CHROM. 4608

Electron-capture gas chromatography of free chlorophenols

A need in this laboratory for a rapid and sensitive method for the determination of chlorophenols in fats, oils, and fatty acids, because of their relationship to chick edema factor[1], prompted an investigation of the use of electron-capture gas chromatography for the determinative step.

Tailing has been a major problem in analyzing free chlorophenols by gas–liquid chromatography. HARVEY AND NORMAN[2] concluded that mixtures of free chlorophenols could not be analyzed and suggested their conversion by diazomethane to methyl ethers. ARGAUER[3] has published a rapid procedure for the chloro-acetylation of microgram quantities of chlorophenols and detection by electron-capture gas chromatography. However, a procedure that would allow a direct analysis of chlorophenols would still be advantageous.

METCALFE[4] used polar polyester-type columns with added phosphoric acid to eliminate heavy tailing in the analysis of free fatty acids. KOLLOFF *et al.*[5] succeeded in analyzing chlorophenol mixtures without sample pretreatment by using thermal conductivity and flame ionization detection. These workers resolved free chlorophenols on a column containing phosphoric acid and Carbowax 20M, a thermally stable polar substrate capable of forming strong hydrogen bonds at elevated temperatures. Addition of phosphoric acid to the solid support deactivated adsorption sites and eliminated peak tailing[6]. BARTHEL *et al.*[7] used a phosphoric acid column for the electron-capture gas chromatography of pentachlorophenol in clothing and biological materials

The purpose of this paper is to demonstrate the use of a variety of polar column substrates containing phosphoric acid for the electron-capture gas chromatographic analysis of free chlorophenols.

Experimental

Apparatus. A Barber-Colman Pesticide Detector Analyzer (Model 5360) gas chromatograph and a Series 8000 Chromocorder were used. A concentric-type Packard electron-capture detector with tritium as a radioactive source (90 mCi) was used as the detecting device. Nitrogen was the carrier gas.

Chemicals. All reagents and chemicals used in this work are commercially available. The chlorophenols studied were orthochlorophenol (OCP), 2,4-dichlorophenol (2,4-DCP), 2,5-dichlorophenol (2,5-DCP), 2,6-dichlorophenol (2,6-DCP), 2,4,6-trichlorophenol (2,4,6-TCP), 2,4,5-trichlorophenol (2,4,5-TCP), 2,3,4,6-tetrachlorophenol (2,3,4,6-TCP), and pentachlorophenol (PCP).

Chromatographic columns. The five liquid substrates investigated were Carbowax 20M, diethylene glycol succinate (DEGS), ethylene glycol succinate (EGS), ethylene glycol adipate (EGA), and ethylene glycol malonate (EGM). The columns were glass coiled, 9 ft. × 4 mm I.D., packed with one of the five liquid substrates and 2% H_3PO_4 on Gas-Chrom P, 60–80 mesh. The packings were prepared according to the method of METCALFE[4]. The packed columns were purged with nitrogen at room temperature and then given a static (no gas flow) 18-h conditioning at the appropriate conditioning temperature (see Table I). After the conditioning, the temperature was

TABLE I

COLUMN OPERATING PARAMETERS

Column	*Conditioning temperature (°C)*	*Operating temperature (°C)*	*Gas flow (ml/min)*
15% Carbowax 20M + 2% H_3PO_4	200	170	150
15% EGA + 2% H_3PO_4	195	165	120
15% DEGS + 2% H_3PO_4	185	155	130
10% EGM + 2% H_3PO_4	185	155	120
15% EGS + 2% H_3PO_4	195	165	120

TABLE II

RETENTION TIMES RELATIVE TO OCP ON POLAR SUBSTRATES

Compound	*Carbowax 20M*	*EGA*	*DEGS*	*EGM*	*EGS*
OCP	1.00	1.00	1.00	1.00	1.00
2,6-DCP	2.42	2.77	2.28	1.64	2.65
2,4-DCP	3.14	2.77	3.17	2.64	2.65
2,5-DCP	3.14	2.77	3.17	2.64	2.65
2,4,6-TCP	5.00	5.33	4.61	3.45	4.78
2,4,5-TCP	9.57	7.54	10.2	9.00	7.22
2,3,4,6-TCP	14.2	14.0	14.0	10.5	12.8
PCP	35.9	33.1	37.1	28.5	30.2

lowered 30° and the columns were again purged with nitrogen until a steady baseline was obtained.

GLC operating conditions and sample analysis. Detector voltage, 40 V; power setting, 100; sensitivity, 1×10^{-9} A.F.S.; injection block temperature, 250°; detector block temperature, 215°. Table I indicates the column conditioning temperature, the column operating temperature, and the gas flow used with each column.

Standard solutions of the chlorophenols, both alone and as mixtures, were prepared by weighing the compounds and dissolving them in benzene, with subsequent serial dilutions. The final concentration of the chlorophenol mixture was approximately 30 ng/μl. One microliter injections were made with a 10-μl Hamilton syringe.

