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Organochlorine Pesticide Contaminants in Human Adipose Tissue Collected in Tebriz (Iran)

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Organochlorine pesticides (OCP) have recevied considerable attention in the last decade since studies have shown extreme persistence of these pollutants in the world-wide environment & accumulation in human tissues(Tanabe et al., 1983; Jensen, 1983; Kutz et al., 1991). For these reasons, during the 1970s, many countries have restricted or banned the use of these chemicals. Thus, in Iran, the use of most of the persistent pesticides, except Lindane (γ-HCH) which is still certified for plant protection, has been prohibited in agriculture since 1985. National Human Milk and Adipose Tissue Monitoring Programs have been used in many countries for assessing levels of environmental pollution by lipophilic OCP compounds in different areas within or between countries and for developing time trends(Karakaya & Özalp, 1987; Karakaya et al.,1987;Skaare et al.,1988;Robinson et al.,1990; Mes et al.,1990;Loganathan et al.,1993;Burgaz et al., 1994).

Although there has been increased concern about the pesticide burden of the human body, only a few reports pertaining to the levels of OCP contaminants in breast milk and adipose fat are available from Iran (Hashemy-Tonkabony & Fateminassab,1977, Hashemy-Tonkabony & Soleimani-Amiri,1978). The aim of this study is to estimate the levels of OCP contaminants in human adipose tissue samples collected in Tebriz(Iran) during 1991-1992 and to investigate whether alterations in levels of these contaminants have occurred since 1974 in Iran.

MATERIALS AND METHODS

Human adipose tissue samples were taken during surgical operations in Tebriz State Hospital from 61 subjects, aged between 14 and 65, with a mean of 36.5. All subjects participated in the study voluntarily and agreed with its aims and all had lived in the Tebriz

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area during the last 5 years. Apart from this selection criteria, they were selected at random. Each survey patient had been interviewed to obtain information regarding sex, age, weight, height, number of children (women), smoking and dietary habits.

Tebriz samples consist of 8 men and 53 women. Most of the donors except four were nonsmokers. There was insufficient data on donors' body mass index. Of the mothers, 7 had one, 6 had two children and 32 were multiparas (more than 3). Those women without children were 8. All the subjects were mixed food consumers. The adipose tissue samples were frozen (-20°C) immediately until analysis.

Standards of HCB, α-HCH, β-HCH, γ-HCH, pp'DDE, pp'DDT, op'DDE were obtained from U.S.Environmental Protection Agency (EPA).

100 mg samples were ground with anhydrous sodium sulphate by using a tissue grinder. op'DDE, confirmed not be present in samples, was used as an internal standard and also added to the adipose tissue before extraction. Then tissue samples were extracted with 4x2x2 ml of n-hexane. The combined n-hexane extract was evaporated to dryness to determine extractable fat content(Smrek&Needham,1982). Using 8 ml n-hexane as the solvent, the sample was cleaned up with 1-2 ml of conc. sulphuric acid (Murphy,1972; Wickström et al.,1983).

Gas chromatography analysis was done on a Packard 438 equipped with ^{63}Ni-EC detector. The column used for quantitation was a 25 m x 0.25 mm fused silica capillary column cp Sil 8 from Chrompack. The operating conditions were: injector temperature 260°C; detector 320°C; column 80°C initial with 1 min. hold 10°C/min to 280°C. A 25 m long glass capillary column cp Sil 5 was also used. Peak heights were used as the basis for quantification. Residue levels are expressed as mg/kg extracted fat (ppm). Recoveries from a fortified sample at a 0.2 ppm each level were in the range of 80.3-92.1 % by this method, including internal standard. Results were not corrected for the percentage recovery.

The different sets of data were examined for significant difference ($p<0.05$) by the Mann-Whitney test. Spearman rank correlation was calculated to measure association between residues.

RESULTS AND DISCUSSION

The residue levels of OCP are shown in Table 1. It can be seen that residues of HCB, β-HCH, and pp'DDE are found to be major contaminants in the samples analyzed. Most of the OCP compounds except for pp'DDE and ΣDDT,

show higher values in females than males, but, there are too few results for males to compare this group with female subjects. Similar disparities have been observed in other studies(Mes et al., 1982; Abbott et al., 1981; Ferrer et al.,1992, Kutz et al., 1991).

