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PCB Congeners, Hexachlorobenzene, and Organochlorine Insecticides in Human Fat in Italy

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The identification and quantification of environmental pollutants and their metabolites in human tissues are very important in determining the extent of exposure to these compounds and in evaluating the hazard to humans. Chlorinated hydrocarbons, particularly polychlorinated biphenyls and organochlorine pesticides have played an important role in the last years since studies have shown the extreme persistence of these contaminants in the environment, their accumulation in human tissues, and their toxicity (Risebrough et al., 1968; Biros et al., 1970; Bleavins and Aulerich, 1983; Splinder, 1983; Jensen, 1984).

The presence of chlorinated hydrocarbons in humans in Italy has been shown by recent papers on this subject (Leoni and Biocca, 1982; Focardi et al., 1984). The aim of this paper is to contribute to our knowledge of the levels of these contaminants in human adipose tissue in Italy, and to identify the PCB isomers involved.

MATERIALS AND METHODS

Subcutaneous adipose tissues were obtained from patients admitted to the Clinic of Surgical Semeiotics of the University of Siena for general surgery in 1983 and 1984. Samples were brought to the laboratory at a temperature of about 4°C, frozen (-20°C), freeze-dried and then broken up. The residual water content in the freeze-dried material was evaluated in subsamples (24h at 110°C).

For the analysis about 0.5 g of the freeze-dried homogeneous material was extracted for 12h in Soxhlet with n-hexane. The extract was subjected to sulfuric acid cleanup (Murphy, 1972) followed by Florisil chromatography. The eluates were analysed with Perkin Elmer Sigma 3 and F 22 gas chromatographs equipped with Ni63 Electron Capture Detectors. Glass columns (200 cm long, 0.3 cm

i.d.) packed with 4% SE-30 + 6% SP-2401 on Supelcoport (100-120 mesh) or with 5% OV-101 on Chromosorb W HP (80-100 mesh) were used. A 50-m-long fused silica capillary column covered with SE-54 was also used.

Calculations were based on standard reference solutions (Aroclor 1254, Aroclor 1260 and single isomers for PCB) after the losses had been evaluated. The capillary chromatograms of typical human adipose tissue and standard for PCB are reported in Figure 1.

RESULTS AND DISCUSSION

The residue levels of organochlorines in the samples are shown in Table 1; all the results are expressed as μg/g dry weight.
The most prevalent compounds are pp'DDE, hexachlorobenzene (HCB) and polychlorinated biphenyls (PCBs); other organochlorines that occurred in all the samples were lindane and pp'DDT. Of these contaminants DDE had the highest average residue concentration (7.3 ppm), followed by HCB (2.2 ppm), PCBs (1.7 ppm), DDT (0.8 ppm) and lindane (0.12 ppm); all these concentrations are the highest yet recorded for Italy (Leoni and Biocca,1982; Minak et al., 1982; Focardi et al., 1984). In particular these figures reveal a marked increase in the concentrations of the PCBs (which are quite regularly distributed) and very high values of hexachlorobenzene even when compared with world-wide literature (Bjorseth et al., 1977; Mes et al., 1982; Jan, 1983; Mori et al., 1983; Peattie et al., 1984).

Table 1. Organochlorines (μg/g dry weight) in adipose tissues. (n=26; $\bar{x}$=mean; S.D.=Standard Deviation; Age=years; PCBs calculated as Aroclor 1260).

	Age	HCB	γ-HCH	pp'DDE	pp'DDT	PCBs	PCBs/DDTs
$\bar{x}$	55	2.26	0.128	7.35	0.83	1.75	0.33
S.D.	16	2.41	0.079	5.36	0.60	0.59	0.23

A strong correlation ($p<0.0005$) exists between DDE and DDT, the equation being

$$y = 0.107\ x + 0.039 \quad (r=0.961)$$

where y is the pp'DDT concentration expressed in μg/g and x is the pp'DDE concentration expressed in μg/g.
The concentration levels for each compound were plotted against the

