

Levels of DDT, HCH, and HCB Residues in Human Blood in Ahmedabad, India

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Received: 6 June 2003/Accepted: 15 October 2003

Organochlorine compounds released into the environment enter living organisms and become biologically concentrated. These chemicals have been implicated in the pathogenesis of many adverse health outcomes (Wolff and Toniolo 1995, Longnecker et al. 1997, Jaga and Brosius 1999). Assessing human exposure to these chemicals through biological monitoring offers a meaningful criteria to examine the magnitude of potential health risk (Nigam et al. 2000). Worldwide, the residues of these persistent chemicals have been identified in humans and environment (ATSDR 2000; Walker et al. 2003), however, higher levels of DDT and its metabolites in human blood samples from India were reported as compared to the other parts of the world (Sharma and Bhatnagar 1996; Bhatnagar 2001).

In view of the fragmentry reports on the serum level of organochlorine insecticides in general population from different cities in India (Bhatnagar 2001), and very importantly, the restriction imposed on the use of DDT and lindane (γ isomer of HCH) in recent years, the present work was an attempt to describe the preliminary results of our analysis on the residues of persistent organochlorine insecticides in the human blood who live in an area of urban zone of Ahmedabad, India.

MATERIALS AND METHODS

The material examined in this study was human blood samples from 18 male healthy volunteers of Ahmedabad (urban) area, who participated after giving their informed consent. Blood samples were analyzed for the presence of residues of DDT, HCH and HCB. Subjects were requested to provide information on their demographics, dietary habits and smoking status.

Standard reference materials (SRM) grade of pp'-DDT, pp'-DDE, op'-DDT, pp'-DDD, α -HCH, β -HCH, γ -HCH and HCB were obtained from M/S AccuStandard, USA. Organic solvents used in the present study were glass distilled and checked for any pesticide contamination. All glassware were washed with detergent, rinsed with water, dipped in chromic acid for 24 hr and finally rinsed with distilled water and then hexane.

Blood sample (6-8 ml) was drawn from medial cubital vein with the help of sterilized syringe into appropriate vial. Serum was obtained by centrifuging blood sample at 3,000 rpm for 10 min and stored at -20°C until analysed. The method of Dale et al. (1966) was employed for the analysis of organochlorine pesticides. Blood serum (2 mL) was extracted with 6 mL hexane in round bottom glass stoppered test tube for 2 hr on slow speed rotating machine. 5 mL aliquot of hexane layer was quantitatively transferred to an graduated stoppered tube and concentrated under stream of N2 in a waterbath at 40°C. The residue was dissolved in an appropriate volume of hexane. A suitable aliquot was injected into Gas Chromatograph equipped with 63Ni electron capture detector. Perkin Elmer GC 8700; Capillary VIT silica column containing 25 QCZ/OV 1701, length - 25 m, id -0.2 mm; carrier gas argon/methane (90/10) at 15 PSIG, injector port temp 220°C, detector temp 275°C, column temp 215°C, chart speed 2 cm/min. Quantitative analysis of the each pesticide residue was determined by retention times on the chromatograms and identified by comparison with known standards. The recoveries of various residues were also calculated which ranged from 88 % to 102 %. In addition, the fortified samples studied as a part of routine, internal analytical quality assurance procedure. Statistical analysis was carried out by using software SPSS Release 6.1.4.

RESULTS AND DISCUSSION

Eighteen serum samples were analysed by GC-ECD for the residues of DDT, HCH and HCB and their results are shown in Table 1. Serum level of pp'-DDE, op'-DDT, pp'-DDD and pp'-DDT ranged from 10.43-38.33, 0.42-2.41, 0.77-4.43, 3.66-24.06 with a mean of 20.85, 1.15, 2.03 and 9.28 µg/L respectively. However, the Σ DDT (equivalent sum of pp'-DDE, op'-DDT, pp'-DDD and pp'-DDT) content in serum samples had a mean of 32.61 µg/L and ranged from 21.17-54.47 $\mu g/L$. pp'-DDE was the major metabolite and it contributes about 64 % of the Σ -DDT. The ratio of DDE to Σ DDT ranged from 0.39 to 0.93 (mean \pm SE; 0.64 \pm 0.03) and followed the normality pattern as shown in Fig 1. The DDE to Σ DDT ratio may represent an indicator of the extent of DDT degradation and its metabolic transformation. Exposure to DDT in nonoccupationally exposed subjects as indicated by their plasma DDE, was most reliably predicted by age and serum cholesterol level (Laden et al. 1999). More recently, DDE has been shown to be a potent androgen receptor (AR) antagonist (Kelce et al. 1995). The possible impacts on human health of these hormonally active substances at the low but with prolonged exposure are less studied. In addition, the findings of Beard et al. (2000) suggested that past exposure to DDT may be associated with reduced bone mineral density in women.

