

## **Total DDT and Dieldrin Content of Human Adipose Tissue**

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The persistence of organochlorines in the environment raises the problem of chronic toxicity caused by frequent low level exposures of animals and humans to these compounds. Since the pesticides DDT and dieldrin, like most organochlorines, are lipid soluble, they and their metabolites tend to accumulate in adipose tissue and lipid-rich organs, such as brain and liver, as well as in the lipid fraction of the biological membranes of all cells (deVlieger et al. 1968; Slater 1984).

The inductive and stimulating effects of organochlorine pesticides such as DDT and dieldrin on mixed function oxidases and cytochrome P 450 were recognized over 20 years ago (Conney et al. 1967). Since then further understanding has been gained, connecting the actions of mixed function oxidases and the formation of free radicals and lipid peroxides with damage to membranes and DNA (Halliwell and Gutteridge 1985).

Pesticides can enter human tissues through the foodchain, by inhalation of dusts, vapours and aerosols into the lungs or by direct diffusion through the skin. Hence it becomes important to establish the concentration of pesticides residing in human tissues.

As far as the authors could ascertain only 4 well-documented analytical studies have been carried out in Australia determining the total DDT (p,p'-DDD + p,p'-DDE + p,p'-DDT) and dieldrin content of human adipose tissue (Bick 1967; Wasserman et al. 1968; Brady and Siyali 1972; Siyali 1972; Table 1). Another unpublished study (Lugg 1970) has been quoted by Wassermann et al. (1975), see Table 1. The latest of these studies was published over 16 years ago,

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therefore it is timely and important to re-examine the total DDT and dieldrin concentration within the adipose tissue of the Australian population.

Particularly since it was found in 1987 that some samples of export beef contained traces of DDT and/or dieldrin.

The present investigation has analysed 290 samples of human adipose tissue obtained from Westmead Hospital situated in an outer suburb of Sydney, New South Wales (N.S.W.) for their content of total DDT and dieldrin.

Table 1. Total DDT and dieldrin concentrations in human adipose tissue. A comparison of Australian studies between the years 1965 and 1986.

Study and Location	Number of samples	Total DDT Mean $\mu\text{g/g}$ (Range $\mu\text{g/g}$ )	Dieldrin Mean $\mu\text{g/g}$ (Range $\mu\text{g/g}$ )
Bick (1967) Victoria	53	1.81 (0.48-6.35)	0.046 (0.01-0.43)
Wasserman et. al. (1968) Western Australia	12* 46**	9.3 (1.2-11.6) 9.5 (2.2-31.7)	0.67 (0.25-0.99)
Lugg (1969)		3.1	0.21
Brady and Siyali (1972) N.S.W.	75	4.94 (0.58-19.6)	0.21 (0.0-2.60)
Siyali (1972) N.S.W.	81***	4.75 (0.18-19.6)	0.21 (0.0-2.60)
Present study N.S.W.	292/290	3.72 (0.0-26.30)	0.13 (0.0-0.61)

\* Determined by GLC

\*\* determined by Schecter-Holler colorimetric analysis

\*\*\*including 75 samples from Brady and Siyali (1972)

## MATERIALS AND METHODS

Human adipose tissue samples were obtained from Westmead Hospital through the good offices of Prof. Stephen R. Leeder. The samples were obtained in 15 batches between August 1985 and November 1986 either post-operatively or from coroners' cases. The samples were frozen at the Hospital as soon as practicable and kept at  $-30^{\circ}\text{C}$  until used for analysis. Because of the high lipid content (in excess of 80% of the wet weight), only 300 to 500 mg of adipose tissue were used for analysis. Most of the adipose tissue samples obtained from Westmead Hospital weighed between 5 - 20 g. For each analysis small samples were taken from at least 3-4 different locations of the tissue sample, to ensure random sampling.

The extractions and analyses of p,p'-DDT, p,p'-DDD and p,p'-DDE were carried out by the one-step method as described previously (Ahmad and Marolt 1986). The average recoveries of p,p'-DDT, p,p'-DDD and p,p'-DDE by this method were previously found to be 81% (Ahmad and Marolt 1986).

In addition, extractions for the total DDT determinations were carried out by a modification of the method of Morton and Pollak (1987) for the determination of total organochlorines. About 400 mg of adipose tissue were sonicated in 0.8 mL distilled water in stoppered 10 mL tubes as described previously.

