Levels of organochlorine pesticides in the blood serum of agricultural workers from Rio de Janeiro State, Brazil

Níveis de pesticidas organoclorados no soro sangüíneo de agricultores do Estado do Rio de Janeiro, Brasil

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Abstract *Serum levels of organochlorine pesticides (OCP) were measured in agricultural workers from Rio de Janeiro State, Brazil. Blood samples from 26 volunteers (24 males, 02 females, 17-60 years old) were taken in October 1997. OCP residues (op'DDT pp'DDT, pp'DDD, pp'DDE, aldrin, dieldrin, endrin, heptachlor, heptachlor-epoxide, α-, β- and γ-hexachlorocyclohexane, and hexachlorobenzene) were analyzed by gas chromatography with an electron capture detector. Tests detected pp'DDE in 16 out of 26 samples, but pp'DDE concentration exceeded 1.4 µg/L (i.e. 1.8, 2.4 and 4.4 µg/L) in only 3 of these. β-HCH was found in 6 (23.1%) out of 26 samples. In one sample β-HCH did not exceed 1.4 µg/L, but in the remaining samples concentrations ranged from 1.4 to 5.3 µg/L. The percentage of positive pp'DDE samples increased from the youngest (≤29 yrs: 30.0%) to the oldest age group (≥40 yrs: 100%). A similar trend was found for β-HCH contamination (≤29 yrs: 0%; 30-39 yrs: 20.0%; ≥40 yrs: 66.7%). Dieldrin (3.7 µg/L) was found in only one sample. No other OCP residue was found in the samples. Serum concentrations of OCPs found in this study are comparable to levels reported for the non-occupationally exposed population in Brazil and elsewhere.*
Key words *Pesticides; DDT; Dieldrin; Occupational Exposure*

Resumo *Os níveis sangüíneos de pesticidas organoclorados (OCP) foram determinados em agricultores do Estado do Rio de Janeiro, Brasil. Amostras de soro de 26 voluntários (24 homens, duas mulheres, entre 17 e 60 anos de idade) foram retiradas em outubro de 1997. Os resíduos de OCP (op'DDT pp'DDT, pp'DDD, pp'DDE, aldrin, dieldrin, endrin, heptaclor, heptaclor-epóxido, α-, β- γ-hexaclorociclohexano e hexaclorobenzeno) foram analisados por cromatografia gasosa com detector de captura de elétrons. O pp'DDE foi detectado em 16 das 26 amostras, mas em apenas três delas os níveis de pp'DDE excederam 1,4 µg/L (1,8; 2,4 e 4,4 µg/L). O β-HCH foi encontrado em seis (23,1%) das 26 amostras. Em uma das amostras o β-HCH não excedeu a 1,4 µg/L, mas nas restantes as concentrações variaram de 1,4 a 5,3 µg/L. A percentagem de amostras positivas para pp'DDE aumentou do grupo mais jovem (≤29 anos: 30,0%) para o mais velho (≥40 anos: 100%). Uma tendência semelhante foi observada para a contaminação por β-HCH (≤29 anos: 0%; 30-39 anos: 20,0%; ≥40 anos: 66,7%). O dieldrin (3.7 µg/L) foi encontrado em apenas uma das amostras. Nenhum outro resíduo de OCP foi encontrado nas amostras. As concentrações de OCPs encontradas nos agricultores são comparáveis aos níveis sangüíneos relatados para a população não exposta ocupacionalmente no Brasil e em outros países.*
Palavras-chave *Praguicidas;DDT; Dieldrin; Exposição Ocupacional*

Introduction

Agricultural workers from Third World countries are at particularly high risk for pesticide-related illnesses. In developing countries, as a rule, agrochemicals are carelessly handled and personal protective equipment as well as adequate clothing are seldom used by workers engaged in mixing, loading, and spraying pesticides (Forget, 1991; Jeyaratnam, 1993). Furthermore, lack of government and public concern, lack of stringent regulations, low standards of living, poor education, and illiteracy in rural areas are factors that contribute to make occupational exposure to pesticides a major public health problem in less developed countries.

In Brazil, as well as in most Latin American countries, pesticides are extensively used for crop protection and to control insect-borne diseases (e.g., malaria, dengue, yellow fever, Chagas' disease, leishmaniasis, and bubonic plague) but health surveillance of agricultural workers and biological monitoring of occupational pesticide exposure have seldom been performed.