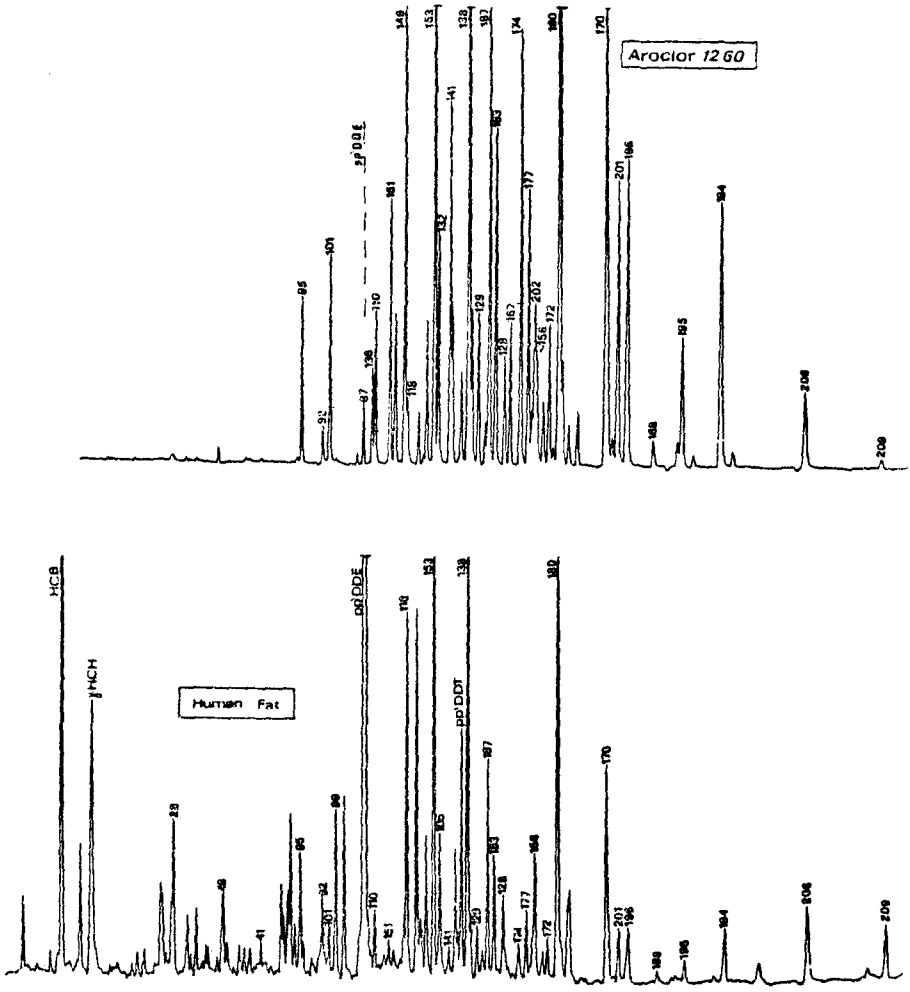

Figure 1. Temperature programmed capillary column ECD chromatogram of a standard (Aroclor 1260) and a sample of human fat. Chromatographic conditions as follow: carrier gas Argon/Methane (95/5) 100KPa; detector 300°C; injector 280°C; temperature programme, isothermal phases at 100°C (2 min) and 280°C (10 min), with intermediate temperature increase rate of 3°C/min.

age of the subjects, showing DDE and DDT to be positively correlated with age ($p<0.05$ both) while the PCBs/DDTs ratio is negatively correlated with it ($p<0.02$). This is consistent with the more recent diffusion of PCB with respect to DDT and derivates.

The concentrations of all the organochlorines were found to be significantly higher in 1984 than in 1983 (Table 2). However it is not possible to evaluate the influence of the different average age of the two samples (higher in 1984 samples than in those of 1983) on this increase.

Table 2. Chlorinated hydrocarbons (μg/g d.w.) in the two years.

