Int. J. Environ. Res., 2(1): 43-48, Winter 2008
ISSN: 1735-6865

Solid phase extraction of pesticide residues in water samples: DDT and its metabolites

Asi, M. R. , Hussain, A. and Muhmood, S. T.*

Nuclear Institute for Agriculture and Biology, NIAB, Faisalabad, Pakistan

Received 15 Jan. 2007; Revised 23 July 2007; Accepted 30 Aug 2007

ABSTRACT: Solid phase extraction (SPE) cartridge embedded with octadecyl C_{18} chain bounded to silica particles were used for the isolation and trace enrichment of pesticides from water samples collected from cotton, rice growing and municipal areas. The eluents were analyzed with high performance liquid chromatography (HPLC) using methanol (100%) as mobiles phase at different UV-Visible wavelengths. DDT and its metabolites were found in all areas but were not present in all samples. Concentration of pesticide residues varies from sample to sample and was in the range of 0.017-1.06 ng ml^{-1}. Overall, recoveries ranged from 84%-91% for all target pesticides.

Key words: SPE, HPLC, DDT, Metabolites

INTRODUCTION

Pesticides are widely used to control pests that affect agricultural crops and pests in homes, yards and gardens. These chemicals generally are man-made organic compounds. The principal processes that influence their potential for loss from soil to groundwater are volatilization (and subsequent diffusion), decomposition, retention by the soil, and transport by water. In some cases, pesticides used for plant protection may be applied to soil before planting, while for others, the application is made after seeding. Usually, most of the applied chemicals come into contact with soil. Application is by spraying a liquid, broadcasting granular formulation, or fumigation. Concern about environmental and health issue related to pest control makes it important to understand the possible hazards associated with their use and to minimize risks.

Pesticides used on agricultural crops for control of weeds and insects can be migrated to surface water, local watersheds and ground water after application to crops or soil. Ground water pollution by agricultural chemicals has become growing concern in the world because 40-50 % of domestic drinking water is pumped from ground water resources. (Hallberg, 1989). Water pollution is the most damaging and has widespread environmental effect on agriculture production and human health. The food contamination study in Pakistan conducted by National Institute of Health, Islamabad in 1984 revealed that organochlorine pesticide residues were the most frequently present in food. There had been widespread distribution of DDT and is metabolite as residues in food Tsipi, *et al.*,(1999). Chlorinated pesticides have also been found by Perveen and Masud (1988) in sample of milk, feed and cattle drinking water drawn from Karachi Cattle Colony. Hussain, *et al.*, 1993, have also reported residues of DDT and its degrading products in human milk and adipose tissue samples in Faisalabad area. Therefore, it is great concern to determine the contamination level of ground and surface water for DDT and its metabolites, which are heavily sprayed in the past. Because of their widespread use, pesticides residues have been also found in drinking water (Kumari, *et al.*, 1996), ground water (Plonke, *et al.*, 1988 and Plonke and Glotfelty, 1989), surface water (Hinkley and Bidle, 1989 and Hussain, *et al.*, 1997), and fog water (Glotfelty, *et al.*, 1987). Insecticides utilized for cotton productions are intensive and can contribute to water quality problem (Crutchfield, *et al.*, 1992).

---

*Corresponding author: Email-asimuhammad@yahoo.co.uk

In the past, heavy use of organochlorine pesticide was practiced on field crops and malaria eradication program. Although the use of majority of them such as DDT and its metabolite have been banned but the long persistency of most of these has resulted in the continuing presence in the environment. However, these days some new less persistent pesticides such as organophosphorus are used for saving cotton from pests.

But these compounds are highly toxic to aquatic organisms (Kendel 1999). As DDT is persistent (Gaeb, *et al.,* 1975 and Billings and Bidleman, 1983) and was heavily sprayed, its residues may move from fields into ground water and pose health risk to ultimate users. They can reach surface water bodies in either dissolved or particulate form, which may impair the water quality and jeopardize the aquatic species. Concern about such adverse effects necessitates the present work for potential environmental impacts of the pesticides.

MATERIALS & METHODS

Agriculture is playing a pivot role in the economy of Pakistan. It has a very good water channel system in World to irrigate agriculture area. Maximum canals flow from Northern to Southern part of the country and irrigate vast area of agriculture farms. Maximum pesticides are used in the cotton, rice belt and vegetables to protect them. It is a great concern to check the water table, surface water and water supplies for pesticide contamination. Therefore, water samples were collected from different area of the country to analyze pesticides residues.

