A Feasibility Study of Miniature Silica Gel Columns for the Separation of Some Polychlorinated Biphenyls, DDT, and Analogs

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Successful rapid cleanup of samples for the determination of certain pesticide residues using silica gel microcolumns has been reported by KADOUM (1967, 1968). LAW and GOERLITZ (1970) reported using silica gel in microcolumns for cleanup in analysis of water for pesticides.

The method of ARMOUR and BURKE (1970) for separating polychlorinated biphenyls (PCBs) from DDT and its analogs employs a 400 x 22 mm i.d. chromatographic column containing a mixture of 5 g Celite 545 and 20 g silicic acid. The purpose of this study was to investigate the feasibility of using miniature columns of silica gel (5 g in a 300 x 10 mm i.d. chromatographic column) for separating PCBs, DDT, and analogs with the solvent system used by ARMOUR and BURKE. PCBs consisting of Aroclors 1242, 1248, 1254, and 1260 and pesticide compounds consisting of p,p'-DDE, p,p'-TDE, and p,p'-DDT were individually tested for recovery from miniature columns. Mixtures consisting of each of the above PCBs individually combined with p,p'-DDE, p,p'-TDE, and p,p'-DDT were tested for separation and recovery on miniature columns. The use of miniature columns, if found acceptable for routine analysis, would reduce analytical time and reagent costs and would be ecologically advantageous in waste disposal.

Experimental

Reagents

(a) Silica Gel.--0.2-0.5 mm, 30-70 mesh (ASTM, EM Laboratories Inc., 500 Executive Blvd., New York, N.Y. 10523). Used for column chromatography as received.

(b) Solvents.--Petroleum ether, hexane, acetonitrile, and methylene chloride suitable for use in pesticide residue analysis.

(c) Eluting mixtures.--Petroleum ether and 1/19/80 (v/v) acetonitrile/hexane/methylene chloride mixture.

(d) Pesticide standards.--p,p'-DDE, p,p'-TDE, and p,p'-DDT supplied by the Environmental Protection Agency.

(e) Polychlorinated biphenyls.--Aroclors® 1242, 1248, 1254, and 1260 produced by Monsanto Co., St. Louis, Mo.

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Apparatus

(a) Chromatographic columns.--300 mm x 10 mm i.d. with 24/40 S outer joints at the top of the column and a coarse fritted disc and Teflon stopcock at the bottom (Kontes Glass Co., K-422450 or equivalent).

(b) Kuderna-Danish concentrators.--125 ml (Kontes Glass Co., K-570000) were used.

(c) GLC equipment.--A Packard Series 7800 Gas Chromatograph equipped with an electron capture detector and 6 ft. x 4 mm i.d. column containing 10% DC-200 on 80/100 mesh Chromosorb WHP was used. Operating conditions: nitrogen, 120 ml/min; column and detector temperatures, 200°C; injection temperature, 225°C. A detector sensitivity of 1/2 full scale deflection for 1 ng heptaclor epoxide was established.

Silica Gel Preparation

KADOUM (1969) reported satisfactory separation of pesticides from biological materials using high purity silica gel activated at 130°C or 300°C for 2 hr or longer periods. The silicic acid-Celite mixture prepared according to ARMOUR and BURKE (1970) gave slow elution rates and could not be used practically with miniature columns. Silica gel as received, silica gel that had been heated to 130°C for 16 hr, and silica gel prepared according to the procedure of ARMOUR and BURKE (1970) for silicic acid were tested for ability to separate PCB, p,p'-DDE, p,p'-TDE, and p,p'-DDT. These tests showed silica gel as received to give separation of tested compounds equal to or better than treated silica gel. A subsequent check showed that silica gel as received lost 3.4% weight on heating at 130°C for 16 hr. An absorbent column containing 5 g of silica gel used as received with the described elution system was chosen because the PCBs and p,p'-DDE eluted reproducibly in the 94/6 fraction which was the criterion of acceptance for the column.

Column Preparation and Elution Procedures

A slurry was prepared by weighing 5 g of silica gel in a 50 ml beaker, covering with petroleum ether, and stirring to remove air bubbles. Using a syringe containing petroleum ether, the silica gel was transferred to the chromatographic column with the stopcock wide open. A wash of 20 ml of petroleum ether was immediately added to the column and the flow rate was adjusted to 4-6 ml/min. After about 15 ml of wash eluted from the column, the column was tapped to settle the silica gel and was marked 1 cm above the settled silica gel. When the wash reached the 1 cm mark, the sample was added in 3 ml of petroleum ether followed with 1 ml of petroleum ether as a rinse. When the rinse reached the column mark, 35 ml of petroleum ether was added to the column and the eluate was collected in a Kuderna-Danish

concentrator. If necessary, the flow rate was readjusted to 4-6 ml/min. When the petroleum ether reached the mark on the column, 40 ml of the acetonitrile/hexane/methylene chloride mixture was added and the eluate was collected in a Kuderna-Danish concentrator. Collected fractions were concentrated to suitable volumes for GLC determination.

