Available online at www.pelagiaresearchlibrary.com

Pelagia Research Library

European Journal of Experimental Biology, 2011, 1 (3):162-168

ISSN: 2248 –9215

Hexachlorohexane (HCH) & Dichlorodimethyltrichloroethane (DDT) in soils from Northern Uttar Pradesh, India

Meenu Mishra, Bhupander Kumar*, S. K. Singh, Gargi Goel, Richa Gaur, Dev Prakash, Sanjay Kumar and C. S. Sharma

National Reference Trace Organics Laboratory, Central Pollution Control Board, East Arjun Nagar, Delhi, India

---

ABSTRACT

HCH and DDT, the organochlorine pesticides (OCPs) were measured in soils from northern Uttar Pradesh, India. The average concentration of HCHs and DDTs was 7.58±1.54 ng g^{-1} *and 0.61±0.21 ng g*$^{-1}$ *(dry wt.). Compositional analysis of HCH and DDT isomers was carried out to identify the possible contamination sources. The* α/γ *ratio of HCH (<0.01 - 8.83) reflects the use of technical as well as lindane formulation. The ratio of p,p'-DDT/p,p'-DDE (0.73) and p,p'-DDT/∑DDT (0.42) indicates the combined effects of past and ongoing usage of DDT. o,p'-DDT/p,p'-DDT (<0.01) ratio suggest the contamination of soils from technical DDT and not from Dicofol type DDT.*

Keywords: HCH, DDT, soil, Accelerated Solvent Extraction (ASE).

INTRODUCTION

Widespread attention has been given to the pollution by persistent organic pollutants (POPs) including some organochlorine pesticides. Organochlorine pesticides (OCPs), such as HCH and DDT are persistent, toxic and bio-accumulative in nature [1-2]. These are long range transport pollutants and can be transported to regions far from their original sources, such as the Arctic [3-4]. OCPs have a wide range of acute and chronic health effects, including cancer, neurological damage, reproductive disorders, immune suppression, birth defects, and are also suspected endocrine disruptors [5].

Their physico-chemical characteristics, which include hydrophobicity and resistance to degradation, make these chemicals ultimately to accumulate in soils and sediments [6-8]. OCPs have a great affinity for particulate matter so they can remain in soil and sediments for very long due to their long half-life times.

In May 2004, Stockholm Convention on POPs entered into force with the intention of reducing, and ultimately eliminating these pollutants. As is well known, DDT was listed by the Stockholm Convention as 1 of 12 persistent organic pollutants (POPs) in 2004, and more recently, α-HCH, β-HCH, and γ-HCH (lindane) were added to the list in 2009 [9]. As a party to the Convention, India is legally obligated to abide by the objectives of the treaty, and is encouraged to support research on POPs. This study was aimed to evaluate persistent organochlorine pesticide (HCH and DDT) concentrations in soils from northern Uttar Pradesh using accelerated solvent extraction (ASE) and GC-ECD.

MATERIALS AND METHODS

Sampling

Soil samples in duplicate collected from Meerut and Ghaziabad districts in northern Uttar Pradesh. Approximately 1 Kg. of soil sample was collected using stainless steel auger, and after removing pebbles and wood sticks the sample was mixed thoroughly to homogenized, then an aliquot was transferred to clean wide mouth amber glass bottle. After proper labeling the sample bottles were transported to laboratory and kept at -4^{0}C until further chemical treatment.

Chemicals and Solvents

Chemicals and solvents were purchased from Merck India. Silica gel 60 (0.063 – 0.100 mm) was from Sigma-Aldrich. Prior to use, silica gel and anhydrous sodium sulphate was cleaned separately with methanol, dichloromethane and acetone in Soxhlet extractor for 8 h each, and stored air tight at 130^0 C. Pesticide standard solutions were obtained from Supelco (Sigma, USA).

Sample Extraction

Soil samples were extracted using extraction procedure as per EPA's SW-846 Method 3545 [10]. Briefly, 15-20 g sample was homogenized and dried by mixing with diatomaceous earth (ASE prep DE, Dionex, USA) until a free-flowing powder was obtained. The extraction was carried out with accelerated solvent extractor (ASE-350, Dionex, USA) [11] using acetone: hexane (v/v, 1:1) in two cycles with 5 min. static time. The ASE was operated at 1500 psi and the oven was heated to 100^0 C. The extracts were concentrated to 2.0 ml using Rotatory Vacuum evaporator (Eyela, Japan). Moisture content was determined to report data on dry weight basis.

