

Organochlorine Pesticide Levels in the Brazilian army conscripts of the city of Rio de Janeiro, Brazil

M.C. Cardeal De Oliveira^{1*}; P. Barrocas^{2**}; J. Moreira^{2**}; V.R. Rossi Lemes¹; T.A. Kussumi¹; V.E. Nakano¹; S. Bio Rocha¹; I. Albuquerque Kimura¹; R. Kuno³; N. Gouveia⁴

1-Instituto Adolfo Lutz (IAL), State Health Department of São Paulo, Brazil;

2- Fundação Oswaldo Cruz , Ministry of Health, Rio de Janeiro, Brazil **Coordinators of Project Brazilian Army Conscripts

3-Environmental Agency of the State of São Paulo. São Paulo, Brazil;

4-School of Medicine, University of São Paulo (USP) São Paulo, Brazil;

**e-mail: maria.oliveira@ial.sp.gov.br; celestecardeal@yahoo.com.br*

INTRODUCTION

Human exposure to toxic chemicals has been occasioned by increased agricultural production, industrialization and urbanization. Taking into consideration their severe effects, a cross-sectional study for the analysis of persistent organochlorine compounds (OCPs) was performed with army conscripts of the metropolitan area of Rio de Janeiro City. It was applied a questionnaire and collected biological material (venous blood). Persistent organochlorines (aldrin, dodecachlor, dieldrin, dichlorodiphenyltrichloroethane (DDT), endrin, heptachlor, hexaclorociclohexane (HCH), hexachlorobenzene (HCB) are included in the POPs list, Stockholm Convention on Persistent Organic Pollutants (POPs⁸, 2001). Rio de Janeiro is one of the world's highest urban population densities. Human biomonitoring is a tool in environmental medicine to assess the level of internal exposure of population, groups and individuals to environmental pollutants. Army conscripts constitute a group of healthy individuals and researches have shown that they adequately represent control individuals (Cerná³, 2007).

OBJECTIVES

The study had as objective to evaluate exposure by persistent organochlorinated residues in blood of Brazilian army conscripts living in the city of Rio de Janeiro and provide subsidies for the realization of the First National Survey of Populations Exposed to Chemicals.

METHODS

Study Population

Biological monitoring was conducted in blood serum (N = 306 analyzed samples) of young men, 18 to 25 years old who are residents of the city of RIO DE JANEIRO, collected in five Army Selection Committees.

Sample collection

Blood samples (about 10 mL were collected in "vacutainer" tube without anticoagulant addition and then sent immediately to the laboratory under refrigeration. After centrifugation, serum samples were transferred to a glass tube with screw cap and PTFE stopper and kept in the freezer until analysis.

Ethical aspects

All procedures complied with the ethical guidelines (Brasil¹, 2012).

Questionnaires

For support the interpretation of the analytical results, questionnaires were applied to obtain socio-demographic and economic characteristics, race, education, birthplace, age, work, residence time and variables (past occupations involving pesticides, animal farming for consumption, greenery garden, living near industrial waste or agricultural area and water source used).

Analytical method

The used method was "Hexane extractable chlorinated insecticides in human blood" (Dale⁶, 1966) with modifications as described in Manual of Analytical Methods for the Analysis of Pesticide Residues in Human and Environmental Samples. External calibration curves at least 7 levels of concentrations for each pesticide were used to quantify pesticide residues, within the linear range of detector (**Table 1**): 0,01 - 0,4 μ g/dL for HCB, α -HCH, γ -HCH, δ -HCH; 0,04 - 1,0 μ g/dL for β -HCH; 0,02 - 0,8 μ g/dL for heptachlor, heptachlor epoxide (cis e trans), pp'DDE, dieldrin; 0,03 - 1,6 μ g/dL for op'DDE, op'DDD, pp'DDD, op'DDT, pp'DDT and 0,04 - 1,7 μ g/dL for dodecachlor.

