Bull. Environ. Contam. Toxicol. (1989) 42:911–918

Environmental Contamination and Toxicology

Levels of Organochlorine Insecticides in Milk of Mothers from Urban and Rural Areas of Botucatu, SP, Brazil

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The use of organochlorine insecticides started to be more widely disseminated in the forties. But this has become a serious problem of public health, due to the fact that insecticides accumulate in tissues and biological liquids owing to their fat-soluble character, their persistence in the environment and their accumulation in the food-chain. Since man is located at the top of the food-chain, he receives and accumulates insecticide residues that vegetables and animals have stored up in various periods of development (Hornabrook et al. 1972, Lara and Barreto 1972).

The continuous development of gas chromatographic techniques allowed the detection of ppb levels of these insecticide residues. On the other side, the research on the chronic effects due to indiscriminated use of these products did not keep track at the same speed, since the observation of the toxic effects in man is misled by a series of external factors, which can not be isolated. Studies with laboratory animals have been useful to establish the toxicity of these compounds, since there is possibility to control these factors. Therefore, it has been observed a change in levels of bovine limphocytes and intracelular calcium due to DDT exposition (McCabe and Yin-Foo 1986), an immunesupressor effect of DDT in mice (Banerjee et al. 1986) and a great evidence of the carcionogenic effects of DDT, HCH, aldrin and heptachlor in several laboratory animals (Zambrone 1986).

Human milk can be used as an evaluation index of environmental contamination by these insecticides, although the main objective of its analysis is to determine the amounts ingested by children, who, without a doubt, will have to face other sources of contamination during their lives. When evaluating the levels of organochlorine insecticides in human milk it is useful to establish where the mothers live. Theoretically, mothers who live in a rural

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area have much more contact with these insecticides, because they work directly in agriculture. Therefore, the risk of exposure by their nursing children will be even greater. This aspect causes even more worries in developing countries where agriculture has always been one of the predominant sectors of the economy. In these countries the need to increase productivity has led to the indiscriminate use of organochlorine insecticides because of their low cost and efficacy. In Brazil, farmers do not have enough knowledge to measure the risks brought about by their indiscriminate use. In addition, government programmes for the control of rural endemic diseases still make use of DDT and HCH on a large scale.

MATERIALS AND METHODS

Forty two nursing mothers were interviewed, 21 from the urban zone, and 21 from the rural. The urban zone mothers were chosen randomly from the population of the city of Botucatu, São Paulo, and the ones from the rural zones were involved in a post-natal care program at the Health Centers in the towns of Anhembi and Pardinho, both of which belong to the township of Botucatu. Each mother had to answer a questionnaire on their age, number of children, education level, whether their houses had been sprayed, smoking habits and domestic use of insecticides. The questionnaire for the women from the rural zone had a specific part related to the use of insecticides in the fields.

Milk samples (10-15 ml) were collected by a manual suction pump, stored in glass flasks and frozen at -20°C until submitted to analysis, within 72 hours. The percent of fat in the milk was also determined (AOAC 1980).

The extraction of organochlorine insecticides was carried out according to the generally accepted procedures (US Environmental Protection Agency 1980). Residues were determined by gas chromatography using a CG-3735 instrument, on a 1.8m x 1/8" id glass column packed with 1.5 OV-17 + 1.95% QF-1 on Chromosorb W HP. Working conditions were: temperatures of injector - 216°C, of column - 204°C, and detector - 218°C. Carrier gas was nitrogen at a flow rate of 40 mL/min.

RESULTS AND DISCUSSION

Table 1 shows the levels of DDT and its metabolites in the samples of whole milk, in ppb, obtained from mothers living in urban and rural areas. The DDTt value refers to the metabolites pp'-DDE, op'-DDT, and pp'-DDT. The average level of DDTt in samples from the urban zone was superior to that of the rural zone, 34.9ppb and 16.5ppb respectively. The reason for these levels may be attributed to the differences in economic class and eating habits

