Bull. Environ. Contam. Toxicol. (1993) 50:247–252

Environmental Contamination and Toxicology

Gas Chromatographic Determination of Pesticides in Natural Waters of Greece

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Analysis of natural waters for pesticides is recognized as complex, since several hundred pesticides with different physical and chemical properties are widely used for agricultural purposes. Analysis is also considered difficult and prone to errors due to the very low concentrations of the compounds. Measurement at the 0.1-μg/L level, that is required by the E.C. Council Directive (1980) for water intended for human consumption, requires the sample extracts to be greatly concentrated. This in turn results in concentration of other substances that are present in the sample. Furthermore, positive identification requires confirmatory evidence from more than one chromatographic procedure.

In the present study, three different gas chromatographic procedures were used to monitor natural water samples for a wide variety of pesticides. Surface river and lake waters originating from various regions of Greece were analysed. To date, very little work has been done on monitoring pesticides in the natural waters of Greece (Albanis et al. 1986).

MATERIALS AND METHODS

Water samples were collected in 2.5-L glass bottles, between October 1991 and January 1992, from the following lakes : Iliki, Paralimni, Marathon, Mornos and Plastira; and the following rivers : Aheloos, Pinios, Gallikos, Enipeas, Kosinthos, Lailia, Drosopigi, Gortsilakas and Bos-Pos. These lakes and rivers provide Athens and other big cities in Greece with potable water. Samples were stored at 4°C prior to extraction, which was normally conducted within 24 hr of sampling.

The solvents used were "pesticide residue" grade. Stock solutions (1000 and 100 μg/mL) and working solutions (10, 1, 0.1 and 0.01 μg/mL) of lindane, aldrin, dieldrin, phorate, parathion ethyl and parathion methyl, as well as

solutions of mixtures of these pesticides were prepared in acetone according to the standard practices (Chau and Lee 1982).

The multiresidue procedure described by Ambrus et al. (1981) was followed for the extraction and cleanup of the samples with minor modifications. According to the method, a 1-L sample of water is cleaned by removal of any floating or insoluble material. The water is transferred into a 2-L separatory funnel and extracted, after the addition of 50 mL of saturated NaCl solution, with 100-, 50- and 50-mL portions of CH_2Cl_2. The extracts are filtered through 30 g of anhydrous Na_2SO_4 which is then rinsed with 20 mL of CH_2Cl_2. The volume of the filtrate is reduced using a vacuum rotary evaporator at 30°C to ca 2 mL. 10 mL of acetone are added, and the solution is evaporated to ca 2 mL. This step is repeated twice. The solution is carefully evaporated to dryness, and 2.5 mL of toluene are added. For the cleanup of the samples, a suspension of 1 g of activated charcoal and 4 g of silanized celite is placed in a 18 mm i.d. column, toluene is allowed to flow through the column; and the sample extract is then added. Pesticides are eluted with 150 mL of CH_2Cl_2. The eluate is evaporated to ca 2 mL and, after adding 2x10 mL of acetone, is evaporated to dryness. 2 mL of acetone are added, and the extract is transferred to a sealed vial for gas chromatographic analysis.

A Varian aerograph model 3700 gas chromatograph was used. Three detector systems were used a) a Ni-63 electron-capture detector (ECD-system) with a 2 m x 2 mm i.d. glass column containing 1:1 mixture of 10% OV-101 and 15% OV-210 on Chromosorb WHP (80-100 mesh) b) a nitrogen-phosphorus detector (NPD-system) with a 2 m x 2 mm i.d. glass column containing 10% OV-101 on Chromosorb WAW (80-100 mesh) c) a flame photometric detector (FPD-system), operating with a phosphorus bandpass filter, with a 2 m x 2 mm i.d. glass column containing 3% OV-17 on Chromosorb WHP (80-100 mesh). Operating temperatures were : injection port 220°C, column oven 200°C, detector 250°C (for ECD 300°C). Nitrogen carrier-gas flow rate was 60 mL/min. 1 µL of the sample extract was injected for each of the three chromatographic systems, and three chromatograms were obtained for each sample. Quantification was achieved by use of a computer integrator.