Table 1. Chlorinated hydrocarbon residues in adipose tissue of Tebriz citizens (The mean levels expressed as mg/kg extracted fat basis)

Compound	Mean (S.D.)	Range	Median	Frequency of residue (%)
HCB	0.055 (0.11)	(ND[a]-0.875)	0.038	99.9
α-HCH	0.021 (0.12)	(ND-0.905)	<ND	11.5
β-HCH	0.728 (0.48)	(0.104-2.328)	0.608	100.0
ɣ-HCH	0.018 (0.04)	(ND-0.267)	<ND	27.9
Σ HCH[b]	0.767 (0.52)	(0.104-2.371)	0.645	
pp'-DDE	2.450 (2.44)	(0.256-10.428)	1.442	100.00
pp'-DDT	0.190 (0.22)	(ND-1.504)	0.132	96.7
Σ DDT[c]	2.921 (2.88)	(0.314-13.132)	1.880	

[a]ND = under the limit of detection
[b]ΣHCH = α-HCH + β-HCH + ɣ-HCH
[c]ΣDDT = 1.115 x pp'DDE + pp'DDT

There is a trend towards higher OCP residues increasing with age. There are significant differences in pp'DDE, pp'DDT and HCB residue levels between 46 plus and 14-24, 25-35, 36-45 age groups, respectively. There are significant differences in β-HCH levels between the 14-24 and 25-35 and also 46 plus year age groups. These findings show that the higher body burden levels for the older population reflect their longer periods of exposure and bioaccumulation and our data are in concordance with those obtained by other authors(Kutz et al., 1991). Mother's milk contains considerable amounts of organochlorine pesticides. Thus, it is to be expected that breast feeding should decrease the amount of chlorinated hydrocarbons in the mother's adipose tissue (Jensen, 1983; Mussalo-Rauhamaa et al., 1984). In our study, no clear decreasing influence of the number of childbirths on the ΣDDT and ΣHCH concentrations was observed. Age dependence was more discernable, the ages of the mothers having 1 and 2 children were on average only 28 years old, whereas the average of mothers having 3 and more children was 39 years old.

Of particular interest is the presence of HCB in all of the tissue samples except one analyzed. HCB has not been used directly in agriculture in Iran, but, recent studies indicate that HCB is an industrial by-product and a contaminant in some pesticides, and may thus find its way into the environment(Tobin, 1986). There are also reports that HCH is converted biologically into HCB

in mammals (Menzie, 1986). By comparison with published data from other countries, the values we obtained for HCB are lower than those found in Italy (2.26 ppm), Spain (2.99 ppm), United Kingdom (0.11 ppm), and India (0.28 ppm) in which HCB has never been massively used as fungicide (Abbott et al., 1985; Focardi et al., 1986; Camps et al.,1989; Nair&Pillai,1989). HCB levels in present study are difficult to interpret because of its diverse and not well known sources. It is unfortunate that no previous data on HCB levels in human adipose tissue exist for several regions in Iran. In Iran, beginning from 1979, restrictions are imposed on the usage of DDT in agriculture, and since 1983 its use has been prohibited, however, DDT has only been used for malaria control. Technical Lindane (%25 γ-HCH) is widely used instead of DDT in this country.