NOTE: Columns were prepared one at a time and given attention to assure that the eluting solvent did not go below the top of the silica gel in order to prevent cracking of the column.

Results and Discussion

Preliminary work reported in Table 1 shows the persent of tested compound eluted into varying volumes of petroleum ether. Based on these data, 35 ml was selected as the optimum volume for elution of PCB.

TABLE 1

Recoveries of Added PCB, DDT, and Analogs from Miniature Silica Gel Columns in Varying Volumes of Petroleum Ether (PE)[a]

		% Compound Eluted[b]			
Compound	µg Added	20 ml PE	25 ml PE	30 ml PE	35 ml PE
Aroclor 1242	10	61	71	85	100
Aroclor 1248	10	49	76	77	100
Aroclor 1254	20	25	88	100	100
Aroclor 1260	20	60	94	100	100
p,p'-DDE	2	13	65	88	95
p,p'-TDE	2	0	0	0	0
p,p'-DDT	2	0	0	0	10

[a]For total petroleum ether used add 4 ml to above volumes.

[b]Recoveries are averages of duplicate determinations.

Table 2 shows range of recoveries, average recoveries, and elution fraction for individually tested compounds. Recoveries ranged from 87 to 99% of added PCB and 90 to 99% of added p,p'-DDE in the petroleum ether fraction and 90 to 100% of added p,p'-TDE and 75 to 87% of added p,p'-DDT in the acetonitrile/hexane/methylene chloride fraction. Occasionally small amounts of p,p'-DDE were found in the acetonitrile/hexane/methylene chloride fraction and small amounts of p,p'-DDT were found in the petroleum ether fraction.

TABLE 2

Recoveries of PCB, DDT, and Analogs Individually Added to Miniature Silica Gel Columns[a]

Compound	Added, µg	Eluate a		Eluate b	
		Range, %	Av., %	Range, %	Av., %
Aroclor 1242	10	87-97	91	0	-
Aroclor 1248	10	88-98	90	0	-
Aroclor 1254	20	92-98	98	0	-
Aroclor 1260	20	96-99	99	0	-
p,p'-DDE	2	90-99	95	0-5	-
p,p'-TDE	2	0	-	91-100	96
p,p'-DDT	2	5-15	10	75-87	80

[a]Eluate a: A total of 39 ml of petroleum ether. Eluate b: 40 ml of 1/19/80 (v/v) acetonitrile/hexane/methylene chloride mixture. Range and average for each compound based on 6 determinations.

Table 3 shows range of recoveries, average recovery, and elution fraction for mixtures of individual PCBs combined with p,p'-DDE, p,p'-TDE, and p,p'-DDT. Recoveries ranged from 86 to 98% of added PCB and 80 to 93% of added p,p'-DDE in the petroleum ether fraction and 93 to 100% of added p,p'-TDE and 69 to 99% of added p,p'-DDT in the acetonitrile/hexane/methylene chloride fraction. Up to 15% of added p,p'-DDT was found in the petroleum ether fraction and small amounts of added p,p'-DDE were found in the acetonitrile/hexane/methylene chloride fraction.

Typical chromatograms obtained before and after separation on miniature silica gel columns are presented in Figures 1-4. This study shows that miniature silica gel columns used with the described elution system are capable of separating total PCB and p,p'-DDE from p,p'-TDE and p,p'-DDT. p,p'-DDE can be quantitated in the presence of PCBs provided the concentrations of latter are not excessive. Background interference of Aroclor 1254 must be taken into account if the PCB concentration is more than twice that of p,p'-DDE.

The miniature silica gel columns are easily prepared, rapid to use, and require small volumes of elution solvents. The author is investigating the use of miniature silica gel columns for the determination of pesticide and PCB compounds in various food commodities.