RESULTS AND DISCUSSION

The extraction method that was used is a well established and validated method with verified applicability to approximately 150 pesticides (Ambrus et al. 1981; Ambrus and Thier 1986). The cleanup requires little time and the

carbon-celite mixed adsorbent used has been found to be efficient on a wide variety of pesticides (Ambrus et al. 1981).

Identification of the unknown peaks found in the samples' chromatograms is made by comparison of the relative retention time (RRT) of the unknown peak to the RRTs given in various RRT data tables. A different data table is used for the ECD-system (U.S. Deptm. of Health 1982, table 333-A), the NPD-system (U.S. Deptm. of Health 1982, table 331-B) and the FPD-system (Prinsloo and DeBeer 1985). Six target pesticides were selected for evaluation of the method. Table 1 lists the RRTs of the target pesticides found for each detector system. As seen from the table, the RRTs found were almost identical with those expected from the data tables.

Table 1. Relative retention times of the target pesticides for the three detector systems. In parentheses are the expect values from the RRT data tables.

Pesticide	ECD-system	NPD-system	FPD-system
Phorate	0.46 (0.45)	0.37 (0.38)	0.33 (0.34)
Lindane	0.59 (0.58)		
Aldrin	1.00*		
Parathion methyl	1.41 (1.42)	0.70 (0.70)	0.78 (0.79)
Parathion ethyl	1.85 (1.88)	1.00*	1.00*
Dieldrin	2.19 (2.22)		

* Reference compound

For statistical evaluation of the overall method's efficiency, 1 L of HPLC-grade water was spiked with the target pesticides at various concentration levels. The concentrations of the pesticides in each spiked sample were chosen so that a similar peak height was obtained for each compound in the chromatograms. A typical chromatogram of spiked water sample is shown in Figure 1a. Compounds in a sample were quantified by comparing the detector response for the sample to that measured for the calibration standard within the linear range. The results of the recovery study are presented in Table 2. The average recoveries were between 84 and 106%, except for phorate with 44-60% and relative standard deviation ranged from 4 to 18%, values satisfactory for residue analysis (Greeve 1984).

The lowest validation level for lindane and aldrin was 0.01 µg/L in this study. Since the worst case standard deviation was about 0.0005 at this level and the Student