Chromatographic column cleanup

The multilayered silica gel column chromatography was performed for fractionation and to remove interfering organic and polar species. Briefly multilayered silica gel column (300 mm x 30 mm) was packed from bottom to up with 2.5 g silica gel, 4.0 g silver nitrate silica gel, 2.5 silica gel, 4.0 basic silica gel, 2.5 g silica gel, 12.0 g acid silica and 5.0 g anhydrous sodium sulphate. The column was pre-rinsed with 100 ml n-hexane before sample was loaded. The elution of analytes was subsequently carried out using 170 ml hexane and concentrated to 2.0 ml.

The eluted extract was concentrated using Rotatory Vacuum evaporator and under gentle stream of pure nitrogen using Turbo Vap (Caliper, USA) to 1.0 ml. The extract was transferred to auto sampler vial and 1 µl was injected onto a gas chromatograph equipped with an electron capture detector (GC-ECD) for quantification.

Instrumental analysis

In the present study DDTs (*p,p'*-DDE, *o,p'*-DDT and *p,p'*-DDT) and HCHs (α-HCH, β-HCH, γ-HCH, and δ–HCH) were analyzed in soils. Separation and Quantification of pesticides was carried out using Gas Chromatograph (Perkin Elmer, Clarus 500) attached with autosampler and equipped with an Electron Capture Detector (ECD, ^{63}Ni), on fused silica column 25 m x 0.20 mm id 0.33 µm film (Elite-1). The column oven temperature was initially maintained at 170^{o} C and programmed to 220^{o}C (7^{0}C min^{-1}) and again ramped to 250^{o}C at 5^{o}C min^{-1} and held for 6.86 min. The injector and detector temperature were maintained at 250^{o}C and 350^{o}C respectively. Purified nitrogen gas was used as carrier at the flow rate of 1.0 ml. min^{-1}.

Analytical quality control

Certified reference standards (Sigma, USA) were used to for quantification of pesticide. The pesticides were identified in the sample extract by comparing the retention time from the standard mixture and quantified using the response factors from five level calibration curves of the standards. Appropriate quality assurance quality control (QA/QC) analysis was performed, including analysis of procedural blanks (analyte concentrations were <MDL 'method detection limit'), random duplicate samples (Standard deviation <5), calibration curves with the r^2 value of 0.999, and matrix spike recovery 100±20%. Each sample was analysed in duplicate and the average was used in calculations. A reporting limit of > 0.01 µg kg^{-1} was taken for calculation. Levels below reporting limit or below MDL (<0.01 ng g^{-1}) were taken as zero (0) in the calculations. The results of the analysis are reported in ng g^{-1} dry weight (dry wt.) basis.

RESULTS AND DISCUSSION

Analytical results of HCH and DDT in soils from northern Uttar Pradesh, India are presented in Table 1 and Table 2. The average concentration of ∑OCPs in soils was 8.19±1.55 ng g^{-1} dry wt. with the range of 0.79-19.85 ng g^{-1} dry wt. Concentrations of ∑HCH in soils were higher than those of ∑DDT. The ∑HCH ranges from <0.01 to 17.68 with a mean value of 7.58±1.54 ng g^{-1} whereas the ∑DDT ranges from <0.01 to 2.97 with a mean value of 0.61±0.21 ng g^{-1}. The properties of higher water solubility, vapor pressure, biodegradability, lower lipophilicity and particle affinity, of HCH relative to DDT [7] could account for the higher concentrations of HCH in soils. HCH isomers have been detected as a worldwide contaminant. Among the analysed OCPs compounds, α-HCH alone was the dominant pollutants (70.31%) with 5.76±1.25 ng g^{-1} (dry wt.) mean concentration, followed by γ-HCH (0.96 ng g^{-1} dry wt.), *p,p'*-DDE (0.32 ng g^{-1} dry wt.) and *p,p'*-DDT (0.25 ng g^{-1} dry wt.).

