table 11 PCDD/F and coplanar levels (expressed as toxic equivalents, lipid-adjusted basis) in blood
serum samples taken from the human general population of 5 model districts of Slovakia and
occupationally exposed persons (MWI, former producer of PCBs). Only average values are
described, for details see ref. (22)

	Sex		вмі		Content (ng/kg, lipid adjusted)					
Area		Age	(kg/m²)	I-TEQ PCDDs	I-TEQ PCDFs	I-TEQ PCDDs/Fs	WHO-TEQ planPCBs	ΣTEQs		
Bratislava	3M/3F	37.0	26.3	5.6	18.2	24.1	9.6	33.7		
Michalovce	3M/3F	35.3	26.9	6.5	13	19.5	12.9	32.4		
Myjava	3M/3F	35.5	24.9	6.9	15.3	22.2	11.6	34.7		
Nitra	3M/3F	33.3	25.7	6.8	12.7	19.4	7.6	27		
Velký Krtiš	3M/3F	40.0	27.0	6.5	8.8	15.4	7.9	23.3		
Average (men)		36.9	26.3	6.6	15.3	22	10	32.1		
Average (women)		35.5	26.0	6.3	12	15.3	9.6	28		
Average (together)		36.2	26.1	6.5	13.5	20	9.9	30		
MWI	4M	41.3	28.3	6	16.6	22.6	9.5	31.9		
Chemko Co.	2M	42.0	30.8	7	68.3	75.3	63.1	138.4		

It is noteworthy that the ratio between I-TEQ_{PCDDs} and I-TEQ_{PCDFs} found in the Slovak general population was less than 1 (about 0.5), whereas reports from other countries showed much higher ratio – from about 1 for Germany and Spain to 2.7 for the USA. This was due to relatively high levels of 2,3,4,7,8-PeCDF (I-TEQQ=0.5) in the Slovak human samples in proportion to the total TEQ's. On the other hand, the levels of 1,2,3,7,8-PeCDD (a congener with the same I-TEF) were found to be very low. Authors assumed that the increased 2,3,4,7,8-PeCDF levels observed in the Slovak samples were due to increased exposure to PCBs. This assumption was supported by findings of a high 2,3,4,7,8-PeCDF content in the blood lipids of the Chemko workers occupationally exposed to PCBs. The ratio between the mean 2,3,4,7,8-PeCDF level in the Chemko and the mean level in general population samples was 6 (121.2:21.1), which was also the ratio of the total PCB levels found in the same samples.

The aim of the above study was to investigate PCB and OCC levels in samples of human population (from the District Michalovce, location of former producer of PCBs and the District Stropkov). The basic topic of that project was environmental and human population load in the area contaminated with PCBs (23). Table 12 brings the first summary of results from measurements of the PCB content in blood samples of human population from these two regions.

Higher content of PCBs determined in various kinds of foods from the District Michalovce led to higher levels of PCBs in human population from this district. The average concentration of PCBs in fat samples from the blood serum of general population of that region (107 men and 108 women) was 4.2 μ g/g, whereas in the District Stropkov (101 men and 104 women) it was 1.2 μ g/g. The PCB level of 8.6 μ g/g was found in occupationally exposed workers in the former PCB producer Chemko Strážské (27 men and 11 women). It is important to note that the exposure was 20–30 years before those measurements took place. A part of the general population was a group of 11 fishermen who consumed fish from contaminated waters of the Laborec River

	Contents (µg/g fat of blood serum)						
	ΣPCB ¹	нсв	нсн	DDE	DDT	% of DDT in SDDTs	
Professional exposure with PCBs (N=38)	8,567	1,699	22	3,867	130	3.3	
– men (N=27)	10,115	1,674	21	4,262	145	3.3	
- women (N=11)	4,767	1,761	22	2,899	94	2.4	
Fishermen (MI, N=11)	12,205	1,453	33	3,733	260	6.5	
General population MI							
(not occupationally exposed, N=215)	4,166	1,921	30	3,890	131	3.3	
- with the occupationally exposed (N=253)	4,827	1,888	29	3,865	141	3.5	
 with the occupationally exposed, 							
randomly selected PCB (N=225)	4,222	1,912	30	3,873	141	3.5	
- without fishermen (N=204)	3,753	1,945	30	3,874	136	3.4	
General population SP (N=205)	1,206	1,622	38	2,571	98	3.7	
Men MI (N=107); general population	4,802	1,419	33	3,528	152	4.1	
Women MI (N=108); general population	3,536	2,418	27	4,199	134	3.1	
Men SP (N=101); general population	1,385	1,217	36	2,940	98	3.2	
Women SP (N=104); general population	1,033	2,015	37	2,503	97	3.7	

Table 12 Levels (means) of PCBs (sum of 9 congeners), HCB, g-HCH, p,p'-DDE and p,p'-DDT in samples of blood serum from population groups of Districts Michalovce (MI) and Stropkov (SP) (23)

¹ Sum of the following congeners: 28, 52, 101, 138, 153, 180, 118, 156, and 170.

and Zemplínská Šírava Dam. This small and not very representative group showed very high (58.7 mg/kg) and relatively low (1.6–2.9 mg/kg) contamination levels. Women showed higher levels of PCB contamination in all evaluated groups from both districts, but not significantly. However, the study confirmed the association between consumption of contaminated homemade food such as eggs or chicken meat, and higher contamination levels in humans and the results were statistically significant. The levels of PCBs, HCB, and p,p'-DDE, (including the Stropkov District) were higher in Slovakia than in the EU countries, the USA, or Canada.

Placental contamination with xenobiotics may act as a biological marker of mother's or the foetus' exposure via placenta (24). In Slovakia, PCB congeners were detected in the human food chain including human breast milk, where the PCB's levels were higher than in cow's milk. The aim of that study was to compare the contamination of human placentas with organic xenobiotics (selected organochlorine compounds) in five environmentally different Slovak regions.

Samples of placental tissue were taken from mothers in five environmentally different regions in the Slovak Republic. The samples were analysed for the concentrations of 21 selected organochlorine compounds – polychlorinated biphenyls (PCBs – PCB-28, PCB-52, PCB-101, PCB-118, PCB-138, PCB-153, PCB-180), chlorinated benzenes (1,4+1,3-DCBz, 1,2-DCBz, 1,3,5-TCBz, 1,2,4-TCBz, 1,2,3-TrCBz, TeCBz, PeCBz, HCB), and organochlorine insecticides (α -HCH, β -HCH, γ -HCH, δ -HCH, p,p'-DDT, p,p'-DDE).