Owing to their environmental persistence, bioaccumulation in food chains, and very slow elimination from the human body, most organochlorine pesticides (OCP) were banned from agricultural use in Brazil in 1985 (Brazil, 1985). However, their use in public health campaigns is still allowed, and it has been reported that the National Health Foundation used 3,000 tons of dichlorodiphenyltrichloroethane (DDT) to control the mosquito *Anopheles darlingi* (one of the vectors of malaria) in the Amazon Region in 1992 (Oliveira-Filho, 1997). Moreover, illegal trade, non-authorized use of OCPs in agriculture and areas highly contaminated with technical grade hexachlorocyclohexane (HCH) have been cause for concern in Brazil.

Recently, there has been growing public health concern related to environmental pollutants which have the potential to interact with the endocrine system, i.e., the so-called "endocrine disruptors". It has been demonstrated that exposure to some persistent organochlorine compounds such as DDT and its metabolites, polychlorinated biphenyls (PCBs), and dioxins may cause reproductive failure in wildlife species (e.g., DDT-induced egg shell thinning in birds of prey) as a consequence of their hormone-like effects. Since certain types of cancer (e.g., breast, prostate, and testicular) have an endocrine-related etiology, debate has heightened concerning the potential link between organochlorine compounds and these tumors. For instance, a study by Wolff et al. (1993) found that breast cancer was closely associated with serum levels of DDE [1,1-dichloro-2,2-bis(p-chlorophenyl)ethylene], the main metabolite of DDT. However, data from other recent studies did not support the hypothesis that exposure to DDT and other persistent organochlorine compounds increases risk of breast cancer (Krieger et al, 1994; Hunter et al, 1997; Schecter et al, 1997).

Whether human exposure to persistent organochlorine compounds has adverse health consequences is still a matter of controversy. However, within this context, exposure assessments and more data on the human populations exposed to organochlorine compounds are urgently needed.

The present study was undertaken to provide data on organochlorine pesticides contamination of agricultural workers from Rio de Janeiro State. It is part of a more comprehensive evaluation of the exposure of the Brazilian population to persistent organochlorine pesticides.

Methods

Paty-do-Alferes (22º15'-22º30' South and 43º 16'-43º31' West) is a small county located in the Serra-do-Mar mountain range, some 130 km from the city of Rio de Janeiro. Most of its 22,500 inhabitants live in the rural area, and farming (tomatoes, green peppers, cabbage, cauliflower, and other vegetables) has been the main economic activity in the region for decades.

Twenty-six volunteers (24 males and 2 females) took part in the present study. Recruitment for voluntary participation involved contact with the four main tomato planters from the rural districts of Caetés, Campo Verde, and Bela Vista. All participants were agricultural workers who had been engaged in mixing, loading, and/or applying pesticides. Data on the use of personal protective equipment, type of exposure to pesticides, and work history are shown in Table 1. Volunteers were asked to sign an Informed Consent form after having received a full explanation of the objectives and procedures to be followed in the study. Afterwards, all participants were interviewed and examined by a medical doctor, and blood samples were drawn from an arm vein. Blood samples were taken in silicone-coated Vacutainer® tubes without anticoagulants and left to stand for 30 minutes. After clotting, serum was separated by centrifugation. Serum samples were immediately frozen and sent to the Adolfo

Table 1

Occupational data from the group of agricultural workers who took part in the present study. All participants are from Paty-do-Alferes, Rio de Janeiro State, where they grow tomatoes, green peppers, cabbage, cauliflower, and other vegetables.

Number	Sex	Age (yrs)	Time (yrs) in agricultural work		Type of exposure	Use of protective equipment
			Total	With pesticides		
1	M	50	40	40	A, M, L	No
2	M	34	27	21	A, M, L	No
3	M	44	37	34	A	No
4	M	36	23	23	A	No
5	M	26	15	12	A	No
6	M	30	22	22	A	No
7	M	31	23	23	M,L	No
8	M	25	15	15	A	No
9	M	26	16	8	A	No
10	M	21	9	9	A	No
11	M	59	41	41	M,L	No
12	M	36	29	29	A, M, L	No
13	M	32	19	19	A	No
14	M	30	20	12	A	No
15	M	18	11	1	M, L	No
16	M	30	18	13	A	No
17	M	17	< 1	< 1	A	No
18	M	18	4	< 1	A	No
19	M	33	23	23	A	No
20	M	60	35	35	A	No
21	M	29	22	22	A, M, L	No
22	M	28	20	20	A	No
23	F	45	30	30	M	No
24	F	49	42	42	M	No
25	M	26	19	19	A, M, L	No
26	M	30	15	15	A	No

M: male; F: female; A: applicator (sprayer); M: mixer; L: loader; No: do not use any protective equipment (gloves, face masks, and adequate clothing).