The attention was focused to do sampling from tube-well; hand pump or well installed near heavily sprayed fields areas. The main purpose of sampling was to assess the present situation of pesticides levels in water. In order to assess possible impact of pesticides on aquatic ecosystem and drinking water supplies, water samples were taken from cotton, rice belts and municipal areas (Table 1). Water samples collected from different localities were processed for extraction and clean up for multi-residues analysis according to the procedure of Tanabe, *et al.,* 2000. Solid phase extraction (SPE) was used for the clean up of the samples. Each sample was processed in triplicate to check reproducibility of results. 500 mL each filter water sample was passed through pre-conditioned SPE at a flow rate of 5 mL/min. After this cartridge was wash with 10 mL of distilled water then the cartridge was dried with air suction at a pressure of 80 mmHg for one hour. In order to check the dryness of pre-weight cartridge, they were weighed again. The target compounds collected on the cartridges were eluted successively with 3 mL acetone; 3 mL hexane and 3 mL ethyl acetate under reduce pressure. All these chemicals were redistilled by fraction distillation apparatus. The combined elutes were concentrated upto dryness under nitrogen stream and volume was adjusted to 1 mL with acetone.

The pesticide residues analyses were carried out by using high performance liquid chromatograph (Shimadzu LC-10 AS) attached with Class 10 A software. The column used was SUPELCOSIL LC-18, 25 cm x 4.6 mm, 5 μL, Supelco, USA; pressure at isocratic system, 59 kg/cm^2; flow rate, 0.5 mL/min; injection volume, 20 μL; oven temperature, 30 ^{o}C and methanol (100 %) HPLC grade was used in mobile phase.

RESULTS & DISCUSSION

The study involves the use of Solid Phase Extraction (SPE) cartridges for the extraction of organochlorine from water samples. The recoveries obtained for DDT and its various metabolites range between 84-91 % (Table 2). The overall recoveries for spiked level (2 ng/mL) in water for o, p$^{/}$-DDD, o, p$^{/}$-DDE, o, p$^{/}$-DDT, p, p$^{/}$-DDD, p, p$^{/}$-DDE, p, p$^{/}$-DDT were 85±5, 89±8, 90±4, 91±6, 90±3, 87±11, respectively and are comparable with the results reported by Ambrus, *et al.,* 1981. An excellent chromatographic separation of all the pesticides was obtained by using Reverse-Phase High Performance Liquid Chromatography (RP-HPLC). The controls were free of HPLC interference for the target compounds. HPLC analyses showed that o, p$^{/}$-DDT and p, p$^{/}$-DDTelute at a retention time of 7.956 and 7.557 min. respectively whereas o, p$^{/}$-DDE, o, p$^{/}$-DDD, p, p$^{/}$-DDE and p, p$^{/}$-DDD were 7.730, 6.975, 8.121, and 7.926 min. respectively. The limit of detection (LOD) for DDT analyses is in the range of 0.02-0.10 ng/mL. The peaks observed for the standards (Fig. 1) and water samples were sharp and minimal tailing.

Table. 1. Water sampling Locations

Sample No.	Location
1	Hand pump (90 feet) Lahore
2	Tube well water, Haroonabad (District Bahawalpur)
3	Water pump (3-40 feet) Haroonabad (District Bahawalpur)
4	Canal water, Haroonabad (District Bahawalpur)
5	Hand pump (30 feet) Sheikhupura (District Lahore)
6	Hand pump, Haroonabad (District Bahawalpur)
7	Tube well water, Mianchannu (District Khanewal)
8	Hand pump water, Mianchannu (District Khanewal)
9	Canal water, Mianchannu (District Khanewal)
10	Tap water, Hyderabad
11	NIAB, Canal water, Faisalabad
12	Hand pump water, Hyderabad
13	Tap water, Railway line, Faisalabad
14	Tube well water, Baidian (District Lahore)
15	Tube well water, Gujranwala
16	Railway station, Hyderabad
17	Hand pump, Hala, Sindh
18	River water, River Indus, Kotari
19	Tube well water, Narowal
20	Water pump, Chak Dena wala, Faisalabad
21	Hand pump, Ali town, Sargodha road Faisalabad