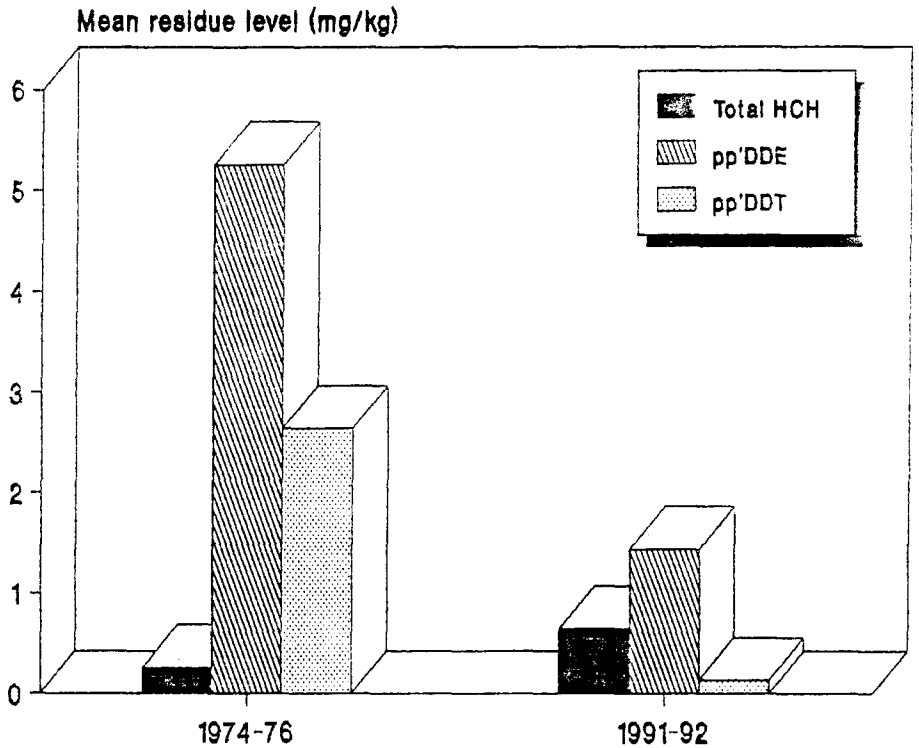

Figure 1. Mean levels of OCP residues in human adipose tissue from Iranian population over the period 1974-1992, based on the following data:

Year	Mean residue level (mg/kg)			References
	ΣHCH	pp'DDE	pp'DDT	
1974-76	0.26	5.26	2.64	Hashemy-Tonkabony & Soleimani-Amiri (1978)
1991-92	0.65[a]	1.44[a]	0.13[a]	Present study

[a]Median

Figure 1 shows the trends for the mean levels of the main residues, pp'DDE, pp'DDT and ΣHCH, over the period 1974-1992 that these studies have been carried out in Iran. It can be seen that there is a trend lower both pp'DDE and pp'DDT residues over 18 years period, even though differences in analytical methodology and statistical treatment make it difficult to directly compare the results of present study with those obtained in Iran by Hashemy-Tonkabony&Soleimani-Amiri (1978). The decreases in levels probably reflect the continuing prohibitions on the use of DDT by Iran. Over this same period total HCH residues appear to have increased about three times in Iranian adipose tissue samples (Figure 1). With continuing release into the environment, residue levels of HCH will probably increase, so, it deserves more attention.

In Table 2, the levels of pp'DDE and pp'DDT in human adipose tissue found in the present study and DDE/DDT ratios are compared with those of other countries as well as those of previous Iranian study. By comparison with published data from other countries, levels of pp'DDE and pp'DDT from Tebriz region are in line with those of most countries with the exception of India, where pp'DDT is probably still used in agriculture (Table 2). The main metabolite pp'DDE is more persistent in the environment than DDT. Thus, when the use of pp'DDT in a country ceases, the levels of this compound decrease more rapidly than the levels of pp'DDE, resulting, in an increasing DDE/DDT ratio (Slorach &

Table 2. Mean levels (mg/kg extracted fat) of pp'DDE and pp'DDT compounds in human adipose tissues collected in different countries

Country	Year	pp'DDE	pp'DDT	DDE/DDT	References
Canada	1979-81	3.26	0.16	20.3	Williams et al. (1984)
India	1982	9.39	9.66	0.97	Ramachandran et al.(1984)
Italy	1983-84	7.35	0.83	8.85	Focardi et al. (1986)
Turkey	1991-92	3.72	0.27	13.77	Burgaz et al. (1994)
United Kingdom	1982-83	1.3	0.11	11.81	Abbott et al. (1985)
Iran	1974-76	5.26	2.64	1.19	Hashemy-Tonkabony & Soleimani-Amiri (1978)
Iran	1991-92	1.44[a]	0.13[a]	11.08	Present study

[a]Median

Vaz,1983). In our study, DDT and DDE correlated significantly ($r=0.679$, $p<0.05$) and DDE/DDT ratio is increased from 1.91 to 11.08 in Iranian samples and this ratio in present study is as high as those found in most western countries where DDT use has been prohibited since 1970's (Table 2). So, our data provide sufficient indications of the effectiveness of the regulatory actions adopted in Iran.

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REFERENCES

Abbott DC, Collins GB, Goulding R, Hoodless RA (1981). Organochlorine pesticide residues in human fat in the United Kingdom. 1976-77. Brit Med J 283: 1425-1428

Abbott DC, Goulding R, Holmes DC, Hoodless RA (1985). Organochlorine pesticide residues in human fat in the United Kingdom, 1982-1983. Human Toxicol 4: 435-445

Burgaz S, Afkham BL, Karakaya AE (1994). Organochlorine pesticide contaminants in human adipose tissue collected in Ankara (Turkey) 1991-1992. Bull Environ Contam Toxicol 53:(in press)

Camps M, Planas J, Gòmez-Catalàn J, Sabroso M, To-Figueras J, Corbella J (1989). Organochlorine residues in human adipose in Spain: Study of an agrarian area. Bull Environ Contam Toxicol 42: 195-201

Ferrer A, Bona MA, Castellano M, To-Figueras J, Brunet M (1992). Organochlorine residues in human adipose tissue of the population of Zaragoza (Spain). Bull Environ Contam Toxicol 48: 561-566