The statistical analysis of the placental organochlorine compounds concentrations revealed that the mean contents of 12 (1,4+1,3-DCBz, 1,3,5-TrCBz, 1,2,3-TrCBz, TeCBz, PeCBz, HCB, α -HCH, β -HCH, γ -HCH, δ -HCH, p,p'-DDT, and PCB-101) out of 21 organochlorine compounds analysed were significantly higher in the Region 1

	Compounds	Region 1 X±SD	Region 2 X±SD	Region 3 X±SD	Region 4 X±SD	Region 5 X±SD
А	1,4+1,3-DCBz 1,2-DCBz 1,3,5-TrCBz 1,2,4-TrCBz 1,2,3-TrCBz TeCBz PeCBz HCB	2.7±3.6 6.3±9.0 25.9±9.9 4.9±10.1 0.8±0.8 0.7±2.0 4.5±14.0 2.0±9.5	1.5±2.3 0.2±0.3 0.0±0.1 0.1±0.3 0.1±0.2 0.6±2.7 0.0±0.1 0.3±0.3	4.10 ± 8.80 0.01 ± 0.04 0.08 ± 0.40 0.20 ± 0.30 0.10 ± 0.20 0.20 ± 0.80 0.01 ± 0.02 0.01 ± 0.02 0.40 ± 1.10	$11.40\pm21.00\\0.03\pm0.10\\0.01\pm0.08\\0.30\pm0.50\\0.20\pm0.30\\0.09\pm0.50\\0.02\pm0.07\\0.40\pm0.50$	2.6±5.4 12.1±14.0 2.7±5.2 6.8±9.9 0.7±0.8 0.3±1.4 1.0±1.6 0.5±0.5
в	α-HCH β-HCH γ-HCH δ-HCH P,p'-DDT P,p'-DDE	0.3±0.6 0.7±1.6 1.5±2.8 0.6±1.1 0.4±0.8 0.4±0.5	0.0±0.0 0.0±0.1 0.0±0.1 0.0±0.0 0.4±1.4 0.5±0.5	$\begin{array}{c} 0.20 \pm 0.30 \\ 0.20 \pm 0.30 \\ 0.60 \pm 0.40 \\ 0.01 \pm 0.05 \\ 0.01 \pm 0.04 \\ 0.40 \pm 0.40 \end{array}$	$\begin{array}{c} 0.10 \pm 0.20\\ 0.30 \pm 0.30\\ 0.50 \pm 0.50\\ 0.00 \pm 0.00\\ 0.04 \pm 0.20\\ 0.30 \pm 0.50\end{array}$	0.2±0.7 0.4±1.4 1.1±4.4 0.2±0.4 0.2±0.9 0.2±*0.3
С	PCB-28 PCB-52 PCB-101 PCB-118 PCB-138 PCB-153 PCB-180	0.2±0.6 0.1±0.2 5.6±17.5 0.7±3.2 2.1±1.7 5.6±22.6 0.2±0.4	0.4±1.7 1.4±1.5 0.5±1.0 - 0.9±2.2 1.3±2.7 0.8±1.9	0.03±0.07 0.04±0.20 0.10±0.30 - 0.10±0.10 0.20±0.20 0.20±1.30	0.08±0.30 0.05±0.20 0.40±8.30 - 0.20±0.40 0.20±0.50 0.80±3.60	n.d. 0.1±0.3 0.5±1.5 n.d.±0.1 0.6±1.4 1.2±3.4 n.d.

Table 13	Concentrations of organochl	orine compounds	(mg/kg x 10 ⁻³)	in human place	ntal samples in
5 reg	ons of Slovakia related to pre	dominant industria	al sources of e	environmental po	ollution (24)

A - chlorinated benzenes; B - organochlorine insecticides; C - polychlorinated biphenyls;

n.d. - below detection limit

Region 1 - industrial region polluted by organic chemical petrol, pesticide and rubber industries;

Region 2 – agricultural region;

Region 3 - region polluted by iron-ore mining;

Region 4 - industrial region polluted by iron-ore processing;

Region 5 - rural region with no point source of industrial pollution

polluted by chemical industry than in other investigated regions. However, concentrations of DDE and the PCB congeners 28, 52, 138, 153, and 180 were the highest in the agricultural region. Among the investigated regions, the lowest concentrations of 9 organochlorine compounds (1,2-DCBz, 1,3,5-TrCBz, TeCBz, PeCBz, p,p'-DDT, PCB-28, PCB-52, PCB-138, and PCB-153) were found in iron-ore mining or processing regions.

Exposure to organochlorine compounds can be determined in samples from various tissues, including foetal and placental tissues. Experimental findings in animals revealed that organochlorine compounds could induce oxidative stress in the foetal and placental tissue with subsequent tissue damage. The findings showed the production of superoxide anion and lipid peroxidation and DNA-single strand breaks. In our previous work we found more frequent pathological microstructural changes in the human placental samples collected from the Region 1 polluted by chemical industry than in the rural Region 5. The embryo/foetus is highly susceptible to adverse impacts of organochlorine compounds. Further research should focus on the mecha-

nisms of these compounds in reproductive pathology. Our findings pointed to contamination of human placenta with various organochlorine compounds, depending on the predominant type of pollution in the selected regions.

Reichrtová and co-workers studied the role of organic xenobiotics in human placenta in the allergic sensitisation in newborns. (25). Although we now understand more about the genetics of atopy and the role of Th1 and Th2 cells in the control of immunoglobulin E (IgE), environmental causes of atopy are still not clear. The evidence linking patterns of foetal growth to adult diseases has focused attention on the role of the placental environment in the aetiology of atopy and atopic diseases. In our study, we gathered fresh samples of full-term placentas and umbilical cord blood n the delivery rooms of hospitals from environmentally different regions (industrial, polluted predominantly by organic xenobiotics and traffic v. rural). Data concerning parameters of placentas and newborns were collected through questionnaires. Placenta samples were analysed for 21 compounds of persistent organic pollutants (POP) using capillary gas chromatography. Concentrations of total IgE (as a biomarker of a sensitisation of newborns) in the umbilical blood samples were also determined using capillary gas chromatography. Laboratory analyses showed significantly higher concentrations of 17 out of 21 POPs investigated in the placental samples collected from the industrialized region than in from the rural region.

Total IgE level in the umbilical blood samples gathered from industrial region were also higher, perhaps as the result of organic xenobiotics. External placental parameters differed too.

Allergic diseases are on the rise in both prevalence and severity, especially in industrialised countries. Developing foetuses and young children, especially those with a genetic propensity may become even more susceptible to the effects of chemical agents, that is more prone to allergic sensitisation (24).

In general, the foetus is protected against external influences by the placental barrier, but the barrier is selective, especially for maternal immunoglobulin G (IgG) antibodies, various antigens, and chemical substances. Contrary to the transplacental IgG transport, only a low amount of immunoglobulin E (IgE) antibodies is present in the newborns, and it seems that these IgE antibodies have a foetal origin. There are environmental chemicals and drugs (such as xenobiotics) that may enhance the sensitisation to allergens of various origin in susceptible persons (due to their modulative effect on T-cells). It is therefore of great importance to understand the pathogenic mechanisms (neoantigen formation, metabolism of xenobiotics into reactive, haptenic metabolites, induction of costimulatory enzymes, and sensitisation of T-cells) involved in the xenobiotics may influence the response to antigen exposure and bring about allergic diseases in the early childhood.