		Age	HCB	γ-HCH	pp'DDE	pp'DDT	PCBs	PCBs/DDT
1983	$\bar{x}$	45	1.29	0.089	5.93	0.79	1.52	0.41
n=9	S.D.	13	0.81	0.011	5.33	0.75	0.34	0.28
1984	$\bar{x}$	60	2.78	0.147	8.10	0.85	1.85	0.29
n=17	S.D.	16	2.82	0.081	5.34	0.52	0.67	0.19

Human fat samples were next examined for sex-related differences; Table 3 shows the average of the five compounds in question. All the chlorinated hydrocarbons showed higher values in females than in males, but the only compounds that show significant differences are lindane ($p<0.02$), DDE ($p<0.01$) and DDT ($p<0.002$).

Table 3. Organochlorines (μg/g d.w.) in males and females.

		Age	HCB	γ-HCH	pp'DDE	pp'DDT	PCBs	PCBs/DDTs
Males	$\bar{x}$	53	1.73	0.099	5.14	0.50	1.72	0.40
n=14	S.D.	17	1.41	0.057	3.17	0.39	0.56	0.22
Females	$\bar{x}$	57	2.84	0.162	9.94	1.13	1.79	0.25
n=12	S.D.	15	3.18	0.089	6.32	0.71	0.64	0.22

The composition of PCB in adipose tissuee does not vary appreciabily from one sample to another; the PCB congeners and isomers identified are mostly penta-, hexa-, hepta- and octachlorobiphenyls (Table 4; Figure 1).
As far as the factors that determine the uptake, distribution and elimination of individual PCB isomers are concerned, many Authors (De Freitas and Norstrom, 1974; Burse et al., 1976; Yakushiji et

Table 4. PCB isomers and congeners identified.

	IUPAC number		IUPAC number
Trichlorobiphenyls		Heptachlorobiphenyls	
2,4,4'	28	2,2',3,3',4,4',5	170
		2,2',3,3',4,5,5'	172
Tetrachlorobiphenyls		2,2',3,3',4,5,6'	174
2,2',3,4	41	2,2',3,3',4',5,6	177
2,2',4,5	49	2,2',3,4,4',5,5'	180
		2,2',3,4,4',5',6	183
Pentachlorobiphenyls		2,2',3,4',5,5',6	187
2,2',3,5,5'	92	2,3,3',4,4',5,5'	189
2,2',3,5',6	95		
2,2',4,4',5	99	Octachlorobiphenyls	
2,2',4,5,5'	101	2,2',3,3',4,4',5,5'	194
2,3,3',4,4'	105	2,2',3,3',4,4',5,6	195
2,3,3',4',6'	110	2,2',3,3',4,4',5',6	196
2,3',4,4',5	118	2,2',3,3',4',5,5',6	201
Hexachlorobiphenyls		Nonachlorobiphenyls	
2,2',3,3',4,4'	128	2,2',3,3',4,4',5,5',6	206
2,2',3,3',4,5	129		
2,2',3,4,4',5'	138	Decachlorobiphenyls	209
2,2',3,4,5,5'	141		
2,2',3,5,5',6	151		
2,2',4,4',5,5'	153		
2,3,3',4,4',5	156		

al., 1978; Mattheus and Tuey, 1980; Safe, 1980) report that most of the lower chlorinated di-, tri- and tetrachloro isomers and selected higher chlorinated PCB, are preferentially eliminated from body tissues and organs; other studies (Kuroki and Masuda, 1977; Wolfe et al., 1982; Bush et al., 1984) suggest that the PCB isomers can be divided into two groups: persistent and nonpersistent isomers.

Our results show that most of the residues (about 60%) included only five distinct components: 2,3',4,4',5, 2,2',3,4,4',5', 2,2',4,4',5,5', 2,2',3,3',4,4',5 and 2,2',3,4,4',5,5', which have the IUPAC numbers 118, 138, 153, 170 and 180 (I.O.C., 1984). According to Bush et al. (1984) and Yakushiji et al. (1984) in recent experiments, the more persistent congeners in these samples of human fat seem to have the 2,4,5 chlorine substitution pattern in one ring and 4 or 2,4 in the other ring of the biphenyl molecule.

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