Focardi S, Fossi C, Leonzio C, Romei R (1986). PCB congeners hexachlorobenzene and organochlorine insecticides in human fat in Italy. Bull Environ Contam Toxicol 36: 644-650

Hashemy-Tonkabony SE, Fateminassab F, (1977). Chlorinated pesticide residues in milk of Iranian nursing mothers. J Dairy Sci 60: 1858-1862

Hashemy-Tonkabony SE, Soleimani-Amiri MJ (1978). Chlorinated pesticide residues in the body fat of people in Iran. Environ Res 16: 419-422

Jensen AA (1983). Chemical contaminants in human milk. Residue Rev 89: 1-128

Karakaya AE, Ozalp S (1987). Organochlorine pesticides in human adipose tissue collected in Ankara (Turkey) 1984-1985. Bull Environ Contam Toxicol 38: 941-945

Karakaya AE, Burgaz S, Kanzık I (1987). Organochlorine pesticide contaminants in human milk from different regions of Turkey. Bull Environ Contam Toxicol 39: 506-510

Kutz FW, Wood PH, Bottimore DP (1991). Organochlorine pesticides and polychlorinated biphenyls in human adipose tissue. Rev Environ Contam Toxicol 120: 1-82

Loganathan BG, Tanabe S, Hidaka Y, Kawano M, Hidaka H, Tatsukawa R (1993). Temporal trends of persistent organochlorine residues in human adipose tissue from Japan, 1928-1985. Environ Pollut 81: 31-39

Menzie CM (1986). Hexachlorobenzene: Uses and occurrences. In: Hexachlorobenzene:Proceedings of an International Symposium, Lyon, France, eds, CR Morris,JRP Cabrel. IARC Publication No 77, Oxford University Press, New York, pp 13-22

Mes J, Davies DJ, Turton D (1982). Polychlorinated biphenyl and other chlorinated hydrocarbon residues in adipose tissue of Canadians. Bull Environ Contam Toxicol 28: 97-104

Mes J, Marchand L, Davies DJ (1990). Organochlorine residues in adipose tissue of Canadians. Bull Environ Contam Toxicol 45: 681-688

Murphy PG (1972). Sulfuric acid for the cleanup of animal tissues for analysis of acid-stable chlorinated hydrocarbon residues. JAOAC 55: 1360-1362

Mussalo-Rauhamaa H, Pyysalo H, Moilanen R (1984). Influence of diet and other factors on the levels of organochlorine compounds in human adipose tissue in Finland. J Toxicol Environ Health 13: 689-704

Nair A, Pillai MKK (1989). Monitoring of hexachlorobenzene residues in Delhi and Faridabad, India Bull Environ Contam Toxicol 42: 682-686

Ramachandran M, Banerjee BD, Gulati M, Grover A, Zaidi SSA, Hussain QC (1984). DDT and HCH residues in the body fat and blood samples from some Delhi hospitals. Ind J Med Res 80: 590-593

Robinson PE, Mack GA, Remmers J, Levy R, Mohadjer L (1990). Trends of PCB, hexachlorobenzene, and β-benzene hexachloride levels in the adipose tissue of the U.S. population. Environ Res 53: 175-192

Skaare JU, Tuveng JM, Sande HA (1988). Organochlorine pesticides and polychlorinated biphenyls in maternal adipose tissue, blood, milk, and cord blood from mothers and their infants living in Norway. Arch Environ Contam Toxicol 17: 55-63

Smrek AL, Needham LL (1982). Simplified cleanup procedures for adipose tissue containing polychlorinated biphenyls, DDT and DDT metabolites. Bull Environ Contam Toxicol 31: 251-256

Tanabe S, Mori T, Tatsukawa R, Miyazaki N (1983). Global pollution of marine mammals of PCBs, DDTs and HCHs (BHCs). Chemosphere 12: 1269-1275

Tobin P (1986). Known and potential sources of hexachlorobenzene. In: Hexachlorobenzene: Proceedings of an International Symposium, Lyon, France, Eds. CR Morris, JRP Cabral, IARC Publication No 77, Oxford University Press, New York, pp 13-22

Wickström K, Pyysalo H, Siimes MA (1983). Levels of chlordane, hexachlorobenzene, PCB and DDT compounds in Finnish human milk in 1982. Bull Environ Contam Toxicol 31: 251-256

Williams DT, LeBel GL, Junkins E (1984). A comparison of organochlorine residues in human adipose tissue autopsy samples from two Ontario municipalities. J Toxicol Environ Health 13: 19-29