Demonstration of benzo(a)pyrene-DNA (B(a)P) adducts in human placenta and cord blood confirmed the metabolic capacity of the placenta, the transfer of B(a)P from the mother to the foetus, and the genotoxicity of B(a)P. Organochlorine compounds accumulate in the body during the lifetime and the individual body burden increases to levels that are toxic to the organism. The offspring is exposed *in utero* through maternal transfer. Organochlorine compounds exert estrogenic effects (endocrine disruption) and a variety of associated effects such as reproductive and immune system dysfunction. Neonatal exposure to polychlorinated biphenyls (PCBs), especially

to their congeners 28 and 52, were found to have persistent neurotoxic effect in adult animals. In Slovakia, these PCB congeners were detected in human breast milk as well as in cow's milk and dairy products.

Placental contamination by chemicals may act as a biological marker of exposure of the mother or foetus via placenta. Placentas collected from delivery rooms of hospitals from two Slovak regions. The samples were then analysed for 21 selected organochlorine compounds. The study was based on 2,050 full-term deliveries, randomly selected in two Slovak regions differing in the industrial potential and environmental pollution. The industrial region was represented by a city (Bratislava) polluted mainly by organic chemical industry (petrol, pesticides, and rubber industry) and by the traffic. The rural region was situated in the mountains (Stará Lubovna) away from industrial sources of environmental pollution, but with traffic that goes with tourism and a border crossing. Women were selected according to the following criteria: residence in the investigated areas at least 3 years before the conception, normal term of delivery (40±2 weeks of gestation), and non-occupational exposure to organochlorine compounds. The data from questionnaires focused on mothers (e. g. residence, smoking habit, occupation). The external parameters were analysed such as placental longest and transverse diameter and thickness and the birth weight and height of the newborns. The average age of the investigated mothers in the industrial region was 24.2 and in the rural region 22.6 years. In the industrial region 26% mothers smoked cigarettes and in the rural region 27%. The incidence rate of atopic eczema (per 10,000 children) in 1995 was 30.82 in Bratislava and 12.78 in Stará Lubovna. Samples of cord blood of newborns (N=2 050) were collected in respective maternity clinics, and sera prepared by centrifugation. Simultaneously, randomly selected samples (N=120) of full-term placentas were taken. Specimens of cord blood from 2,050neonates were gathered for the determination of levels of total immunoglobulin E (IgE).

Table 14 shows the contents of 21 substances of organochlorine compounds in the human placental samples collected from the industrial and the rural region. Comparisons revealed that both the placental contamination with 16 (out of 21) organochlorine compounds and the cord serum IgE levels were significantly higher in the industrial region. Furthermore, the percentage of non-contaminated placental samples was significantly higher in the rural region than in the industrial. The content of all congeners of polychlorinated biphenyls (mainly PCB congeners 101 and 153) and organochlorine insecticides analysed in the placental samples was higher in the industrial region. The neonates were divided according to their cord serum IgE concentration in 3 groups: <0.7 kU/L (e.g. negative newborns), 0.7-3.5 kU/L, and >3.5 kU/L. The 2nd and the 3rd group were largely dominated by IgE-positive neonates from the industrial region (expressed in percentage) (P < 0.001). Quite expectedly, there were less IgE-negative newborns from the industrial region (68.7%) than from the rural region (82.4%). The findings pointed to an association between organochlorine compounds and the higher levels of total IgE in newborns, suggesting a higher risk of allergic sensitisation in the industrial region. This association was supported by the higher incidence of atopic eczema recorded for population from the industrial region. The positive Spearman correlations for p,p'-DDE (r=0.3294, P=0.01) and for PCB 118 (r=0.3482, P=0.006) and cord serum total IgE level were found.

		Concentration (µg/kg)								
	Compounds	Indus	strial re	gion	Rur	Rural region				
		Median	Max.	% n.d	Median	Max.	% n.d			
	1,4+1,3-DCBz	1.4	218.0	19	0.8	26.9	21			
	1,2-DCBz	0.8	46.9	18	7.6	64.3	18			
	1,3,5-TrCBz	10.2	310.4	11	0.7	31.5	14			
^	1,2,4-TrCBz	0.5	41.9	14	1.9	50.5	13			
А	1,2,3-TrCBz	0.7	3.0	14	0.5	3.5	32			
	TeCBz	0.1	12.7	28	n.d.	10.4	68			
	PeCBz	0.4	102.2	23	0.2	7.0	32			
	HCB	0.6	72.0	2	0.4	2.0	25			
	α-HCH	0.2	4.0	19	n.d.	5.4	70			
	β-НСН	0.3	12.0	12	0.1	11.2	59			
-	γ-HCH	0.6	17.5	5	0.2	33.1	40			
в	δ-HCH	0.3	5.4	25	n.d.	3.1	65			
	P,p'-DDT	0.1	3.5	26	n.d.	5.2	54			
	P,p'-DDE	0.1	2.2	12	0.1	2.0	24			
	PCB-28	0.1	4.0	14	n.d.	0.2	73			
	PCB-52	0.1	0.6	12	n.d.	2.0	67			
	PCB-101	0.2	109.0	26	n.d.	8.9	76			
С	PCB-118	0.1	23.5	18	n.d.	0.4	64			
	PCB-138	0.2	7.9	5	n.d.	6.4	60			
	PCB-153	0.2	124.8	9	0.1	24.4	46			
	PCB-180	0.1	1.9	32	n.d.	0.1	81			

Table 14 Median and maximum concentrations of organochlorine compounds and the percentage of samples below detection limits in human placentas collected from industrial and rural regions (24)

Industrial region: N=57; Rural region: N=63; A – chlorinated benzenes; B – organochlorine insecticides, C – polychlorinated biphenyls; Max – maximum; n.d. – below limit of detection,

% n.d. - percentage of samples below limit of detection

External parameters of human term placentas and birth parameters of neonates between the rural and the industrial region are compared in Table 15. In spite of even

 Table 15
 External parameters of human full-term placentas and birth parameters compared between industrial and rural regions (24)

Industrial region X±SD	Rural region X±SD	Difference
586.9±135.7	572.5±107.3	n.s.
18.9±2.8	16.3±2.9	P<0.001
15.7±2.2	12.8±2.3	P<0.001
2.3±0.6	1.8±0.7	P<0.001
3,287.9±595.7	3,347.2±462.0	n.s.
49.9±2.5	49.6±2.1	n.s.
	Industrial region X±SD 586.9±135.7 18.9±2.8 15.7±2.2 2.3±0.6 3,287.9±595.7 49.9±2.5	Industrial region X±SDRural region X±SD586.9±135.7572.5±107.318.9±2.816.3±2.915.7±2.212.8±2.32.3±0.61.8±0.73,287.9±595.73,347.2±462.049.9±2.549.6±2.1

n.s.-not significant

average weights of placentas in both regions, other parameters (e.g. longest diameter, transverse diameter and thickness) were significantly higher in the rural region. The comparison of birth parameters (weight and height) revealed that there were no significant differences between the regions. However, the industrial region (Bratislava) had a higher percentage of microstructural lesions diagnosed in human placentas than the rural region (Stará Lubovna). These microstructural lesions may be responsible for different external parameters of human placentas.