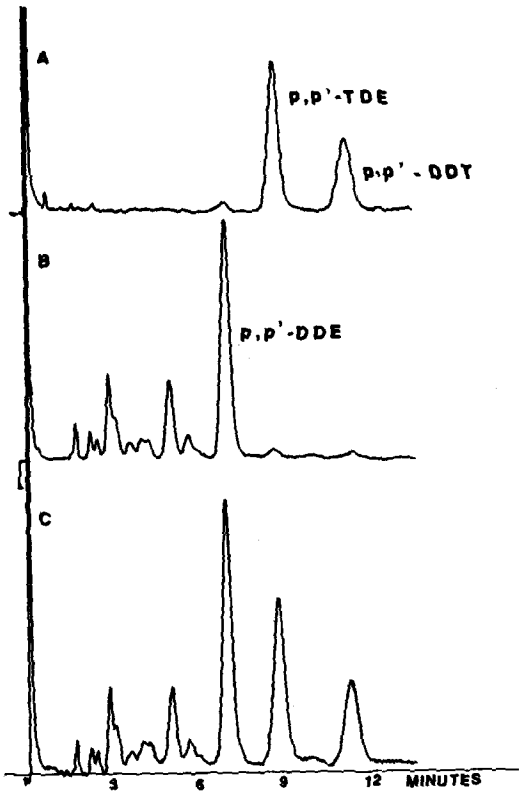

FIG. 1 - GLC chromatograms of Aroclor 1242 and p,p'-DDE, p,p'-TDE, and p,p'-DDT before and after separation on miniature silica gel column: A, eluate of acetonitrile/ hexane/methylene chloride; B, eluate of petroleum ether; C, before column separation. Injected amount equivalent to 20 µg Aroclor 1242 and 2 µg each of p,p'-DDE, p,p'-TDE, and p,p'-DDT.

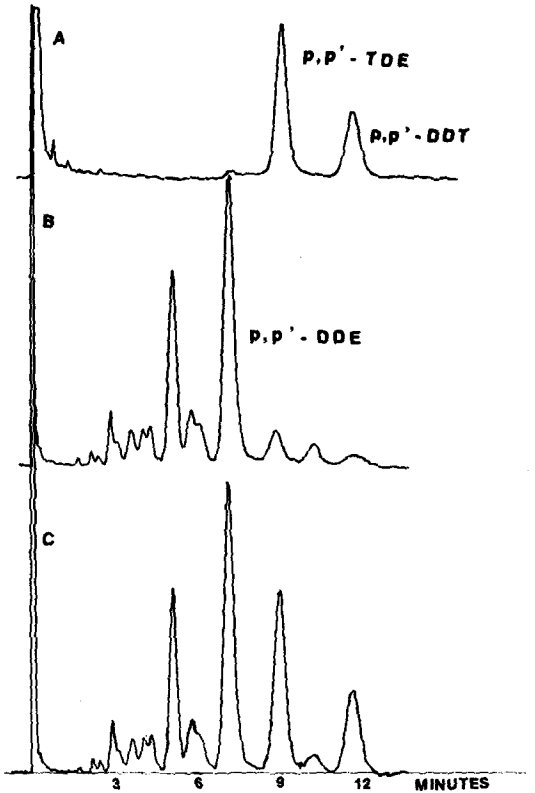

FIG. 2 - GLC chromatograms of Aroclor 1248 and p,p'-DDE, p,p'-TDE, and p,p'-DDT before and after separation on miniature silica gel column: A, eluate of acetonitrile/ hexane/methylene chloride; B, eluate of petroleum ether; C, before column separation. Injected amount equivalent to 20 µg Aroclor 1248 and 2 µg each of p,p'-DDE, p,p'-TDE, and p,p'-DDT.