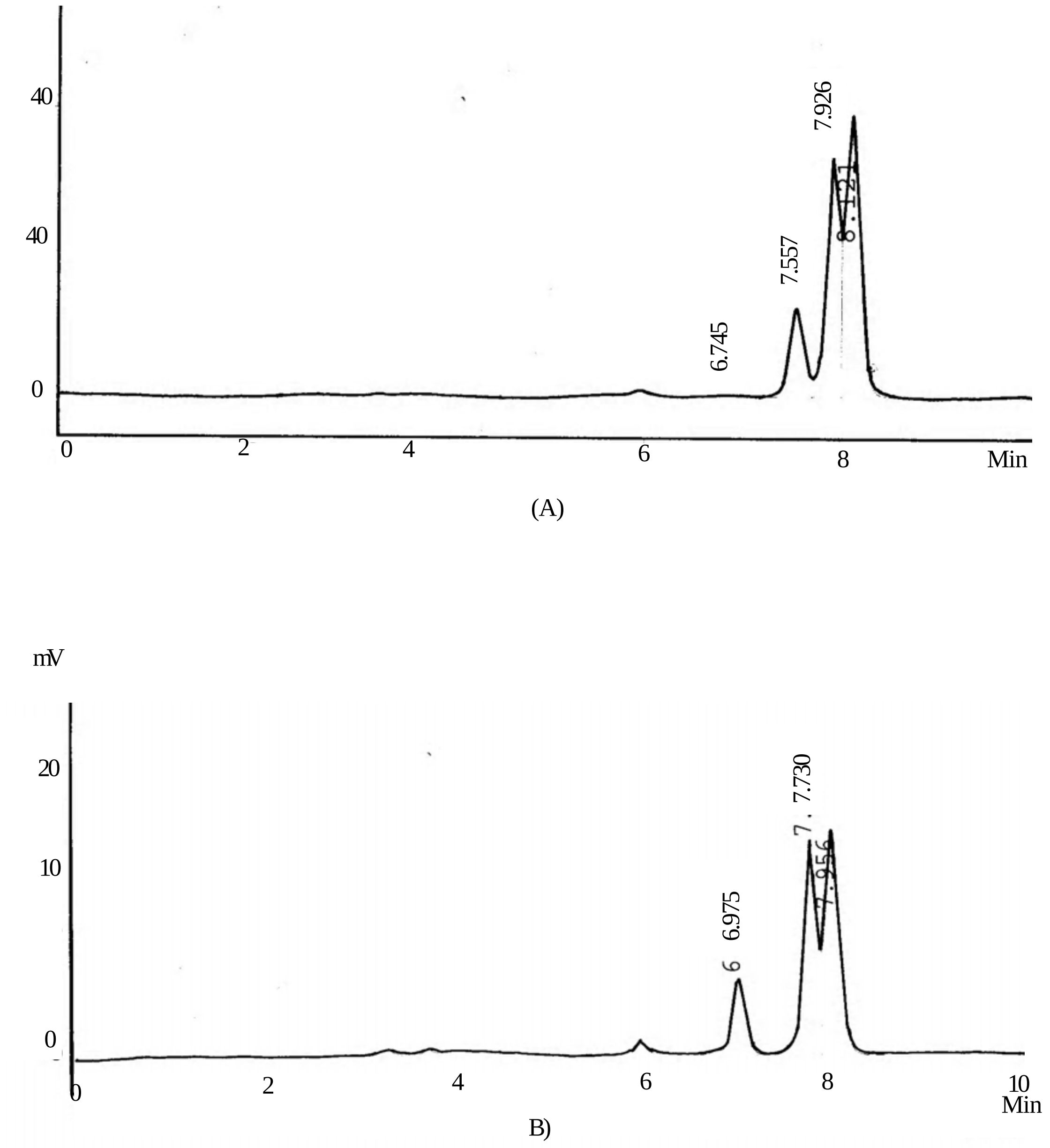
Table 2. Recovery of pesticides residues from water samples

Standard	Spiked concentration	Recoveries %
o, $p^{/}$-DDD	2 ppb	85±5
o, $p^{/}$-DDE	2 ppb	89±8
o, $p^{/}$-DDT	2 ppb	90±4
p, $p^{/}$-DDD	2 ppb	91±6
p, $p^{/}$-DDE	2 ppb	84±3
p, $p^{/}$-DDT	2 ppb	87±11

Identification was made by comparing the peaks of samples with that of standards for qualitative and quantitative analysis. With the help of peak areas and retention times of standards, the concentrations of samples were calculated (Table 3).