Table 1: Range and mean of HCH and DDT concentrations in soil

ng g^{-1} (dry wt.)	∑HCH	∑DDT	∑OCPs
Range	<0.01-17.68	<0.01-2.97	0.79-19.85
Mean±SE*	7.58±1.54	0.61±0.21	8.19±1.55

**SE (standard error) = SD/√n*

Table 2: HCH and DDT concentrations in soils

Compound	Range		Mean	Std. Err	%
	Min	Max			
α-HCH	<0.01	13.73	5.76	1.25	70.31
β-HCH	<0.01	1.48	0.55	0.12	6.68
γ-HCH	<0.01	2.27	0.96	0.19	11.77
δ-HCH	<0.01	0.99	0.31	0.10	3.75
p, p'-DDE	<0.01	1.44	0.32	0.12	3.96
o, p'-DDT	<0.01	0.73	0.04	0.01	0.47
p, p'-DDT	<0.01	2.00	0.25	0.03	3.06

**Std. Err (standard error) = SD/√n*

Differences in composition of HCH or DDT isomers in the environment could indicate different contamination sources [12]. Technical HCH has been used as a broad spectrum pesticide for agricultural purposes, which has been banned since 1996-97 in India. Technical HCH consists principally of four isomers, α-HCH (60-70%), β-HCH (5-12%), γ-HCH (10-15%), δ-HCH (6-10%), while lindane contains >99% of γ-HCH [13-14]. . The average concentration of α-HCH, β-HCH, γ-HCH and δ-HCH in this study was 5.76±1.25 ng g^{-1}, 0.55±0.12 ng g^{-1}, 0.96±0.19 ng g^{-1} and 0.31±0.10 ng g^{-1}, respectively (Table 2). The composition of HCH isomers observed in the present study reveals α=76%; β=7%; γ=13%; δ=4% was similar to the technical grade composition, which indicates technical HCH usage. The ratio of α-HCH to γ-HCH has been used worldwide to identify the possible HCH source. The ratio of α-HCH to γ-HCH between 3 and 7 is indicative of fresh input of technical HCH [15]. However, a lindane source will show the reduce ratio close or <1 [16-17]. In this study the ratio of α-HCH to γ-HCH isomers (α/γ ratio) is ranged between <0.01 and 8.83, with pooled mean value of 5.76 (Table 3). These ratios reflect the regular usage of technical HCH and lindane. Similar ratios of α/γ were reported for Indian environment by other workers [18-21]. The technical mixture of HCH was produced and used in India until it was banned in 1997, and lindane formulations are registered for use in public health practices to control vector borne diseases and for pest control in selected crops [22].

It is to note that, in the mixture of past technical HCH and current lindane application, a significant correlation between α–HCH and γ–HCH was still observed in the studied soil samples (Figure 1). Moreover, the α–HCH in higher concentration indicated a fresh source of α–HCH, contrary to the past application of technical HCH. The explanation of transformation of γ–HCH to α–HCH is the best in terms of maintaining the correlation between each other. In particular, γ–HCH may be transformed under ultraviolet radiation and through biological degradation in soil into α–HCH [23-24]. The studied area is located under the Tropic of Cancer line with strong ultraviolet radiation. Our results suggested that the transformation from γ–HCH into α–HCH under such conditions may be significant.