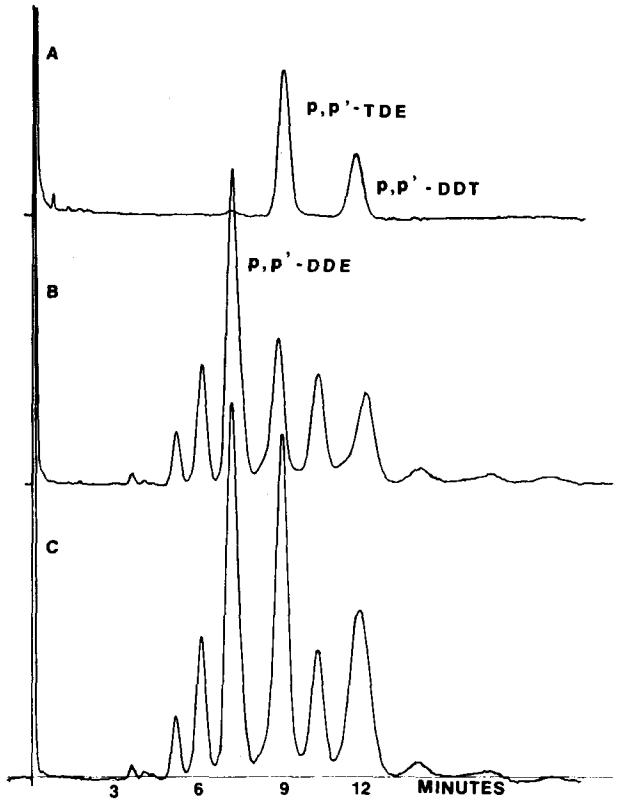

FIG. 3 - GLC chromatograms of Aroclor 1254 and p,p'-DDE, p,p'-TDE, and p,p'-DDT before and after separation on miniature silica gel column: A, eluate of acetonitrile/ hexane/methylene chloride; B, eluate of petroleum ether; C, before column separation. Injected amount equivalent to 20 µg Aroclor 1254 and 2 µg each of p,p'-DDE, p,p'-TDE and p,p'-DDT.

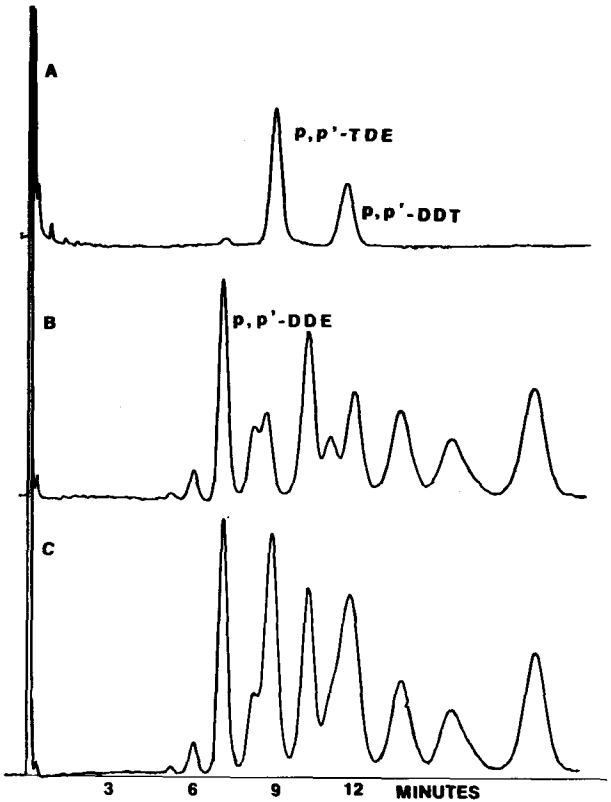

FIG. 4 - GLC chromatograms of Aroclor 1260 and p,p'-DDE, p,p'-TDE, and p,p'-DDT before and after separation on miniature silica gel column: A, eluate of acetonitrile/ hexane/methylene chloride; B, eluate of petroleum ether; C, before column separation. Injected amount equivalent to 20 µg Aroclor 1260 and 2 µg each of p,p'-DDE, p,p'-TDE and p,p'-DDT.

TABLE 3

Recoveries of PCB, DDT, and Analogs in Mixtures Added to Miniature Silica Gel Columns[a]

Compound	Added, µg	Eluate a		Eluate b	
		Range, %	Av., %	Range, %	Av., %
Aroclor 1242	20	87-97	91	0-5	-[b]
p,p'-DDE	2	80-89	85	0	-[b]
p,p'-TDE	2	0	-	93-100	95
p,p'-DDT	2	0-10	-	75-84	80
Aroclor 1248	20	87-97	93	0	-[c]
p,p'-DDE	2	88-93	91	0-5	-[c]
p,p'-TDE	2	0	-	95-100	98
p,p'-DDT	2	0-10	-	87-99	93
Aroclor 1254	20	86-93	90	0	-[d]
p,p'-DDE	2	80-87	85	0-7	-[d]
p,p'-TDE	2	0	-	94-99	98
p,p'-DDT	2	0-15	-	87-94	91
Aroclor 1260	20	88-98	93	0	-[e]
p,p'-DDE	2	85-93	88	0-5	-[e]
p,p'-TDE	2	0	-	95-100	98
p,p'-DDT	2	0-15	-	69-77	75

[a] Eluate a: A total of 39 ml of petroleum ether. Eluate b: 40 ml of 1/19/80 (v/v) acetonitrile/hexane/methylene chloride mixture. Range and average for each compound based on 6 determinations.
[b] See Fig. 1. [c] See Fig. 2. [d] See Fig. 3. [e] See Fig. 4.

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