The comparison of biomarkers of mothers' chemical exposure (e. g. different placental metal and organochlorine content in placental samples) with the cord sera IgE levels between the two regions has led to a hypothesis of possible foetal allergic sensitisation evoked by organochlorine compounds in the placenta.

Poland

Most persistent organochlorine pesticides, excluding lindane, were banned in Poland in 1975/76. The first restrictions of use and marketing of lindane became effective in 1980 and were gradually extended until its agricultural use was ultimately banned in 1989. Unfortunately, there are no detailed data on the use and environmental release of PCBs in Poland. OCCs and PCBs in human adipose tissues were studied in Warsaw (26), Gdaňsk (a chemically more detailed study), and an inland province (27).

Subcutaneous adipose tissues were taken from surgically treated patients in Warsaw hospitals (26). Samples were collected between 1989 and 1992 from male and female persons aged between 10 and 80. The total number of analysed samples was 277 (142 from male, and 135 from female persons) (Table 16).

Pollutant	Mean	Median	Maximum
PCBs	0.856	0.500	36.000
HCB	0.310	0.120	9.020
α-HCH	0.016		0.160
β-НСН	0.228	0.120	5.097
γ-HCH	0.074	0.030	2.727
p,p'-DDE	5.745	4.382	35.850
p,p'-DDT	0.537	0.478	9.600

Table 16Levels of organochlorine compounds in human adipose tissue from 277 patients from
Warsaw hospital, Poland in 1989–1992 (mg/kg of fat) (26)

The study has hown that age may considerably contribute to organochlorine concentrations in human adipose tissue. The same authors studied the excretion of these pollutants from human body by lactation (28). Lactation is an important factor in disposing organochlorine compounds from the female body. That is why the authors followed up the number of deliveries per woman and subsequent lactations and sought to understand their role in OCCs excretion through milk. The relationship between age, number of deliveries, and the concentrations of OCCs in mother's milk was identified by examining the findings from the analysis of 253 samples of human

breast milk. The analysis included 108 milk samples from primiparae and 145 samples from multiparae (2–7 deliveries).

No decline was found in the mean concentrations of OCCs in multiparae as compared to primiparae. The reason may lie in the age of the studied women. Mean HCB and Σ HCHs concentrations were similar between the groups, and the mean p,p'-DDT and PCBs concentrations were even higher in multiparae (statistically significant only for PCBs; P \leq 0.05).

Women with the highest number of deliveries (over four) were reported the highest DDT levels, DDT metabolite levels, and PCB levels. Quite expectedly, the average age of those donors was also the highest – 33 years.

The concentrations of chlorinated hydrocarbons identified in human milk are the result of two processes: bioaccumulation of such compounds in the adipose tissue and the excretion of those compounds to human milk in the course of lactation. Older women may be expected to have higher concentrations of OCCs due to longer exposure. On the other hand, lactation is an important way to dispose of such compounds from the body. Daily disposal during lactation is much greater than the daily intake. Thus, the concentrations of chlorinated hydrocarbons in human milk may be expected to fall as the number of deliveries increases (28–30).

Newborns become chronically exposed to organochlorine compounds mainly through food (28, 29). Bioavailability of these compounds in breast-fed infants is higher than that in formula-fed infants. It is therefore critical that one should determine whether these compounds are hazardous to the health of infants and small children in the high risk category and identify the safety margin between the current concentrations of the OCPs in human milk and the limit beyond which health hazard is no longer acceptable.

That was the rationale of another study of infant exposure to PCBs and OCPs (HCB, HCHs, DDTs) from mother's milk (31). Samples of human milk were collected in lactarium and maternity clinic in Warsaw and from donors from different regions in Poland. The analysis comprised 462 human milk samples. To assessment of exposure of breast-fed children to the above mentioned compounds required data on ADI and FAO Guidelines to calculate the theoretical maximum daily intake (TMDI) and estimated daily intake (EDI) for the tested compounds. Table 17 shows the average concentrations of the observed compounds.

НСВ	α-HCH	β-НСН	γ-НСН	p,p'-DDT	p,p'-DDD	p,p'-DDE	PCBs
		Mi	lk collected	on the 4 th d	ay		
0.0013± 0.0008	0.0002± 0.0002	0.0014± 0.0008	0.0002± 0.0001	0.0050± 0.0024	0.0009± 0.0003	0.0211± 0.0140	0.0076± 0.0041
			Matur	e milk			
0.0020± 0.0025	0.0005± 0.0017	0.0033± 0.0068	0.0004± 0.0014	0.0034± 0.0093	0.0004± 0.0025	0.0282± 0.0250	0.0544± 0.0814

 Table 17 Average concentrations of organochlorine compounds in human milk

 in Poland (mg/L milk ± SD) (31)

It is worth noting that higher concentrations of PCBs and b-HCH were reported for mature milk than for milk collected on the 4th day (statistically significant, P \leq 0.05). In Poland SDDTs and PCBs had the highest share in EDI by breast-fed infants. The relation between ADI, TMDI, and EDI was calculated only for Σ DDTs and Σ HCHs, as these were the only compounds for which ADI was identified. The above mentioned calculations were then used to estimate the safety margins for Σ DDTs and Σ HCHs. These amounted to 4.2 (ADI for Σ DDTs=20 µg/kg bw per day) and 13.3 (ADI for g-HCH=8 µg/kg bw per day), respectively. The estimated EDIs for Σ DDTs and Σ HCHs did not exceed the values assumed to be safe, that is, ADI and TMDI.

While the PCBs intake in the initial period of lactation (4th day) was relatively low and did not exceed the TMDI FAO/WAO guidelines, EDI of PCBs in the mature milk exceeded the relevant TMDI in some regions. Nevertheless, the average concentration of PCBs (1 mg/kg of fat) in human milk in Poland was lower than that found in highly developed countries. According to the literature sources, the infant average PCBs intake from human milk is 4.4 μ g/kg bw per day. The EDI of PCBs in this study averaged 2.8 μ g/kg bw per day.

These values of EDI for PCBs exceed the reference values of $1 \mu g/kg$ bw per day (FDA, US) or 0.6 $\mu g/kg$ bw per day proposed by the National Food Agency of Denmark. However, in the light of the recent toxicological research, there is no reason why the levels of organochlorines detected in human milk should provide grounds for altering the recommendations in favour of breast-feeding. The benefits of breast-feeding for the child far outweigh possible negative impacts of compounds found in human milk (28, 29, 31).