TABLE 1. LEVELS OF DDT AND ITS METABOLITES IN WHOLE MILK FROM MOTHERS LIVING IN URBAN AND RURAL AREAS

Sample	Urban area (ppb)[a]				Rural area (ppb)[a]			
	pp'-DDE	op'-DDT	pp'-DDT	DDTt[b]	pp'-DDE	op'-DDT	pp'-DDT	DDTt
1	3.1	64.3	ND	101.2[c]	1.0	ND	ND	1.0
2	1.0	1.0	39.1	41.0	33.3	ND	ND	33.3
3	ND	ND	ND	ND	ND	ND	ND	ND
4	35.0	ND	ND	35.0	19.2	ND	ND	19.2
5	16.1	17.0	19.1	52.2	16.6	ND	ND	16.6
6	1.0	19.4	29.7	50.0	ND	ND	ND	ND
7	17.0	ND	31.7	48.7	3.5	ND	ND	3.5
8	18.1	21.6	34.6	95.9	6.8	ND	ND	6.8
9	3.5	5.8	23.2	32.5	ND	ND	ND	ND
10	10.4	5.9	21.7	38.0	62.1	ND	ND	62.1
11	21.3	13.4	ND	34.7	1.1	ND	ND	1.1
12	21.6	10.4	20.9	52.9	21.1	ND	1.0	22.1
13	9.4	3.9	23.1	36.4	1.0	ND	ND	1.0
14	17.3	11.8	19.2	48.4	1.0	ND	1.0	2.0
15	16.7	ND	ND	16.7	1.0	ND	ND	1.0
16	12.6	ND	ND	12.6	23.0	ND	ND	23.0
17	11.0	ND	ND	11.0	13.3	ND	ND	13.3
18	10.8	1.0	1.0	12.8	15.3	ND	1.0	16.3
19	1.0	ND	ND	1.0	20.6	15.2	ND	35.8
20	9.9	ND	ND	9.9	12.6	1.0	29.1	42.7
21	1.0	ND	ND	1.0	43.7	ND	1.0	44.7
$\bar{x}$	11.3	8.4	12.5	34.9	14.1	0.8	1.6	16.5

a - ND: not detected; 1 ppb represents the detectable limit of the method.

b - Represents the sum of metabolites.

c - Contains also 33.8 ppb of pp'-DDD.

between the two areas under research.The diets of mothers in rural areas contained less meat and more vegetables.

Fifteen of the 21 mothers surveyed in the rural area worked harvesting cotton, the only crop where it is customary to use DDT after planting. However, the average level of DDTt in the milk of the mothers who worked harvesting cotton was 16.7 ppb, a level which does not differ from the average total level encountered in the milk of the rural zone mothers. These results suggest that DDT has not been much used in these cotton crops and thus the contamination of human milk by this insecticide may be attributed to contaminated foodstuffs and the domestic use of this insecticide.

Table 2 presents the levels of the isomers α,β and γ-HCH (in ppb) in whole milk samples of mothers from urban and rural zones. The HCHt value refers to the sum of the isomers. The average level of HCHt was higher in the samples from the rural zone than the urban zone (46.3 and 14.4 ppb respectively). As previously indicated, among mothers from the rural zone eighteen worked in crop-areas, the great majority in cotton, sugar-cane and coffee plantations that frequently made use of HCH. The higher levels of HCHt detected in the samples from the rural zone are due to its utilization in crop-areas and other sources of contamination, such as the food-chain and house spraying against rural endemic diseases.

The relation between the average levels of DDTt in the milk and the age of the nursing mothers was verified using three age-groups, 15-25, 26-35 and over 35. The following levels were found: 25.7, 50.0 and 48.7ppb respectively in the samples of the urban zone, and 16.9, 18.1 and 8.7 ppb respectively, in the samples from the rural zone (Table 3). These results indicate that there is no relation between the levels of DDTt encountered and the age of the mothers from both zones.

As lactation is a form of excretion of insecticides,lower levels in more advanced ages are expected, provided that they had a larger number of children. Apart from this consideration, there are two other factors which may influence the levels of insecticide found in the milk of these mothers: first, women tend to get fatter as they grow older, with a consequent dilution of insecticides in the organism; and second, in the more elevated age groups - over 35 years - the mothers probably were in contact with the insecticide at the peak of its use, during the first post-war decade, having thus been more exposed to these chemical compounds. Both Matuo (1978) and Currie et al. (1979) suggest that the number of children a woman has had before is the most preponderant factor in the reduction of the level of DDTt in the milk. The data available and the present research

TABLE 2. LEVELS OF HCH AND ITS ISOMERS IN WHOLE MILK FROM MOTHERS LIVING IN URBAN AND RURAL AREAS

Sample	Urban area (ppb)[a]				Rural area (ppb)[a]			
	α-HCH	β-HCH	γ-HCH	HCHt[b]	α-HCH	β-HCH	γ-HCH	HCHt[b]
1	ND	ND	ND	ND	1.0	1.0	1.0	3.0
2	1.0	1.0	1.0	1.0	1.0	15.9	ND	16.9
3	ND	31.0	ND	31.0	ND	36.7	ND	36.7
4	1.0	1.0	ND	2.0	ND	ND	ND	ND
5	ND	ND	ND	ND	ND	ND	ND	ND
6	1.0	29.0	1.0	31.0	1.0	1.0	ND	1.0
7	ND	9.8	ND	9.8	ND	ND	ND	ND
8	ND	ND	ND	ND	ND	120.4	ND	120.4
9	ND	11.8	ND	11.8	ND	78.1	ND	78.1
10	ND	15.8	1.0	16.8	ND	111.7	ND	111.7
11	ND	10.9	ND	10.9	1.0	13.5	ND	14.5
12	ND	ND	17.4	17.4	ND	73.7	1.0	74.7
13	1.0	1.0	ND	2.0	ND	21.9	ND	21.9
14	ND	ND	21.9	21.9	ND	11.3	ND	11.3
15	ND	64.1	ND	64.1	ND	1.0	1.0	2.0
16	9.3	ND	ND	9.3	ND	25.2	ND	25.2
17	ND	ND	ND	ND	ND	97.5	ND	97.5
18	ND	ND	ND	ND	1.0	23.4	1.0	25.4
19	ND	26.3	ND	26.3	ND	93.3	ND	93.3
20	ND	43.5	ND	43.5	ND	52.7	ND	52.7
21	1.0	ND	ND	1.0	1.0	184.2	ND	185.2
$\bar{x}$	0.7	11.7	2.0	14.4	0.3	45.8	0.2	46.3