Table 1 CG/ μ ECD conditions for organochlorine pesticides and metabolites assays

	Column	Instrument	Chromatographic conditions
Identification and Quantification	VF-5MS (5% phenyl 95% dimethylsiloxane) fused-silica capillary column (30 m, 0.25 mm i.d., 0.25 μ m film thickness)	GC Agilent 6890/ Detector μ ECD	Injector temperature: 250°C, Detector temperature: 310°C, Oven temperature programmed for quantification: 60°C (3min.), 20°C/min to 200°C, 3°C/min to 280°C, 290°C (20min); Flow of carrier gas N ₂ : 1mL/min, Mode and injection volume: splitless, 2 μ L.
Confirmation	VF-35MS (35% phenyl 65% dimethylsiloxane) fused-silica capillary column (30 m, 0.25 mm i.d., 0.25 μ m film thickness)	CG Thermo Scientific Trace GC Ultra / Detector ECD	Injector temperature: 250°C, Detector: ECD, temperature: 310°C, Oven Temperature programmed for quantification: 60°C (3min.), 10°C/min to 220°C, 3°C/min to 280°C; Flow of carrier gas N ₂ : 1mL/min, Mode and injection volume: splitless, 2 μ L.

RESULTS AND DISCUSSIONS

It was investigated the correlation of data collected through questionnaires with exposure and risk factors. The results obtained did not reveal a health-risk situation for conscripts. The conscripts blood sampled showed a similar profile in terms of ethnic groups, birthplace, and socioeconomic variables. Almost all conscripts lived in urban areas and therefore were exposed to the impacts and activities characteristics of urban centers (industrial, automobile traffic, etc.) as opposed to activities developed in agricultural areas. Most of the participants did not live near factories nor own a garden/orchard in their home. All medians results had lower levels of organochlorine pesticides than LOQs (0.02 to 0.16 $\mu\text{g}/\text{dL}$) (**Table 2**); β -HCH, and pp'DDE were found with high frequencies and maximum levels of 0.01 $\mu\text{g}/\text{dL}$ and 0,15 $\mu\text{g}/\text{dL}$, respectively (**Table 2**). After body absorption, DDT is distributed into tissues and the highest concentrations are found in adipose tissues due to high lipid solubility. DDT is slowly metabolized to op' and pp'DDE, DDA and DDD isomers. DDE is stored in adipose tissue and excreted via the bile, urine, and milk (Klaassem⁷, 2007). No other organochlorines were found. Studies have shown that pp'DDE levels are in blood serum of exposed and unexposed population (Cruz⁵, 2001; Charlier⁴, 2002). Higher levels of pp'DDE regarding DDT isomers indicate previous exposure (Zhou¹⁰, 2006). In Brazil, there are no reference values for organochlorines pesticides in human serum (Brasil², 1994).

Table 2. Concentrations of persistent organochlorine pesticides in army conscripts blood serum ($\mu\text{g}/\text{dL}$)

Organochlorine pesticides	Mean (x)	SD	Median (Md)	Geometric Mean (MG)	Minimum value	Maximum value	N	LOQ	Adult population not exposed	
									References	
									Turci ⁹ et al, 2010 (Novafeltria, Pavia, Italia)	Cruz ⁵ and Lino 2001 (Coimbra, Portugal)
Hexachlorobenzene	0.01	0.000	0.01	0.01	0.01	0.01	306	0.02	0.01 (Md)	-
α -HCH	0.01	0.000	0.01	0.01	0.01	0.01	306	0.02	ND (LD=0.02)	0.76 (mean)
β -HCH	0.02	0.000	0.02	0.02	0.02	0.02	306	0.04	0.05 (Md) 0.12 (máx)	0.16 (mean) 0.15 (max)
γ -HCH	0.01	0.000	0.01	0.01	0.01	0.01	306	0.02	0.01 (Md) 0.01 (max)	0.07 (mean) 0.48 (max)
δ -HCH	0.01	0.000	0.01	0.01	0.01	0.01	306	0.02	-	-
Heptachlor	0.02	0.000	0.02	0.02	0.02	0.02	306	0.04	-	-
Heptachlor epoxide	0.04	0.000	0.04	0.04	0.04	0.04	306	0.08	ND (LD=0.005)	-
Dieldrin	0.02	0.000	0.02	0.02	0.02	0.02	306	0.04	-	-
o,p' DDE	0.04	0.000	0.04	0.04	0.04	0.04	306	0.08	0.04 (max)	-
p,p' DDE	0.02	0.01	0.02	0.02	0.02	0.15	306	0.08	0.01 (Md) 0.20 (max)	0.16 (mean)
o,p' DDT	0.04	0.000	0.04	0.04	0.04	0.04	306	0.08	0.01 (Md)	1.54 (Md)
p,p' DDT	0.04	0.00	0.04	0.04	0.04	0.04	306	0.08	0.01 (Md) 0.07 (max)	1.88 (mean)
o,p' DDD	0.04	0.000	0.04	0.04	0.04	0.04	306	0.08	-	-
p,p' DDD	0.04	0.000	0.04	0.04	0.04	0.04	306	0.08	ND (LD=0.005)	1.20 (mean)
Dodecachlor	0.08	0.000	0.08	0.08	0.08	0.08	306	0.16	-	-