All the samples had presence of the three residues of HCH. Levels of α -HCH, β -HCH and γ -HCH in serum samples had a mean of 4.49, 35.06 and 1.69 μ g/L respectively. β -HCH contributed about 85 % of the total HCH (equivalent sum of α -HCH, β -HCH and γ -HCH). HCB was identified in 7 samples in range of 0.13 -

Table 1. Organochlorine insecticides in human serum samples ($\mu g/L$).

Compound	No*	Range	Median	Mean ± SE
pp'-DDE	18	10.43 - 38.33	20.74	20.85 ± 1.84
op'-DDT	15	0.42 - 2.41	0.99	1.15 ± 0.12
pp´-DDD	18	0.77 - 4.43	1.60	2.03 ± 0.28
pp'-DDT	17	3.66 - 24.06	7.65	9.28 ± 1.30
∑ DDT	18	21.17 - 54.47	29.63	32.61 ± 2.32
α-НСН	18	1.00 - 9.16	3.62	4.49 ± 0.73
β-НСН	18	20.11 - 82.09	30.25	35.06 ± 3.50
у-НСН	18	0.72 - 3.09	1.54	1.69 ± 0.15
ΣHCH	18	22.55 - 91.06	37.77	41.23 ± 3.77
HCB	7	0.13 - 0.27	0.21	0.20 ± 0.02

^{*} Number of positive samples

 $0.27~\mu g/L$ having an average of $0.2~\mu g/L$. In a study conducted in general population in Canada, Mes (1992) reported median and maximum whole blood levels of HCB as 0.11 and 0.34~ppb.

The observed trend for total DDT and total HCH are comparatively lower than the earlier reports from India (Bhatnagar 2001) which may be due to the restriction on use of these pesticides in agriculture but the levels are still higher than the studies originating outside India (Mes 1992; Gammon et al. 1997; Hanrahan et al. 1999; Laden et al. 1999; Hanaoka et al. 2002) In fact, we could not find any age dependent trend on the accumilation of these chemicals, however, the factors that may influence the storage and bioacculmlation of these chemicals are intensity and duration of exposure, efficiency of absorption, sex, nutritional status and lipid content of the organ. Certain persistent environmental chemicals termed as endocrine disruptors are known to elicit their adverse effects by mimicking or antagonizing natural hormones in the body which are responsible for maintaining homeostasis and controlling normal development (Crisp et al. 1998). The toxicological implications of the observed findings could not be examined very accurately as the sample size is small, however, it serves as a basis of diagnostic values for epidemiological work on exposure assessment to persistent organic pollutants in general population in developing countries. The food chain is the main source of exposure to organochlorine residues in human body and the available data on food samples including dairy products, livestock meat and water samples indicate the presence of these contaminants in significant amounts (Kannan et al. 1992; Kashyap et al. 2002). Although the benefits of pesticides are undeniable, the preventive measures such as use of protective devices and educational modules should be adopted to reduce the existing body burden in order to avoid any potential health effects subsquent to their use. These results, coupled with the

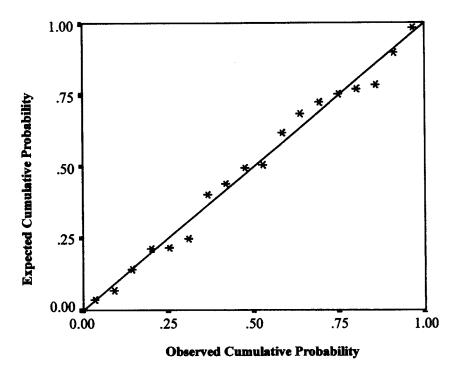


Figure 1. Relationship between observed cumulative probability and expected cumulative probability for the ratio of pp'-DDE to \sum DDT

paucity of data on health effects following low dose long term exposure of these pollutants warrant initiatives to reduce the use and release of organochlorine contaminants.

Acknowledgments. We thankfully acknowledge participation of study volunteers and the technical assistance of Mr. M.R. Variya. Thanks are due to Mr. P.M. Chako for secretarial assistance.

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