Lutz Institute, São Paulo, where they were analyzed for organochlorine pesticide residues. Except for Vacutainer® tubes, all vials were prewashed with residue grade *n*-hexane. Additional blood samples were collected for assessing basic hematological parameters as well as for biochemical liver and kidney function tests. Organochlorine residues were extracted with *n*-hexane and analyzed by gas chromatography with an electron capture detector (Ni63) as previously described by Dale et al. (1970). Nitrogen (high purity grade) was the carrier gas at 40 ml/min. Chromatographic parameters were as follows: column temperature: 60ºC, 25ºC/min, 190ºC, 5ºC/min, 280ºC (2 min), injector temperature: 250ºC, and detector temperature: 320ºC. A capillary column (25m x 0.25mm x 0.33µm) with 5% phenyl methyl siloxane was used. Limits for residue quantification which were validated for the present method are as follows: *op*'DDT, *pp*'DDT and *pp*'DDD = 2.8 µg/L; *pp*'DDE = 1.4 µg/L; aldrin, dieldrin, endrin, heptachlor, and heptachlor-epoxide = 1.4 µg/L; α- and γ-hexachlorocyclohexane (HCH) = 0.7 µg/L, β-HCH = 1.4 µg/L; hexachlorobenzene (HCB) = 0.7 µg/L. All blood samples were taken and analyzed in October 1997.

Results and discussion

DDT derivatives

The derivatives *op*'DDT, *pp*'DDT and *pp*'DDD were not found in any of the samples analyzed. On the other hand, as shown in Table 2, *pp*'DDE

Table 2

Concentrations (µg/L) of β-HCH, dieldrin, and pp'DDE in the blood serum of agricultural workers from Paty-do-Alferes, Rio de Janeiro State, Brazil (October 1997).

Number	Sex	Age (yrs)	β-HCH	dieldrin	*pp*'DDE
1	M	50	nd	nd	< 1.4
2	M	34	1.4	nd	< 1.4
3	M	44	< 1.4	nd	< 1.4
4	M	36	1.7	nd	4.4
5	M	26	nd	nd	nd
6	M	30	nd	nd	< 1.4
7	M	31	nd	nd	< 1.4
8	M	25	nd	nd	nd
9	M	26	nd	nd	nd
10	M	21	nd	nd	nd
11	M	59	nd	nd	< 1.4
12	M	36	nd	nd	nd
13	M	32	nd	nd	< 1.4
14	M	30	nd	nd	nd
15	M	18	nd	nd	< 1.4
16	M	30	nd	nd	nd
17	M	17	nd	nd	nd
18	M	18	nd	nd	nd
19	M	33	nd	nd	< 1.4
20	M	60	1.7	nd	< 1.4
21	M	29	nd	nd	nd
22	M	28	nd	nd	1.8
23	F	45	1.7	nd	< 1.4
24	F	49	5.3	3.7	2.4
25	M	26	nd	nd	< 1.4
26	M	30	nd	nd	< 1.4

M: male; F: female; nd: not detected. Limit for quantification (β-HCH, dieldrin, and *pp*'DDE) = 1.4 µg/L.

chromatographic peaks were detected in 16 (61.5 %) out of 26 serum samples, but *pp*'DDE concentration exceeded 1.4 µg/L (*i.e.* 1.8, 2.4 and 4.4 µg/L) in only in 3 (11.5%). Technical DDT consists predominantly of *pp*'DDT and some *op*'DDT. In the human body, *pp*'DDT is slowly dechlorinated to *pp*'DDD and *pp*'DDE, the latter being even more persistent than the parent compound (WHO, 1979). Therefore, since no *pp*'DDT was found in the blood samples, contamination of agricultural workers appears to have resulted from past exposure to technical grade DDT, years ago, and/or from intake of *pp*'DDE residues present in food (e.g., meat, milk, eggs). It has been demonstrated that *pp*'DDE, expressed as a percentage of total DDT-related material, increases in individuals after DDT ingestion decreases and also increases in successive steps of the food chain (WHO, 1979). It is of note that, in the present survey, the percentage of *pp*'DDE-positive samples increased from the youngest (=29 yrs: 30.0%) to the oldest age group (=40 yrs: 100%) (Table 3).