N = number of analyzed samples; SD: Standard Deviation; LOQ: limit of quantification; ND: Not detected; LD: limit of detection.

CONCLUSIONS

The data obtained in this study represent significant contribution to the knowledge of the levels of organochlorine pesticides in Brazilian army conscripts of the city of Rio de Janeiro, Brazil, and characterize a considerable information database of pesticide levels of control individuals residing in the metropolitan area of Rio de Janeiro. The results provide integrated actions to health control authorities in order to reduce human exposure to chemical contaminants, to prevent or minimize the risks and to encourage the implementation of the First National Survey of Populations Exposed to Chemical Substances, under general coordination of CGVAM / SVS / MS.

REFERENCES

1. Brasil Resolução CNS 466, de 12 de dezembro de 2012. Dispõe sobre as diretrizes e normas regulamentadoras de pesquisas envolvendo seres humanos. Diário Oficial da União 2011; 11 jun.
2. Brasil, Ministério do Trabalho. Secretaria de Segurança e Saúde no Trabalho. Portaria nº 24 (NR-7), de 29/12/1994, D.O.U. de 10/12/94, Seção 1: 21278-280.
3. Cerná M, Spěváčková V, Batáříová A, et al. Human biomonitoring system in the Czech Republic. *Int J Hyg Environ Health*. 2007; 210(1-4):495-9.
4. Charlier JC, Plomteux GJ. Determination of organochlorine pesticide residues in the blood of healthy individuals. *Clin Chem Lab Med*. 2002; 40 (4): 161-164.
5. Cruz S, Lino C, Silveira MI. Evaluation of organochlorine pesticide residues in human serum from in a urban and two rural populations in Portugal. *The Science of the Total Environment* 2001; 117:21-15.
6. Dale, W.E., Curley, A. and Cueto, C., Hexane extractable chlorinated insecticides in human blood., *Life Sciences*, 5:47, 1966. In *Manual of Analytical Methods for the Analysis of Pesticides in Human and Environmental Samples*, Environmental and Protection Agency, 5A, (1), (a), rev. 1980.
7. Klaassem CD editor. *Casarett and Doull's Toxicology The Basic Science of Poisons*, New York, 17th Ed., Mac Graw Hill Medical, 2007.
8. POPs - Persistent Organic Pollutants, 2001: Text of the Convention of POPs, Stockholm, Sweden, 22 May 2001. <http://chmpps.int/TheConvention/Overview/TextotheConvention/tabid./2212/Default.aspx>. Access: nov. 2018
9. Turci R. et al. A simple and fast method for the determination of selected organohalogenated compounds in serum samples from the general population. *Toxicology Letters*, 192 (2010) 66-71.

10. Zhou R, Zhu L, Yang K, Chen Y. Distribution of organochlorine pesticides in surface water and sediments from Qiantang River, East China. *Journal of Hazardous Materials A*. 2006; 117: 68-75.

Acknowledgments: Irani C. Silva and Valéria AC Diago, research trainees, Reinaldo A. Ribeiro, Ana LR de Faria, Kennia C. Waldhelm and Antonia L. Silva, IAL technical team; Rogério R. Prado, statistical analyzes and teams of sample collection and interviewers.