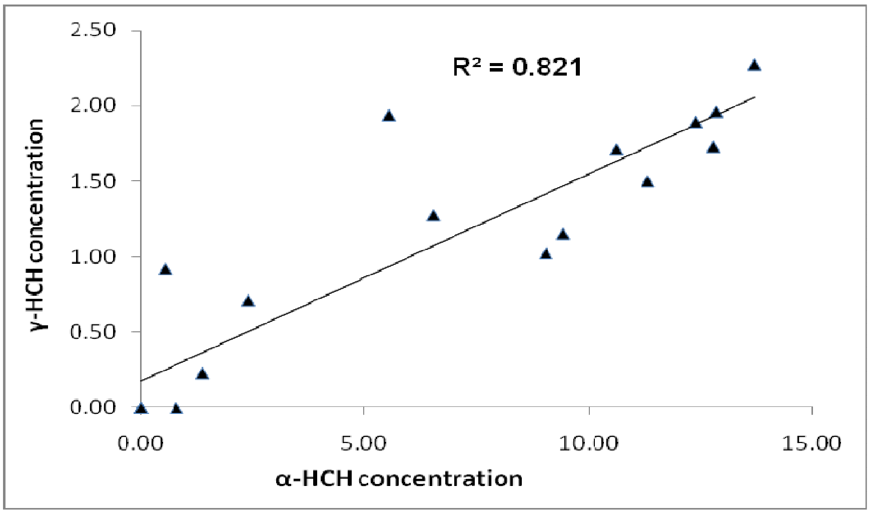

Figure 1: Relationship of α–HCH and γ–HCH in soils samples

In the present study, occurrence of DDT isomers was on order as: *p,p'*-DDE > *p,p'*-DDT > *o,p'*-DDT with average concentration of 0.32±0.12 ng g^{-1}, 0.25±0.03 ng g^{-1} and 0.04±0.01 ng g^{-1}, respectively (Table 2). The relatively high concentration of DDTs than DDE in this area indicated either that there was minimal degradation of DDT or there has been more recently input of technical DDT. DDTs can be volatilized to an ambient environment in a few days [25]. The vapor pressure of *o,p'*-DDT is 7.5 times higher than *p,p'*-DDT leading to greater volatilization of *o,p'*-DDT to the atmosphere [26], and *p,p'*-DDT metabolizes much faster in a subtropical environment. After DDT application, much of the DDT may be converted to *p,p'*-DDE. Elevated concentrations of *p,p'*-DDE have been interpreted as a result of its conversion to *p,p'*-DDE by UV radiation during atmospheric transport [27]. The higher concentration of *p,p'*-DDT and *p,p'*-DDE have been observed in all the samples, reflects aged mixture of DDT usage.

Table 3: Range and mean ratio of HCH and DDT isomers in soils

Isomer ratio	Ratio			
	Min	Max	Mean	Std. Err*
α/γ HCH	0.58	8.83	5.76	0.53
p,p'-DDT/∑DDT	<0.01	1.00	0.42	0.11
p,p'-DDT/*p,p'*-DDE	<0.01	2.30	0.73	0.26
o,p'-DDT/*p,p'*-DDT	-------------------- <0.01 ---------------------			

**Std. Err (standard error) = SD/√n*

The residence time of *p,p'*-DDT could be estimated using the ratio of *p,p'*-DDT to ∑DDTs. The ratio for technical DDTs was reported to be 0.77 [28]. The mean ratio of *p,p'*-DDT to ∑DDTs in present study was 0.42, which indicates that these areas have been contaminated with past usage of DDTs. The ratio of *p,p'*-DDT and *p,p'*-DDE used to estimate whether recent inputs of technical DDT exist. Generally, ratio of *p,p'*-DDT and *p,p'*-DDE higher than 0.33 indicates continuous usage of DDT. In this study the ratio of *p,p'*-DDT/*p,p'*-DDE was 0.73 (mean) so, we anticipate that inputs of DDTs are still exists in the study area. The ratio of *o,p'*-DDT/*p,p'*-DDT can be used to distinguish technical DDT from "Dicofol-type DDT". The *o,p'*-DDT/*p,p'*-DDT ratio was reported to be 0.2~0.26 in technical DDT and ~7.5 in dicofol products [29]. In our study the ratio of *o,p'*-DDT/*p,p'*-DDT is less than unity, which is different from the scenario in

China where dicofol usage is a major source of DDT [30]. Thus, the DDT contaminations in this region may be from technical DDT and obviously not from dicofol usage. Since 1996, DDT was banned as an agricultural pesticide [31], however, permitted to use up to 10,000 tonnes per year, under the Stockholm Convention, until an alternative can be found to combat vector borne diseases which are particularly prevalent in monsoon season [32]. Thus, the possible sources of DDTs are the combined effect of past and ongoing use in vector control or from sprays on open dumping sites.

CONCLUSION

The analytical data indicates, besides lindane, technical HCH mixtures are also used in the study area, and possible sources of DDTs are the combined effect of past and ongoing usage. Intensive assessment for persistent organic pollutants (POPs) is recommended, due to environmental health concern.

Acknowledgements
The authors express their sincere gratitude to the Member Secretary and Chairman of Central Pollution Control Board, Ministry of Environment & Forest Government of India to conduct the study.