CONCLUSION

Ortho, para- DDD, o, $p^{/}$-DDE, o, $p^{/}$-DDT, p, $p^{/}$-DDE, p, $p^{/}$-DDT have been found in all the water samples except few locations where they were not present (Table 3). Para, para- DDD was not present in all the water samples collected from different places of the country. The concentration of o, p- DDD was highest (0.820 ng/mL) in hand pump, cotton belt area, Haroonabad. While lowest concentration (0.032 ng/mL) was found in hand pump, Ali town, Faisalabad. The higher concentration at Haroonabad may be due to the application of DDT in the past because in this area DDT was applied to control the pest against cotton or in malaria eradication program. All the other samples have the concentration in the range between 0.820 and 0.032 ng/mL. It was seen that Haroonabad canal water also contain higher (0.766 and 0.935 ng/mL) amount of o, $p^{/}$-DDE, and o, $p^{/}$-DDT, while lowest concentration (0.017 and 0.027 ng/mL) of these metabolites in hand pump, Sheikhopura, and NIAB canal water, Faisalabad. The lowest concentration is due to the non-use of DDT at these localities. The highest concentration (0.127 ng/mL) of p, $p^{/}$-DDE was found in tube water, Baidian, Lahore while its lower concentration was at NIAB canal water, Faisalabad. In Mianchannu, District Khanewal, highest concentration (1.06 ng/mL) of p, $p^{/}$-DDT was present i.e. 10 times more than the maximum admissible limits. Para, $p^{/}$-DDT was also present in nearly all localities except some where its concentration was lower i.e. below permissible limits. The concentration of detected pesticides, particularly in the cotton growing areas were certainly above the maximum admissible concentration of individual pesticide (0.1 ng/mL) set by European Community (David R Morgan, 1992). Generally, it was observed that DDT or its metabolites residues were under acceptable quantity in Municipal area like Lahore, Hyderabad while higher concentration were present in cotton growing belts particularly Bhawalpur, Khanewal etc. River Indus has higher concentration of DDT and its metabolites at the place of Kotari at the time of sampling.



(A)

B)

Fig. 1. Resolution of DDT and its metabolites.
a) p, p$^{/}$-DDT, 7.557; p, p$^{/}$-DDD, 7.926; p, p$^{/}$-DDE, 8.121*min
b) o, p$^{/}$-DDT, 7.956; o, p$^{/}$-DDD, 6.975, o, p$^{/}$-DDE, 7.730 min
*** Data shows the retention times**

Table 3. DDT and its metabolites residues (ng ml^{-1}) in water samples

Sample No.	o,p$^{/}$-DDD	o,p$^{/}$-DDE	o,p$^{/}$-DDT	p,p$^{/}$-DDD	p,p$^{/}$-DDE	p,p$^{/}$-DDT
1	NF	0.038	0.080	NF	0.089	0.101
2	NF	0.086	0.238	NF	0.096	0.947
3	0.201	0.278	0.521	NF	NF	NF
4	NF	0.726	0.935	NF	NF	NF
5	0.272	0.017	NF	NF	NF	0.040
6	0.820	NF	NF	NF	NF	NF
7	0.706	NF	0.715	NF	NF	0.147
8	NF	0.051	0.058	NF	NF	0.840
9	0.283	0.135	NF	NF	NF	1.060
10	0.330	0.022	0.029	NF	0.025	0.042
11	0.318	0.033	0.027	NF	0.022	NF
12	0.362	0.428	NF	NF	0.057	NF
13	NF	0.463	0.064	NF	0.058	0.156
14	0.197	0.054	NF	NF	0.127	0.085
15	0.272	0.026	NF	NF	0.092	0.046
16	0.096	0.021	0.031	NF	0.039	NF
17	0.243	0.033	0.048	NF	0.018	NF
18	0.172	0.437	NF	NF	0.123	0.186
19	NF	NF	NF	NF	0.181	NF
20	0.386	0.029	0.073	NF	NF	NF
21	0.032	NF	NF	NF	0.096	NF

The occurrence of potentially hazardous chemicals such as DDT and its degrading products in crop growing areas/ground water/ drinking water, even in low concentration, is of real concern because of its potential for long term persistence and wide spread exposure to public. At the present time, only the agriculture land is affected, however, this may simply be a function of time when the other matrices will also contaminated.