After sonication, 3 mL of Dole mixture (Dole 1955) were added, the tubes were shaken for 10 min, and 2 mL water were then added. The tubes were centrifuged for 10 min to break the emulsion. The yellow fat-hexane layer was quantitatively removed and made up to 1 mL with acetone. A 0.2 mL portion was placed onto an aluminium oxide: silicic acid column as described previously (Morton and Pollak 1987) and eluted with hexane. The eluates from the columns were adjusted to a convenient volume.

The quantitative determination of total DDT was carried out by Gas Liquid Chromatography as described previously (Ahmad and Marolt 1986), using a  $^{63}\text{Ni}$  electron capture detector and a 1.5m x 2mm glass column packed with 80-100 mesh Gas Chrom Q coated with 5% QF-1 and 5% DC 200. The operating temperatures were: Injector  $220^{\circ}\text{C}$ , column  $190^{\circ}\text{C}$ , detector  $230^{\circ}\text{C}$ ; carrier gas flow was kept at 45 mL/min. Samples were diluted so that injection volumes were able to be kept between 1-2  $\mu\text{L}$ . After every 4-6 samples a standard containing known amounts of p,p'-DDT, p,p'-DDD and p,p'-DDE were injected into the GC column to facilitate quantitation and to check on the constancy of the retention times

and detector responses. Quantitation was carried out by comparing peak heights of samples with those of standards.

After the extraction of p,p'-DDT, p,p'-DDD and p,p'-DDE by the one-step method of Ahmad and Marolt (1986) had been completed, dieldrin was eluted from the column with 90 mL n-hexane:acetone (96:4). It has been shown that this sequential use of solvents yielded recoveries of dieldrin between 81-102% (Ahmad et al. 1988). The quantitative determinations of dieldrin were carried out by GLC as described by Ahmad et al. (1988).

## RESULTS AND DISCUSSION

When human adipose tissue was extracted by the one-step method (Ahmad and Marolt 1986), it was found that in these tissue samples, variation of the total DDT residues from sample to sample was high, as evidenced by the large standard deviation from the mean (Table 2). To check on the reliability of the one-step extraction procedure, the same adipose tissue samples were also extracted by an alternative method (Morton and Pollak 1987). Total DDT residue values obtained by the two methods showed a strong correlation ( $r=0.707$ , Table 2) and the difference between the two methods was not statistically significant (Table 2), suggesting that the variability of the different tissue samples was not an artefact of the extraction procedure. In extracts obtained by both methods, p,p'-DDE accounted for all of the total DDT in more than 90% of the adipose tissue samples.

The frequency distribution of total DDT residues (using both methods) and dieldrin residues as obtained by the one-step method are presented in Figs. 1a and 1b. Out of a total of 292 samples, no total DDT was detected in 3 samples and dieldrin was below the detectable limit of 0.001  $\mu\text{g/g}$  in 2 samples. The results of the present investigation are compared in Table 1 with those of previous surveys in Australia. These data indicate that in spite of partial restrictions and/or the diminishing use of DDT and dieldrin, the residues of these organochlorines have not significantly decreased over a 20 year span. The more recent (1987) complete banning of DDT and dieldrin from agricultural use in Australia should lead eventually to a decrease in the residues of these organochlorines in the environment and also in human adipose tissues. However aldrin (which is epoxidised to dieldrin) and dieldrin are still registered and used as termiticides.

Table 2. Comparison of Total DDT concentrations in human adipose tissue as obtained by two extraction procedures.

	Ahmad and Marolt (1986)	Morton and Pollak (1987)
Number of samples analysed	292	283
Mean	3.72 µg/g	3.42 µg/g
Standard deviation	3.869	3.992
log Standard deviation	0.5876	0.6012
Standard error	0.226	0.237
Paired T-test (degrees of freedom=280)		
Mean	3.69	3.39
Standard deviation	3.827	3.985
Correlation coefficient	0.707	
Mean (dif.)	0.305	
Standard deviation (dif.)	2.995	
Students-T	1.706	
Probability (Nul hypothesis)	0.089	

The total DDT residue levels found in this investigation are of the same order of magnitude as the total DDT residues in adipose tissue of a Canadian study (Williams et al. 1984) which analysed samples collected in Kingston between 1979 and 1981 and in Ottawa between 1980 and 1981. The dieldrin concentration of adipose tissue in the present study was however significantly higher than the residues of dieldrin found in the Canadian study. The mean value for dieldrin of 0.13 ug/g obtained in the present study compared with the mean of 0.036 ug/g from 91 samples obtained in Kingston, and with the mean of 0.043 ug/g detected in 84 samples from Ottawa (Williams et al. 1984).