REFERENCES

[1] Kannan K., Tanabe S., Tatsukawa R. *Environ. Sci. & Tech.*, **1995,** 29:2673-2683.
[2] Shegunova P., Klanova J. and Holoubek I. *Environ. Poll.*, **2007,**146: 257-261.
[3] Halsall C. J., Bailey R., Stern G. A., Barrie L. A., Fellin P., Miur D. C. G., Rosenberg B., Rivinsky F. Y., Kononov E. Y. and Pastukhov B. *Environ. Poll.*, **1998,**102: 52-62.
[4] Becker B., Halshall C. J., Tych W., Kallenborn R., Schiabch M., Mano S. *Atmospheric Chemistry and Physics: Discussions*, **2009,** 9: 515-540.
[5] Wang X., Wang D. Qin X. and Xu X. *J. Environ. Sci.*, **2008,** 20: 1090-1096.
[6] Yang R., Jiang G., Zhou Q., Yuan C., Shi J. *Environ. Int.*, **2005,** 31 (6): 799-804.
[7] Hong S. H., Yim U. H., Shim W. J., Oh J. R., Viet P. H., Park P. S. *Chemosphere*, **2008,** 72: 1193-1202.
[8] Hu L., Zhang G., Zheng B., Qin Y., Lin T., Guo Z. *Chemosphere*, **2009,** 77: 663-672.
[9] Hu W., Wang T., Khim J. S., Luo W., Jiao W., Lu Y., Laile J. E., Chen C., Zhang X., Giesy J.P. *Arch. Environ. Conta. Toxicol.*, **2010,** doi:10/1007/a00244-009-9455-z.
[10] US, EPA method 3545, **1995,** EPA SW-846, U.S. Government Printing Office, Washington, DC.
[11] Application Note 352, **1999,** Dionex, Sunnyvale, CA.
[12] Doong R. A., Peng C. K., Sun Y. C., Liao P. L. *Mar. Poll. Bull.*, **2002,** 45: 246-253.
[13] Qui X. H., Zhu T., Li J., Pan H. S., Li Q. L., Miao G. F., Gong J. C. *Environ. Sci. Technol.*, **2004,** 38: 1368-1374.
[14] Zhou R., Zhu L., Yang K., Chen Y. *J. Haz. Mat.*, **2006,** Doi: 10.1016/jmazmat.2006.02.005
[15] Yang Y., Li D., Mu D. *Atmospheric Environment*, **2008,** 42: 677-687.
[16] Iwata H., Tanabe S., Sakai N., Tatsukawa R. *Environmental Science & Technology*, **1993,** 27: 1080–1098.
[17] Willet K. L., Ulrich E. M., Hites H. A. *Environ. Sci. Technol.*, **1998,** 32 (15): 2197-207.

[18] Zhang G., Chakraborty P., Li J., *et al*. *Environ. Sci. Technol*., **2008**, 42 (22): 8218-8223.
[19] Chakraborty P., Zhang, G., Li, J., Xu, Y., Liu, X., Tanabe, S., and Jones, K. C. *Environ. Sci. Technol*., **2010**, 44: 8038-8043.
[20] Kumar B., Kumar S., Mishra M., *et al. Organohalogen Compounds* **2008**, 70:1120-1123.
[21] Kumar B., Gaur R., Goel G., Mishra M., Dev Prakash, Singh S. K., Lal R. B., Sanjay Kumar, and Sharma C. S. *Asian J. Scientific Res*., **2011,** 4 (3): 271-280.
[22] Gupta P. K. *Toxicology,* **2004,**198 (1-3):83-90
[23] Benezet H. J., Matsumura F. Nature, **1973,** 243: 480-501.
[24] Malaiyndi M., Shah S. *J. Environ. Sci. & Health*, **1980,**19: 887-910.
[25] Atlas, E., Giam, C. S. *Water, Air, Soil, Pollution*, **1988,** 38: 19-36.
[26] Spencer W. & Cliath M. M. *J. Agric. Food Chem.*, **1972,** 20(3): 645-649.
[27] Talekar N. S., Sun L. T., Lee E. M., Chen J. S. *J. Agric. Food Chem*., **1977,** 25: 348-352.
[28] World Health Organization (WHO). *Environ. Health Criteria,* **1989,** 83.
[29] Qui X. H., Zhu T., Tao B., Hu J., Hu S. *Env. Sci. Technol*., **2005,** 39: 4385-4380.
[30] Liu J., Zhu, T., Wang, F., Qiu, X. H., Lin, W. L. *Ecot. Env. Saf.*, **2006,** 63: 33-41.
[31] Battu R. S., Singh B., Kang B. K. *Ecotoxicol. Environ. Safety,* **2004,** 9: 324-331.
[32] Sharma V. P. *Current Science*, **2003,** 84: 513-515.