Czaja and co-workers have also studied the differences between more and less industrialised areas of Poland (29). Higher concentrations of HCB and DDT were reported in milk of women from more industrialised areas (Table 18). Among eight chlorinated hydrocarbons examined, the concentrations of PCBs and p,p'-DDE were always the highest. Statistically significant differences were found between the mean concentrations

	НСВ	α-HCH	β-нсн	ү-НСН	DDT	DDD	DDE	PCBs			
	More industrialised areas (N=158)										
x	0.0016	0.0004	0.0019	0.0005	0.0055	0.0012	0.0254	0.022			
SD	0.0012	0.001	0.0015	0.0014	0.0109	0.0033	0.0222	0.0447			
Min	0.0002	0.0002	0.0004	0.0002	0.0008	0.0005	0.0009	0.001			
Max	0.011	0.0108	0.0101	0.0156	0.1355	0.0381	0.1388	0.32			
			Less ir	ndustrialise	ed areas (I	N=199)					
x	0.0022	0.0007	0.0041	0.0004	0.0028	0.0006	0.0275	0.0131			
SD	0.0028	0.0018	0.008	0.0011	0.005	0.0004	0.0251	0.0145			
Min	0.0002	0.0002	0.0004	0.0002	0.0008	0.0005	0.0017	0.001			
Max	0.0152	0.0155	0.0835	0.0083	0.0409	0.0048	0.185	0.0741			

 Table 18
 Mean concentrations of organochlorine compounds in human breast milk collected from donors living in more and less industrialised areas of Poland (mg/L) (29)

X - mean concentration, SD - standard deviation

of b-HCH, HCB, p,p'-DDT, and PCBs examined in milk samples of women from more and less industrialised areas (P<0.05). In general, the concentrations of p,p'-DDT (banned in Poland 20 years ago) were consistently lower than those of its metabolite p,p'-DDE. The DDT/DDE ratio in the biological material dropped with time.

The β -HCH isomer showed the highest concentrations although its fraction in commercial lindane-containing pesticide products usually does not exceed 2–3%. The reason is that the excretion of the b-isomer from the human body takes five times longer than of other HCH isomers, and the ability of β -HCH to accumulate in fatty tissue is 10–30 times higher than of other isomers. The α -HCH and γ -HCH levels approximated the detection limit of the method.

Mean hexachlorobenzene concentrations were reported relatively low in human breast milk, comparable to those found in other European countries. Mean concentrations of PCBs were lower than those found in other countries (1 mg/kg of fat).

Bioavailibility of persistent organochlorine compounds found in human breast milk all over the world is very high, and their absorbtion in the infant's alimentary tract is almost complete (32). In the course of lactation, persistent organochlorine compounds dissolved in milk fat are removed from a woman's body. The infant is exposed to the compounds in question throughout the breast-feeding period.

The aim of this study was to identify possible trends in excretion of HCH isomers α - HCB, β - HCB, γ -HCB, p,p'-DDT, p,p'-DDD, p,p'-DDE, and polychlorinated biphenyls with human milk during lactation. The milk was sampled from eight lactating women from Warsaw (Table 19). The results showed individual differences in the excretion of the compounds. Subsequent milk samples collected from the same woman

	НСВ	-HCH	p,p'-DDT	p,p'-DDE	PCBs
Lowest concentration	0.0003±0.0002	<0.0002	0.0033±0.0019	0.0075±0.003	0.0039±0.0033
Highest concentration	0.0025±0.0008	0.005±0.0076	0.0129±0.0098	0.1387±0.1916	0.0384±0.0854

Table 19 Range of average concentrations in samples of eight donors (mg/L milk \pm SD) (28)

once a week differed markedly. The levels of compounds excreted with milk were reported to exceed significantly the daily intakes of those compounds by breast-feeding women. The findings of this study are not conclusive enough to claim that the infants' exposure to those compounds decreases or increases as breast-feeding continues. In light of the current knowledge, the advantages of natural breast-feeding outweigh the potential risk resulting from action of toxic compounds in human breast milk.

The concentrations of organochlorine compounds in breast milk depend on two processes: their lifetime bioaccumulation in the adipose tissue and the excretion with milk during lactation (33). Assuming that lactation can be an important process for eliminating organochlorine compounds from the human organism, an attempt was made to understand how successive childbirth(s) and lactation affect the organochlo-

rine compound levels detected in human milk. The excretion of the examined compounds during lactation is much higher than the intake of these compounds with the diet, which may imply that the tissue deposits of the organochlorine compounds in women decrease with consecutive lactation.

Literature gives different perspectives on the effect of successive lactation on levels of organochlorine compound in breast milk. Some decrease in DDE, HCHs, and PCB levels in the donors' milk following consecutive deliveries was reported. The basic factors affecting the concentrations of these compounds include the donors' age, number of children, and duration of breast-feeding after each childbirth.

Another study analysed breast milk samples of two consecutive lactations taken from two donors (33). The interval between the lactations was 8 months in the first donor and 2 years in the second. Milk samples were analysed for the presence of p,p'-DDT, p,p'-DDE, p,p'-DDD, α -, β -, γ -HCH, and Σ PCBs using gas chromatography with an electron capture detector. The donor with the shorter interval between lactations showed higher mean concentrations of the examined compounds in the milk of the first lactation than in that of the second (Figure 1). These variances were statistically significant for each examined compound.



Figure 1 Human breast milk OCP and PCB levels in a woman with the eight-month interval between two lactations (32)

The second donor with the two-year lactation interval showed a different distribution pattern of the examined compounds. Just as the first, the second donor showed higher mean levels of HCB, β -HCH, DDD, and PCBs during the first lactation (Figure 2), but the differences were not statistically significant. At the same time, the concentrations of DDT and DDE in the second lactation were slightly higher, with the variance being statistically significant. Also noted was the absence of a decrease in DDT and DDE levels in the second donor's milk during the second lactation. This may be related to a relatively fast migration of tissue deposits of these compounds during a longer (2-year) interval between lactations.



Figure 2 Human breast milk OCP and PCB levels in a woman with the two-year interval between two lactations (32)

The ability of a woman's organism to quickly activate tissue deposits of the organochlorine compounds has been confirmed in earlier studies (28) where mean concentrations of these compounds in primiparae and multiparae were compared, but also depended on the donors' age. No decrease in mean concentrations of the studied compounds was reported for multiparae versus primarae. Mean concentrations of HCB and Σ HCHs were similar in both donor groups, with DDT and PCBs being even higher for multiparae. Such results may be related to the age of the studied women. The number of deliveries did not affect mean concentrations of HCB and Σ HCHs. In women with the same number of children DDT, its metabolites, and PCBs increased with age. Other authors reported similar results showing that organochlorine compound concentrations in breast milk increase with the donor's age.