ACKNOWLEDGEMENT

Authors would like to thank to Dr. Charles S. Helling, Lead Scientist, USDA, Beltsville, MD, USA for gifting the Manifold SPE-10 and also grateful to IAEA for providing the reference standards.

REFERENCES

Billings, W. N. and Bidleman., T. F., (1983). High volume collection of chlorinated hydrocarbons in urban air using three solid adsorbents. Atmos. Environ. **17**, 383.

Crutchfield, S. R., Ribaudo, M. O., Hansen, L. T. and Quiroga, R., (1992). Cotton production and water quality: Economic and environmental effect of pollution prevention. USDA-ERS, Agriculture economic reports number 664. Herrdon, VA; USDA-ERS.

David, R., Morgan (Ed.), (1992). BMA guide to pesticide chemicals and health, published on behalf of the British Medical Association by Edward Amold. 121.

Dogheim, S. M., EL-Moattassem, M., Macklad, F., Shaker, N. and Hassan, S., (1992). International conference in the protection and development of the Nile and other major rivers, Cairo, Egypt.

Gaeb, S. Nitz, S., Parlor, H. and Korte, F., (1975). Photomineralisation of certain aromatic xenobiotica. Chemosphere **4**, 251.

Glotfelty, D.E., Selber J.N., and Lijedahl, L.A., (1987). Pesticides in fog. Nature, **325**, 602.

Hallberg, G. R., (1986). Overview of agricultural chemicals in groundwater, In Proc. Conf. Agr. Impact on Groundwater. Nat. Well Water Assoc., Dublin, Ohio. 1-63.

Hallberg, G. R., (1989). Agric. Ecosyst. Environ., **26**, 299-367.

Hinkley, D.A. and Bldle, T.F., (1989). Analysis of pesticedes in seawater after enrichment onto C8 bonded-phase cartridges. Environ. Sci. Technol., **23**, 995.

Hussain, A., Ali, S. W. and Asi, M. R., (1997). Low-level determination of pesticides in environmental water samples using solid phase extraction and high performance liquid chromatography. Proc. NSMTCC97 ON environmental pollution February 24-25,1997, Islamabad-Pakistan.

Kendel, W., Chquin, J. C., Perez, J. and Rosales, S., (1999). Determination of pesticides residues in surface and ground water used for human consumption in Guatemala. J. AOAC, **82**, 41-59.

Kumari, B., Singh, R., Madan, V. K., Kumar, R. and Kathpal, T. S., (1996). Monitoring of pesticide resdiues in fruits. Bull. Environ. Contam. Toxicol., **56**, 787.

Petty, E. E., Johnston, J. J. and Volz, S. A., (1997). Solid phase extraction methd for the qualitative analysis of organochlorine pesticides in wild life urine. J. Chromatogr. Sci., **35**, 430-434.

Peveen Z. and Maud S. Z., (1988). Organ chlorine pesticide residues in cattle drinking water. Pak. J. Sci. Ind. Res., **31**, 53-56.

Pionke, H. B., Glotfelty, D. E. (1989). Nature and extent of grounfwater contamination by pesticides in an agricultural watershed. Water Res., **23**, 1031.

Pionke, H. B., Glotfelty, D. E., Lucas, A. D. and Urban, J. B., (1988). Pesricide contamination of graoundwaters in the Mahantano Creek Watersged. J. Environ. Qual., **17**, 76.

Richard, R.P., Kramer, J.W., Baker, D.B. and Krieger, K.A., (1987). Pesticides in rainwater in northeastern United States. Nature, **327**, 129.

Tanabe, A., Mitobe, H., Kawata, K. and Sakai, M., (2000). New monitoring system for ninety pesticides and related compounds in river water by solid phase extraction with determination by gas chromatography/ mass spectrometry. J. AOAC., **83**, 61-71.

Tsipi, D., Triantafylluo, M. and Hiskia (1999). Determination of organochlorine pesticides residues in honey, applying solid phase extraction with RP-C_{18} column. Analyst, **124**, 473-475.

Younos, T. M. and Weighmann, D. L., (1988). Pesticides in groundwater data base. 1988 interim Rpt. U. S. Environmental Protection Agency, Washington D. C.