There are only a few studies on blood levels of DDT in the Brazilian population. As shown in Table 4, levels of *pp*'DDE and total-DDT in blood samples of non-occupationally exposed Brazilians in 1972 are considerably higher than serum concentrations of *pp*'DDE in agricultural workers from Paty-do-Alferes, Rio de Janeiro State, in 1997. Similarly, levels found in the present study are lower than serum concentrations of *pp*'DDE and total DDT in samples of the general population from São Paulo, Goiás, and Bahia States in the 1980s and early 1990s (Table 4).

Hexachlorocyclohexane isomers

β-HCH was detected in 6 (23.1%) out of 26 samples. In one sample β-HCH did not exceed 1.4 µg/L, but in the remaining samples, concentrations ranged from 1.4 to 5.3 µg/L (Table 2). As shown in Table 1, traces of *pp*'DDE were present in all samples in which β-HCH was found. No β-HCH-positive sample was found in the youngest group (≤29 yrs), and the percentage of positive samples in the intermediate age group (30-39 yrs: 20.0%) was lower than that in the oldest group (≥40 yrs: 66.7%) (Table 3). Murphy & Harvey (1985) found a similar trend analyzing 6252 blood samples (17-74 years of age) from the United States population. β-HCH was detected in 13.9% of the samples (mean: 1.7 µg/L; range: 1.0-28.0 µg/L), and the percentage of positive samples increased with age from 3.2 to 26.8% (Murphy & Harvey, 1985). Levels of β-HCH in blood samples of agricultural workers from Paty-do-Alferes were not higher than those that had been reported for the general population in Brazil and elsewhere. Likewise, even higher serum concentrations were found in samples of the general population from São Paulo and Bahia States in the 1980s and 1990s (Table 5). Blood serum levels of β-HCH were also higher in Brazilian adults living in Cidade-dos-Meninos (Rio de Janeiro State), a highly contaminated area near the ruins of a former technical-HCH manufacturing plant (Braga, 1990). Blood concentrations of β-HCH measured in the present study were much lower than those reported for occupationally-exposed workers. As shown in Table 5, serum levels of β-HCH in workers from cacao plantations in Bahia State (Carvalho, 1991) are higher than those found in agricultural workers from Paty-do-Alferes in 1997. It should be borne in mind, however, that technical-

grade HCH was still extensively used in cacao plantations in Southern Bahia when blood samples were taken in 1983-1985 (Carvalho, 1991). Serum concentrations of β-HCH in Paty-do-Alferes workers are also much lower than those found by Minelli & Ribeiro (1996) and Carvalho (1991) in mosquito control sprayers from São Paulo and Bahia States, respectively (Table 5).

No α- or γ-HCH was detected in blood samples analyzed in the present study. β-HCH is only a minor component (7-10%) of technical BHC but it is the most persistent HCH isomer in the human body (WHO, 1992). It has been reported that after exposure ceases in humans, β-HCH levels in fatty tissues decrease only slightly over several years (WHO, 1992).

Aldrin and dieldrin

Aldrin was not detected in any serum sample in this survey. This is not surprising, since aldrin is rapidly converted to dieldrin in the body. On the other hand, dieldrin (biological half-life = 267 days; Tordoir & van Sittert, 1994) was found in only one sample (a female worker, 49 yrs old) at a concentration of 3.7 µg/L (Table 2). This value is within the variation range found for members of the general populations in different countries until the 1980s (WHO, 1989), and it is well below the Biological Limit Value (blood: 100 µg/L) recommended by Tordoir and van Sittert (1994). Higher levels of dieldrin in Brazilian workers were reported in the 1980s. Lara et al. (1981) analyzed the blood serum of 18 agricultural workers from São José do Rio Preto (São Paulo State) and found a mean concentration of dieldrin of 490 ± 360 µg/L (mean ± S.D.), with values ranging from 50 to 1,370 µg/L. In 1983-1985, Carvalho (1991) found a high mean concentration of dieldrin of 1.3 ± 0.5 µg/L (mean ± S.D.) in serum samples of 10 agricultural workers from cacao plantations in Bahia State. Lara et al. (1987) detected dieldrin at concentrations of <10 to 80 µg/L in the blood serum of 51 people living in areas where Chagas' disease was endemic in Goiás State.

Table 3

Age-dependence of contamination by DDT (*pp*'DDE) and β-HCH in agricultural workers from Paty-do-Alferes, Rio de Janeiro State. Serum samples were taken from 26 volunteers in October 1997.