Age, more than the number of deliveries, seems to affect the concentrations of organochlorine compounds in breast milk. This phenomenon may imply that the deposits of these compounds recover quickly in women's tissues after lactation. This is confirmed by the results of the study in which higher DDT and DDE concentrations were detected in the second lactation milk of a woman with the longer interval between lactations.

Adipose tissue of inhabitants from Gdaňsk, located at the southern coast of the Baltic Sea, and from the province of Skierniewice of the inland Poland were investigated for PCB congeners using HRGC-HRMS technique and for OCCs using HRGC- ECD. The samples were collected during post-mortem from randomly selected donors in Gdaňsk in 1990 and in the province of Skierniewice in 1979. The older data showed high concentrations of DDTs and total PCBs and much lower concentrations of HCH, HCB, and chlordane isomers (27, 34).

PCBs	Range of concentration (ng/kg fat wt)	TEF	Range of TEQ (ng/kg fat wt)	Mean of TEQ
77	54–500	0.01	0.54–5.0	2.3
126	41-850	0.10	4.1–85	34
169	50-390	0.05	2.5–27	13
Total	145–1,740		7.1–107.3	49.3

Table 20	Range of levels of non-ortho coplanal	r PCBs and range of their 2378-TCDD TEQ in humai	n
	tissues from two localities in Poland (Gdaòsk 1990 and Skierniewice 1979) (27)	

PCB-153 was a high contributor of the congener occupying 23% of the total PCB content, and together with PCB-138 (18%) and PCB-180 (13%) were the most prevalent members. Samples taken in Gdaňsk in 1990 contained $1.5\pm1.3 \ \mu g/g$ of total PCBs on a fat basis while the 1979 samples from the inland province contained $1.2\pm0.4 \ \mu g/g$, which seemed to indicate persistent PCB exposure in Poland.

Randomly selected post-mortem samples of liver cancer taken in Gdansk contained 4.7 μ g/g of PCBs, while all other samples showed 0.75–1.9 μ g/g. TCDD toxic equivalent of 13 detectable coplanar members of PCBs in adipose tissues from Gdaňsk and Skierniewice was 210 pg/g and 190 pg/g on a lipid weight basis, respectively, including 45 pg/g and 59 pg/g of non-ortho, 142 pg/g and 110 pg/g of mono-ortho and 24 pg/g and 16 pg/g of di-ortho chlorobiphenyls, respectively. A fingerprint of chlorobiphenyl composition in the examined samples was virtually the same in the 1990 Gdansk human adipose tissue and the 1979 Skierniewice tissue, in spite of geographic differences and the sampling interval.

The baseline non-ortho coplanar PCB concentrations in human adipose tissue from Poland for the two localities were 10.10–20.

The recorded concentrations were equal and higher than those reported in other countries including the Czech and Slovak Republics. Both sites had comparable levels. This finding suggested a slower elimination rate of coplanar congeners and/or a continuing exposure of Polish population to PCBs.

Tanabe and Falandysz (34) also compared the previous data on organochlorine contamination in human adipose tissue of Polish cadavers reported by various researchers so far. Concentrations of PCBs were found somewhat higher than those reported earlier in other regions of Poland (Table 21). This was probably due to identification and determination of large numbers of PCB components using capillary GC in this investigation. Other organochlorine insecticide levels were rather comparable among various study periods reported for various regions in Poland. The results showed that DDT levels in the Polish environment and biota were declining at a slower rate after its withdrawal, and that the existing levels in Polish human fat were

Pollutant	Warsawa	Skierniewice	Poznaň	Gdaňsk
	1977–8 (100)	1979 (12)	1980s (53)	1990 (9)
PCBs	0.19	1.2±0.44	NA	1.5±1.3
p,p'-DDE	3.330±0.199	22±16	14.095±15.798	15±13
p,p'-DDD	0.049±0.013	0.67±0.57	NA	0.013±0.007
o,p'-DDT	0.082±0.008	0.060±0.063	0.433±0.369	0.0098±0.0064
p,p'-DDT	1.011±0.078	2.0±1.2	1.772±1.767	0.67±0.38
DDTs	4.470±0.276	25±16	16.30±17.93	15±13
α-HCH	0.008±0.001	0.040±0.026	0.013±0.003	0.007±0.003
β-HCH	0.148±0.017	0.42±0.13	0.211±0.154	0.23±0.09
γ-HCH	0.022±0.003	0.068±0.025	0.020±0.018	0.004±0.002
δ-HCH	NA	0.0045±0.0057	0.045±0.025	0.002±0.014
HCHs	0.216±0.015	0.53±0.17	0.328	0.25±0.09
HCB	NA	0.36±0.22	0.221±0.143	0.26±0.23
Chlordanes	NA	0.11±0.08	NA	0.06±0.03

Table 21	Comparison of concentrations (means \pm SD: $2\mu g/g$ lipid weight) reported in human fats in
	Poland (the number of samples is given in parenthesis) (34)

still higher on the international level. On the other hand, human PCB contamination corresponds to that of other industrialised nations.

Croatia

Croatia also conducted investigations of organochlorine pesticide residues and PCBs in human milk (35). The first evaluation of OCCs in breast milk took place in midseventies. For the last two decades OCCs and PCBs have been observed to drop in human milk samples collected in the city area of Zagreb. The first analyses of these pollutants in breast milk from two small towns in the Northern Adriatic area were carried out in 1986/87 and 1989. Both locations were approximately 50 km away from the city of Rijeka, one situated in the nearby Istrian peninsula, while the other on the island of Krk. *Frković and co-workers* (35) described OCC and PCB levels in 31 breast milk samples collected in Clinical Hospital of Rijeka.

The organochlorine pesticide levels determined in human milk from nursing women living in the Northern Adriatic region were the lowest in Croatia since the beginning of their survey in mid-seventies. Such decline could be a part of an overall tendency observed in other countries as well. Contrary to OCCs, PCB levels in breast milk were the highest ever observed in Croatia, a fact that was hard to explain at the time. Although PCB use has been restricted, there were some sources of PCBs in the environment. No statistically significant difference in OCC and PCB levels in human milk was seen between the donors' age, parity, residence, smoking habits, weight during pregnancy, and milk fat content. Good correlation was obtained between OCCs and PCBs (r=0.527, P=0.05), indicating the similar fate of those lipophilic compounds in the human body.

Generally, the levels of organochlorine pesticide residues in the breast milk from women living in that area were 10–50 times lower than was reported earlier and were comparable to the lowest levels observed in Italy and Canada. This could be the result of a constant drop observed in Croatia over the 20 years.

High levels of PCBs were found in human milk from industrialised countries, while they mostly kept below the detection limit in milk from developing countries. Average concentrations of PCBs in human milk were typically between 500 and 2,000 μ g/kg milk fat (36). The mean level of PCBs (such as Aroclor 1260) reported by *Frković* (35) was 898 μ g/kg milk fat and was the highest reported in Croatia. Compared to earlier results from the Nothern Adriatic region, the median value (778 μ g/kg milk fat) was 56% higher than in samples from Istria location, and three times higher than in samples from Krk.