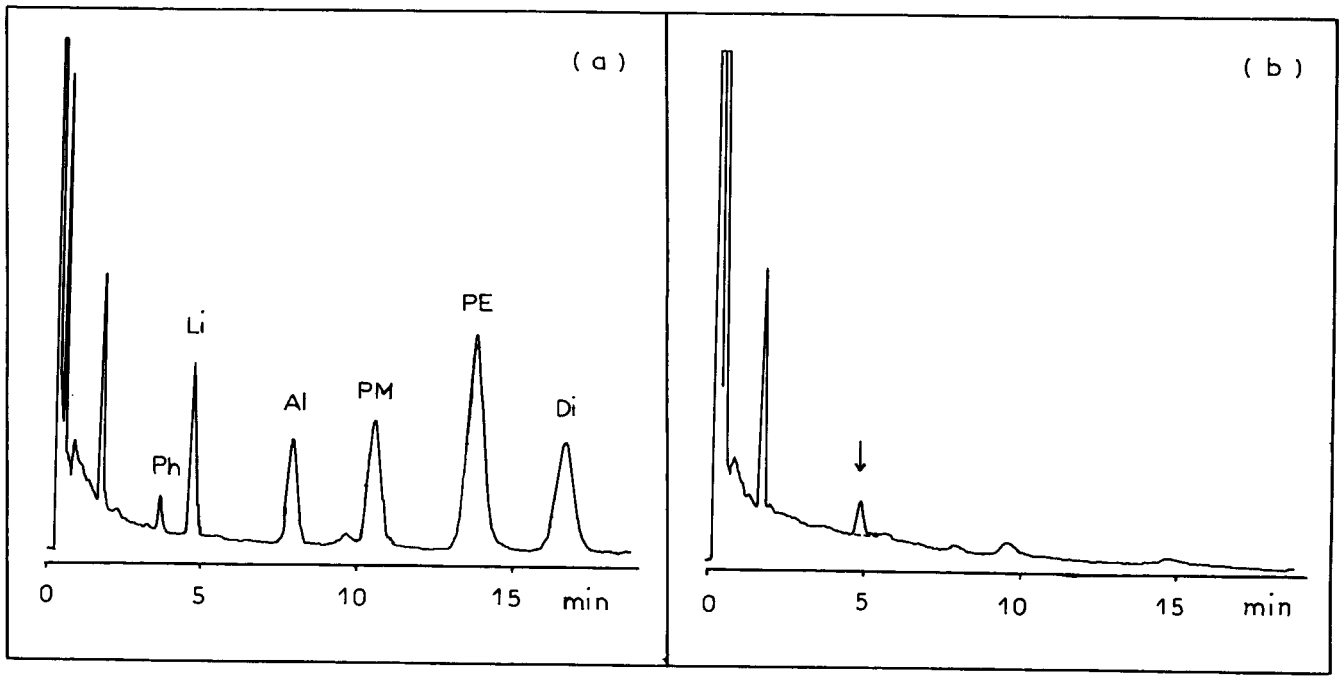

Figure 1. GC chromatograms with the ECD-system of a) High purity water spiked with 0.1 µg/L lindane (Li) and aldrin (Al), 0.2 µg/L dieldrin (Di), 1 µg/L phorate (Ph), 2 µg/L parathion methyl (PM) and parathion ethyl (PE). b) Iliki lake water (19 Nov. 91 sampling). Lindane is indicated with the arrow. The dotted line is the method blank.

Table 2. Mean recoveries (%) ± relative standard deviation (N=3) for the target pesticides in fortified water samples, at various fortification levels (μg/L). Li = lindane, Al = aldrin, Di = dieldrin, Ph = phorate, PM = parathion methyl, PE = parathion ethyl.

μg/L	Li	Al	Di	Ph	PM	PE
0.01	91±5	102±4				
0.02	85±5	95±11	90±9			
0.04			93±8			
0.05	88±7	89±6				
0.1	85±9	89±9	95±6	44±9		
0.2	84±7	84±13	94±10	54±18	86±9	101±8
0.4			88±11		95±12	106±8
0.5				50±9		
1				61±9	99±10	93±12
2				60±12	96±17	92±14
4					98±10	96±12

t-value at 99% confidence level is 6.96 for 2 degrees of freedom (3 replicates), a conservative estimate of the method detection limit (U.S., E.P.A. 1984) would be equal to the amount of SD x t-value or about 0.003 μg/L in the case of lindane and aldrin. The detection limit for dieldrin is about 0.01, for phorate 0.06 and for parathion methyl and parathion ethyl 0.1 μg/L.

Thirty water samples collected from the mentioned rivers and lakes of Greece were analyzed according to the method. A method blank (1 L of high purity water) was analyzed with each set of samples. This was found to be necessary since small peaks appeared in the chromatograms of the blanks. No pesticides were detected in any water samples, except those from Iliki lake. Lindane was detected in these samples at the following concentrations : 0.015 μg/L (1 Oct. 91 sampling), 0.01 μg/L (19 Nov. 91 sampling) and 0.005 μg/L (24 Jan. 92 sampling). Figure 1b shows a chromatogram using the ECD-system for Iliki lake water. Confirmation was achieved using a 30 m x 0.25 mm i.d. DB-5 fused-silica capillary column with the ECD-system. Lindane (gamma-hexachloro-cyclohexane) is an insecticide which is applied to the soil of potato fields lying adjacent to Iliki lake. However, concentrations of lindane found are much lower than the EEC maximum acceptable concentration of 0.1 μg/L.

Acknowledgments. The author thanks the Ministry of Environment that supported the project, Kathrerine Bourou for her assistance in processing samples, and Dr. E.J. Abramides for reviewing the manuscript.

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Received April 15, 1992; accepted August 20, 1992.