	Age (years)			
	≤29	30 to 39	≥40	17 to 60
Serum samples				
Total analyzed (n)	10	10	6	26
Positive samples				
pp'DDE				
n	3	7	6	16
%	30.0	70.0	100.0[a]	61.5
β-HCH				
n	0	2	4	6
%	0	20.0[a]	66.7[ab]	23.1

Data were analyzed by the chi-square test or, alternatively, by Fischer's exact test. Percentages significantly different (P<0.05) from the youngest group (≤29 yrs) and from the intermediate age group (30-39 yrs) are indicated by superscripts [a] and [b] respectively.

Table 4

Literature data on blood serum levels (µg/L) of *pp*'DDE and total DDT in Brazilian adults.

Exposure	State	Sample size	*pp*'DDE	Total DDT	Reference
GP	RJ	30	155.0+	336.0+	Almeida, 1972
GP	ES	11	117.0+	194.0+	Almeida, 1972
GP	SP	42	23.9+	–	Fernícola & Azevedo, 1982
GP	GO	51	10.0-1000.0++	–	Lara et al, 1987
GP	BA	50	8.3+	14.3+	Carvalho, 1991
GP	SP	16	14.3+	16.1+	Minelli & Ribeiro, 1996
AW	BA	10	16.0+	18.1+	Carvalho, 1991
AW	BA	19	18.0+	18.0+	Carvalho, 1991
VC	BA	15	47.7+	112.8+	Carvalho, 1991
VC	BA	14	344.4+	702.7+	Carvalho, 1991
VC	SP	26	64.3+	76.9+	Minelli & Ribeiro, 1996
AW	**RJ**	**26**	**nd - 4.4++**	**nd - 4.4++**	**This study, 1997**

+ mean values ++ minimum and maximum values; GP: general population; AW: agricultural workers; VC: vector control workers; SP: São Paulo; BA: Bahia; RJ: Rio de Janeiro; ES: Espírito Santo; GO: Goiás; nd: not detected.

Table 5

Literature data on blood serum levels (µg/L) of β-HCH in Brazilian adults.

Exposure	State	Sample size	β-HCH	Reference
GP	SP	42	7.3+	Fernícola & Azevedo, 1982
GP	BA	50	2.6+	Carvalho, 1991
GP	SP	16	3.4+	Minelli & Ribeiro, 1996
HCA	RJ	31	1.0 - 207.3 ++	Braga, 1990
AW	BA	6	20.2+	Carvalho, 1991
AW	BA	7	86.7+	Carvalho, 1991
AW	BA	6	84.0+	Carvalho, 1991
AW	BA	10	3.7+	Carvalho, 1991
AW	BA	19	8.0+	Carvalho, 1991
VC	BA	15	6.0+	Carvalho, 1991
VC	BA	14	13.8+	Carvalho, 1991
VC	SP	26	32.0+	Minelli & Ribeiro, 1996
AW	**RJ**	**26**	**nd - 5.3++**	**This study, 1997**

+ mean values ++ minimum and maximum values; GP: general population; HCA: people living in a highly contaminated area (Cidade dos Meninos, Duque de Caxias, RJ); AW: agricultural workers; VC: vector control workers; SP: São Paulo; BA: Bahia; RJ: Rio de Janeiro; nd: not detected.

Other organochlorine pesticide residues

Other cyclodiene pesticide residues, such as endrin (biological half-life = 20 hours) and heptachlor-epoxide, the half-life of which is long and comparable with that of dieldrin (Tordoir and van Sittert, 1994), were not found in any sample of the present study. The fungicide hexachlorobenzene was not detected in any sample either.

Conclusions

In conclusion, data from the present study suggest that agricultural workers from Paty-do-Alferes, Rio de Janeiro State, are not occupationally exposed to highly persistent organochlorine pesticides such as DDT (biological half-life in adipose tissue = 3.4 years; Tordoir and Sittert, 1994), technical grade HCH, aldrin/dieldrin, heptachlor, or HCB. The low levels of *pp*'DDE and β-HCH found in Paty-do-Alferes workers probably resulted from intake of residues found in food. It is well known that baseline levels of persistent OCPs vary from place to place and that dietary habits (i.e., daily intake of meat, eggs, milk, and other dairy products) play an important role in the contamination of non-occupationally exposed individuals. At any rate, serum concentrations of organochlorine pesticide residues found in agricultural workers from Rio de Janeiro State are comparable to or even lower than blood levels reported for the general population in Brazil and elsewhere.

Acknowledgments

This study was supported by grants from PADCT-II (CIAMB-FINEP), DESUSMO/European Union, and FNMA. IFD and FJRP are recipients of research fellowships from CNPq (Brazilian National Research Council). The stimulating cooperation with the staff of the Municipal Health Secretariat in Paty-do-Alferes is gratefully acknowledged.

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