a - ND: not detected; 1 ppb represents the detectable limit of the method.

b - Represents the sum of the isomers.

Table 3. DDTt and HCHt average levels in whole milk samples of mothers living in urban and rural areas, according their ages, number of children, house spraying and smoking habit

Parameters	DDTt (ppb)		HCHt (ppb)	
	Urban	Rural	Urban	Rural
Age (years)				
15 - 25	25.7	16.9	15.9	53.5
26 - 35	50.0	18.1	12.2	41.7
> 35	48.7	8.7	9.8	13.7
Number of children				
1 - 2	45.2	18.9	33.1	49.8
> 3	20.5	12.0	12.8	44.4
House spraying				
Yes	32.7	21.3	15.8	61.9
No	35.5	14.0	14.7	38.5
Smoking habit				
Yes	45.6	18.2	-	-
No	29.5	14.5	-	-

seem to confirm this hypothesis. Thus, mothers of urban and rural zones presented higher average levels of DDTt when they had nursed 1 to 2 children, that is, 45.2 and 18.9 ppb respectively, when compared with mothers who had nursed 3 or more children (20.5 ppb for the urban zone, and 12.0 ppb for the rural zone).

House spraying with insecticide in rural areas seems to have contributed to the higher levels of DDTt in the milk of mothers living in the areas (21.3 ppb) when compared with that of mothers living in houses that had not been sprayed (14.0 ppb). However, in the urban zone,the fact that houses had or not been sprayed with insecticide did not alter the average level of DDTt (32.7 and 35.5 ppb, respectively).

The average levels of DDTt were higher in the milk of mothers who smoked, in both areas under research, when compared with that of non-smokers.The mothers who smoked, in the urban zone,presented 46.5ppb of DDTt against 29.5 ppb for non-smokers,while in the rural zone the levels were 18.1 and 14.5ppb respectively.The mothers who smoked presented higher levels of DDTt in their milk because cigarette is a source of contamination of these insecticides, since they are used on tobacco plantations.

In relating average levels of HCHt with age of the women (Table 3), it was observed that the levels are higher in the first age group (15-25), for both urban and rural zones. The levels of HCHt were respectively 15.9 and 53.5 ppb for ages 15-25; 12.2 and 41.7ppb from 26 to 35 years, and 9.8 and 13.7 for those over 35.

The relation between the levels of HCHt and the number of children showed higher levels for those mothers with one or two children in both urban and rural zones (33.1 and 49.8 ppb respectively) similar to the relation of DDTt levels / number of children. It is interesting to observe that the average level was also higher in the milk of mothers from rural zones (44.4 ppb) with 3 or more children than in those of the urban zone (12.8ppb).

When comparing average levels of HCHt in samples related to house spraying, it was observed that in both areas, under research, the levels were higher in samples from mothers living in houses that had been sprayed (urban zone: 15.8 and rural zone: 61.9ppb) than those from mothers living in houses not sprayed (14.7 and 38.5ppb, respectively). The average values of HCHt in the samples from mothers living in sprayed houses in the rural zone were much higher than those of the urban zone (approximately four times) or even than those who live in houses in the same area but that have not been sprayed (about 50%). A possible explanation for such high values could be that in the rural zone the application of HCH in

houses, is made by public health authorities as part of a campaign, where this insecticide is more widely used, while in the urban zone house spraying is in the hands of private companies that employ a formula which does not necessarily include HCH.

In relation to other insecticides, the presence of dieldrin was detected in only three samples of human milk, all from the urban zone (21.4, 4.6, and 13.2 ppb). This contamination must have been caused by the ingestion of food or through the domestic use of dieldrin or aldrin.

HCB, which is a fungicide and not an insecticide, was detected in only one sample from the urban zone (9.6ppb). The mother in question had used the product in research work.

The analysis of the levels of insecticides studied on a fat-basis presented average values in proportion to those encountered and discussed for whole milk. Milk fat from the urban zone samples presented 1800 ppb of DDTt and 730 ppb of HCHt, and those from the rural zone 770 ppb of DDTt and 2130 ppb of HCHt.

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Received September 12, 1988; accepted December 31, 1988.