When medical records become available from Westmead Hospital for the subjects who provided the adipose tissue samples, the total DDT and dieldrin residue levels will be evaluated in relation to the site from which the tissue sample originated, age, sex, body weight, occupation and morbidity to see if any correlations may be established.

Fig. 1a

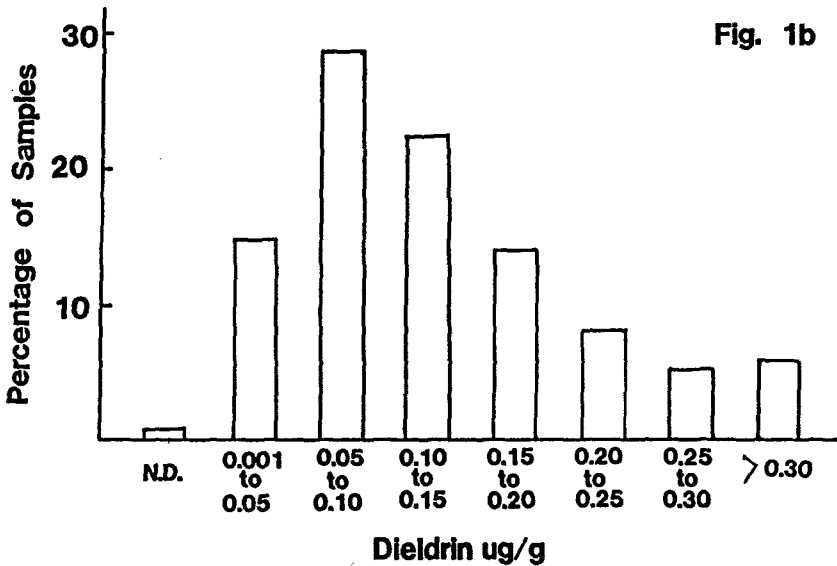
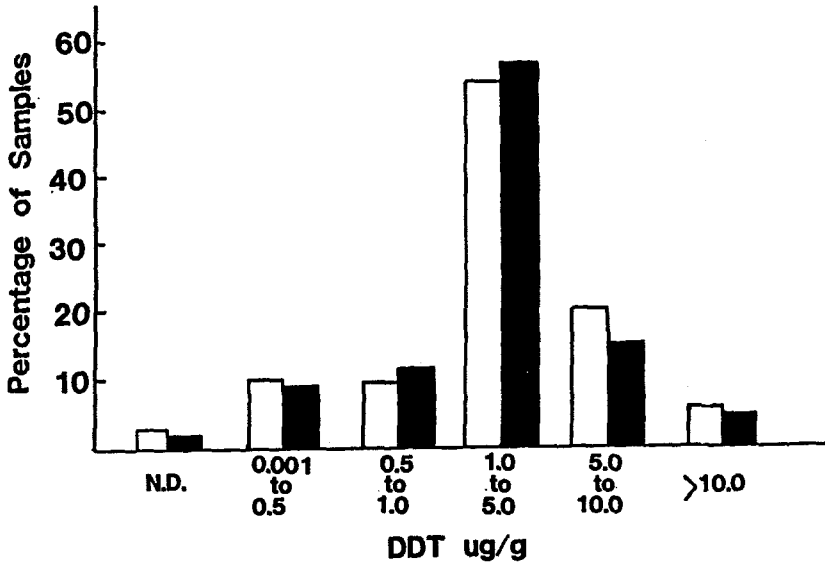


Figure 1a. Distribution pattern of total DDT residues in human adipose tissue. Open columns, extracted by the method of Ahmad and Marolt (1986). Solid columns extracted by the method of Morton and Pollak (1987).

Figure 1b. Distribution pattern of dieldrin residues in human adipose tissue.

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