OCCs in human milk were analysed in samples collected over nine years (1987– 1995) from 139 nursing mothers whose children were hospitalised for various disorders at the Department of Pediatrics, Clinical Hospital Centre in Zagreb, Croatia (37). Mothers were not occupationally or accidentally exposed to organochlorine pesticides or PCBs.

All samples contained p,p'-DDE and PCBs; the median concentrations were 318 μ g/kg milk fat and 220 μ g/kg milk fat, respectively. Higher levels were found in mothers (N=12) nursing neonates with impaired neurodevelopmental competencies or an inappropriate arousal reaction. No difference was observed between mothers nursing children with respiratory or gastrointestinal diseases, urinary tract infections or other infectious diseases, anaemias, prolonged neonatal hyperbilirubinaemias or when children were with dermatological findings, congenital malformations or healthy.

Levels of OCPs in human serum samples collected in Croatia have been monitored since 1975 and the levels of total PCBs since 1985. However, no data have been available on the content of individual PCB congeners (38). That is why samples of human blood serum were collected from one group of 14 donors (3 men and 11 women; between14 and 83 years old) from the general population and from another group of occupationally exposed workers (15 samples) employed between 3 months and 28 years in repairing transformers and capacitors. All donors were residents of Zagreb, Croatia. Six indicator PCB congeners (28, 52, 101, 138, 153, and 180) and some OCPs (HCB, HCHs, DDTs) were determined.

Table 22 shows median concentrations of the analysed organochlorines. All serum samples contained PCB-138, PCB-153, HCB, and p,p'-DDE. The six PCB conge-

Compound	General population (N=14)	Exposed workers (N=15)	Compound	General population (N=14)	Exposed workers (N=15)
PCB-28 0 PCB-52 0 PCB-101 0 PCB-138 0 PCB-153 0 PCB-180 0 ΣPCB congener 7 Total PCBs 1	0.1 (11) 0.7 (14) 0.4 (13) 0.5 (14) 0.5 (14) 0.3 (14) 2.4 (14) NA	0.4 (10) 1.6 (14) 0.6 (11) 0.9 (15) 1.3 (15) 0.9 (14) 6.6 (15) 9 (15)	HCB α -HCH β -HCH γ -HCH p,p'-DDE p,p'-DDD p,p'-DDT o,p'-DDT	0.3 (14) 0 (0) 1.2 (14) 0.3 (14) 3.4 (14) 0.2 (10) 0.6 (14) 0 (6)	$\begin{array}{c} 0.2 \ (15) \\ 0 \ (3) \\ 0.5 \ (12) \\ 0.3 \ (14) \\ 4.9 \ (15) \\ 0 \ (0) \\ 0 \ (0) \\ 0 \ (0) \end{array}$

Table 22 Concentrations (median, µg/L serum) of PCBs and OCPs in human serum samples collected in Zagreb, Croatia (the number of positive samples is given in parenthesis) (37)

ners were present at higher levels in the exposed workers than in the general population, but there was no difference between the two groups concerning the concentration of OCPs. All levels of OCPs and PCBs were fully within the concentration range found for the same compounds in the general population over the past ten years. The distribution pattern of PCB congeners in human serum and human milk was shown to correlate well with Aroclor 1260 (39). The sum of the six PCB congeners found in the analysed serum samples was lower than the total PCB content determined with Aroclor 1260. The same was found for human milk samples collected in Zagreb in the same year (40). This indicates that more than six analysed PCB congeners should be determined in order to assess the total PCB body burden.

Estonia

Studies concerning the concentration of OCPs in human breast milk were started in Estonia in 1971, but data about the content of PCBs in human breast milk were missing. The determination of PCBs in the breast milk of Estonian women was per-

Year		DDE x 100 (%)			
	DDT	DDE	ΣDDT	PCBs	ΣDDT
1971	0.026 0.004–0.050	0.099 0.021–0.230	0.125	-	79
1974	0.021 0.002–0.080	0.063 0.008–0.180	0.084	-	75
1984	-	-	0.006 0.003–0.011	0.012 0.006–0.017	95

Table 23 Concentrations of OCPs and PCBs in Estonian human milk (41)

formed in the 80s and the results were compared with other Baltic Sea countries (41). Table 23 shows the levels of OCPs and PCBs in Estonian human milk.

During the years 1974–1984, decomposition of DDT into its main metabolite DDE increased in the human breast milk from 79% to 95%. As DDE is less toxic to human organism than DDT, in 1984 much less OCPs and less toxic compounds got into the organism of newborn children than between 1971 and 1974 (41).

The average OCP intake of a newborn child, weighing 5 kg and consuming 1 kg of milk a day, is 0.001 mg/kg of total DDT and 0.002 mg/kg of PCBs, that is, 5–20 times less of DDT than in the beginning of seventies. Six years after the total ban of DDT in Estonia in 1968, the content of its metabolite DDE in human milk rose up to 75–79%. The amounts of DDT and DDE consumed by newborn children in the first week of life were 5.6 μ g of DDT and 15.9 μ g of DDE a day. Two to four weeks after the birth, the respective amounts rose to 11.2 μ g and 35.4 μ g a day.

It was found that in mid-80s the average daily intake of total DDTs and PCBs by newborn children did not exceed the ADI proposed by WHO (for DDTs 0.05 mg/kg

and for PCBs concentration 0.07 mg/kg per day, the exceeding of which caused the <code>"Yusho"</code> disease in Japan in 1968 and in Taiwan in 1979).</code>

The potential human exposure of DDTs and PCBs through diet was also studied (41). Some authors claimed that in the Baltic Sea countries human organism received 90% of toxic compounds from fish. By the end 60s, the average DDT daily intake through diet by the residents of Estonia was 0.036 mg. According to certain data, the concentration of DDT in fish reached its peak 10–11 years after the maximum usage of DDT (in Estonia 1965).

If an average 60-kg Estonian ate 30–50 g of fish a day and if the average concentration of biphenyls and DDT in fish is not higher than it was in mid-80s (0.2 mg/ kg and 0.1 mg/kg), then the concentration of DDTs and PCBs in the daily nutrition of Estonian residents was 3–5 μ g/day and 6–10 μ g/day, respectively (that is, if the herring accounted for the main part of the fish diet), that is 0.05–0.08 μ g/kg of DDT a day and 0.10–0.17 μ g/kg of PCBs a day. Similar results have been obtained in Finland and Sweden (41).

In the beginning of the 80s, the content of DDT in the daily nutrition was 10–75 μ g in the former Soviet Union,2–145 μ g in the USA,27–44 μ g in Great Britain, 18 μ g in Canada, 52 μ g in Italy, 60 μ g in the Netherlands, and 60 μ g in Bulgaria. At the same time, the content of total DDTs and PCBs in the Baltic fish ranged from 0.017–0.030 mg/kg and from 0.035–0.060 mg/kg, respectively. Daily concentrations of DDTs and PCBs in fish diet decreased to 0.5–1.5 μ g/day and 1.1–3.0 μ g/day respectively, or 0.008–0.025 μ g/kg of DDTs a day and 0.018–0.050 μ g/kg of PCBs a day.

These data and an overall no-effect level for toxicity of DDT of 0.25 mg/kg body weight per day in humans suggest that the daily intake on this scale would not involve human cancer risk. Thus, if a 60-kg person consumed 30–50 g/day of fish containing 17–30 μ g/kg of DDT and its metabolites, the maximal daily intake would be 0.025 μ g/kg body weight. This would constitute 1/800 of the ADI and 1/12,000 of the lowest dose of non-metabolised DDT which caused liver tumours in male mice of the most sensitive strain tested (41).

Polychlorinated dibenzo-p-dioxins and dibenzofurans

Although the world was engaged in professional discussion about the sources of PCDDs/Fs, their fate in the environment, and their influence and risk to various organisms in the CEE countries, concrete information about contamination of human tissues practically does not exist (42). From 1965 to 1968, 80 workers who had worked with 2,4,5-sodium trichlorophenoxyacetate and butylester of trichlorophenoxy-acetate acid became ill (43). The cause of the illness was 2,3,7,8-tetrachlorodibenzo-p-dioxin. This intoxication occurred during the time when alkaline hydrolysis of tretrachlorobenzene at atmospheric pressure was used to increase and shorten the reaction time, and probably during the same time the mother liquor, which was originally disposed of, was put back into production. The concentration of TCDD in the work atmosphere was never measured; however, TCDD was found in final product – Arboricide E – and several years later, was also found in the building and on wall paintings. This contamination is still measurable. It was something as small »Seveso« in the former Czechoslovakia. A 10-year study was conducted for 55 exposed individuals. Most patients developed chloracne and 11 manifested porphyria cutanea tarda. Approximately one-

half of the patients suffered from metabolic disturbances, i.e. pathologically elevated lipids with abnormalities in the lipoprotein spectrum, and two-fifths of the patients had pathological changes in the glucose tolerance test. One third of the patients had biochemical deviations indicative of a mild liver lesion. Histological examination revealed light steatosis, or periportal fibrosis, or activation of Kupffer cells. In 17 persons symptoms of nervous system focal damage existed, with predominance of peripheral neuron lesion of the lower extremities. Most patients suffered from various psychological disorders. By 1981 when the results were published, two patients died of bronchogenic lung carcinoma; one of liver cirrhosis; one of a rapidly developed, extremely unusual type of atherosclerosis precipu cerebri, and patients have died in traffic accidents. The conditions of most other patients improved. The measurements of air in the working hall and the content of PCDDs/Fs in walls and soil, were performed a few years ago, but results have never been published.

Czech and Slovak scientist carried out preliminary, informative measurements of contamination of human tissues by PCDDs/Fs and PCBs-dioxin in co-operation with people from CDC Atlanta, USA (42). Samples of human adipose tissue from two different sites of the former Czechoslovakia were used. Seven samples (from Prague and Michalovce) were used to determine PCDDs/Fs, PCBs and HCB, HCHs, and DDTs. The results of PCB and OCC determination confirmed the trends described by *Schoula and co-workers* (4, 11) concerning the higher level of contamination in Michalovce region. The level of planar PCBs were lower than in Poland (44).

Only 2,3,7,8-substituted congeners of PCDDs and PCDFs, especially hepta- and octachlorinated in adipose tissue, were found. This was in agreement with previous studies (45). While there were some differences in congener patterns from some countries (46), the general pattern and levels were similar to previous CDC studies (47). The I-TEQ values for the coplanar PCBs made a major contribution to the total TEQs in these samples which were similar to reports from Japan and Sweden (46). The percentage of PCDF contribution to the total TEQ in these samples was 2–3 times higher than the PCDD contribution, which was very different from data from other countries. The high I-TEQ values for the PCDFs in these samples from the Czech and Slovak Republics were due to the relatively high levels of 2,3,4,7,8-pen-taCDF (range 21–44 ng/kg). The average concentration of 2,3,7,8-TCDD was 1.9 pg/g, which is lower than in other industrialised countries. Relatively high concentrations of octa-CDD (100 to 460 ng/kg) were found in human tissues, having in mind that the

Table 24 Range of concentrations of PCDDs/Fs, PCBs, and OCCs in human adipose tissue from the Czech and Slovak Republics (N=7) (42)

I-TEQ (ng/kg)		Total I-TEQ	ΣHCHs	НСВ	ΣDDTs	ΣPCBs	
PCDDs	PCDFs	Planar PCBs	(ng/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)
6.4–10.7	12.0–25.2	6.0–32.6	31.3–59.4	0.039–0.251	0.253–3.395	0.565–9.966	1.157–3.525

ΣHCHs – sum of isomers α -HCH, β -HCH, and γ -HCHs

ΣDDT – sum of isomers and metabolites of DDT (o,p'-DDT, p,p'-DDE, o,p'-DDD, p,p'-DDD, p,p'-DDT)

isomer was found in low concentrations in pork and beef. This suggests another source on contamination (Table 24).

WHO/EURO measured the levels of PCBs and PCDDs/Fs in human milk in various European countries including the CEE region. The values of TEQ in pg/g fat for PCDDs/Fs were 18.4 pg TEQ/g fat in Uherské Hradiště and 12.1 pg TEQ/g fat in samples from Kladno. In Slovakia, the values of TEQ were 15.1 pg TEQ/g fat in district Michalovce and 12.6 pg TEQ/g fat in Nitra.

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Sažetak

PERZISTENTNI, BIOAKUMULATIVNI I TOKSIČNI SPOJEVI U ZEMLJAMA SREDNJE I ISTOČNE EUROPE – NAJSUVREMENIJI IZVJEŠTAJ – IZLOŽENOST LJUDI

Ovaj pregledni rad dio je opsežna izvještaja koji opisuje probleme vezane uz perzistentne, bioakumulativne i toksične spojeve (PBT) u srednjoj i istočnoj Europi. Opisani su problemi izloženosti ljudi u zemljama srednje i istočne Europe perzistentnim i bioakumulativnim organskim spojevima koji imaju toksična svojstva, a koji mogu uzrokovati nepovoljne učinke na zdravlje ljudi.

Ključne riječi: bioakumulativni spojevi, izloženost ljudi, perzistentni spojevi, toksični spojevi

Requests for reprints:

Ivan Holoubek RECETOX-TOCOEN & Associates Veslarská 230B, 637 00 Brno, Czech Republic E-mail: *holoubek@chemi.muni.cz*