Results and discussion

In the present work, high-loaded polar columns were tested for the direct chromatography of chlorophenol mixtures. Table II shows the retention times of the compounds on the various columns.

A problem of peak identification arose during the course of this study. Two peaks appeared when an OCP sample was chromatographed on these columns, using electron-capture detection; however, only one peak was observed using flame ionization detection. The first peak was trapped (preparative gas chromatography) and was shown to be OCP by IR spectrophotometry. When rechromatographed using electron-capture detection, the identified OCP produced one peak which was used

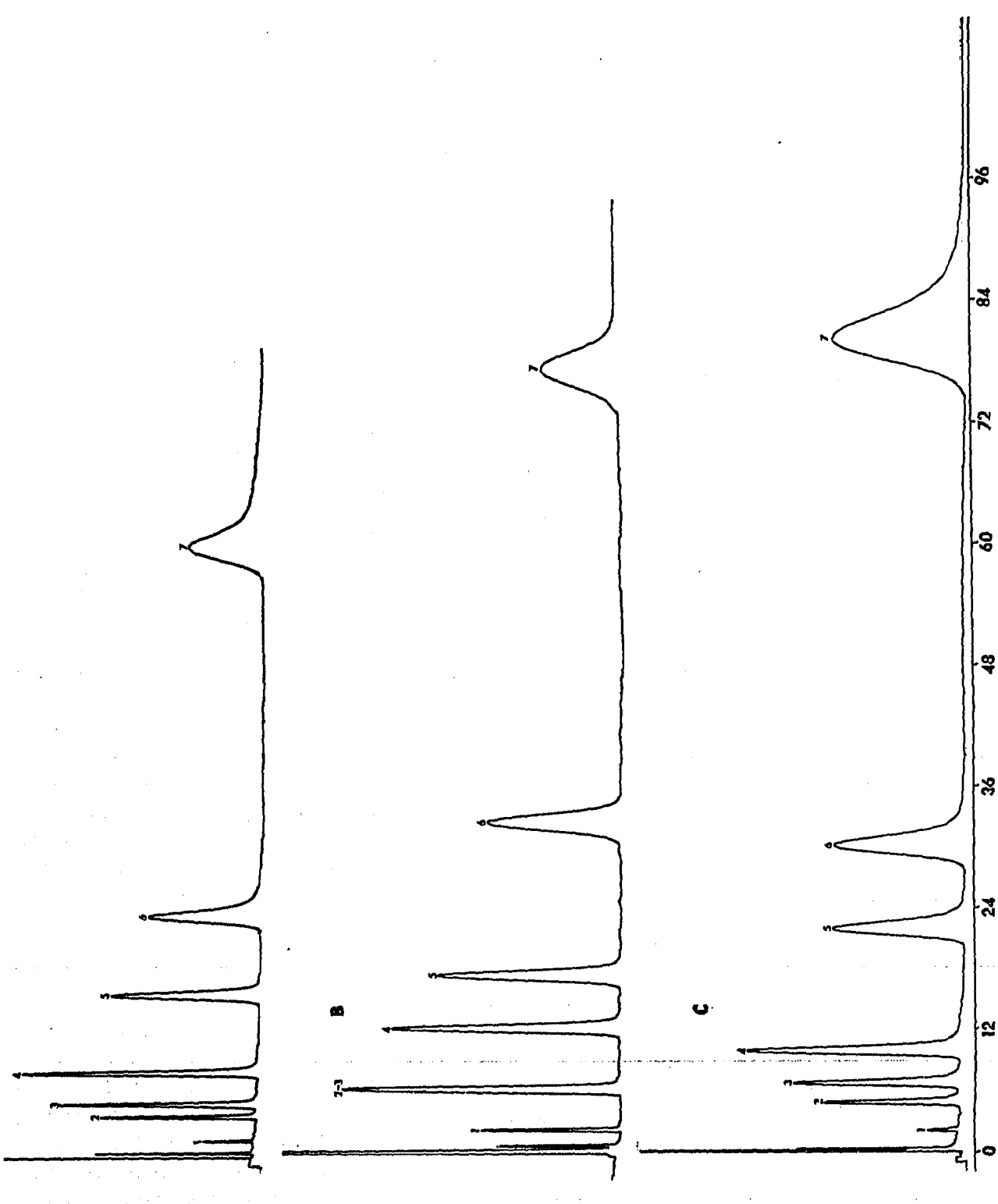

Fig. 1. Chromatograms of chlorophenol mixtures on 9 ft. × 4 mm I.D. glass coiled columns packed with liquid substrate and 2% H_3PO_4 on Gas-Chrom P, 60–80 mesh. Key: (A) 15% Carbowax, (B) 15% ethylene glycol adipate, (C) 15% diethylene glycol succinate. Compounds: (1) OCP, (2) 2,6-DCP, (3) 2,4- and 2,5-DCP, (4) 2,4,6-TCP, (5) 2,4,5-TCP, (6) 2,3,4,6-TCP, and (7) PCP.

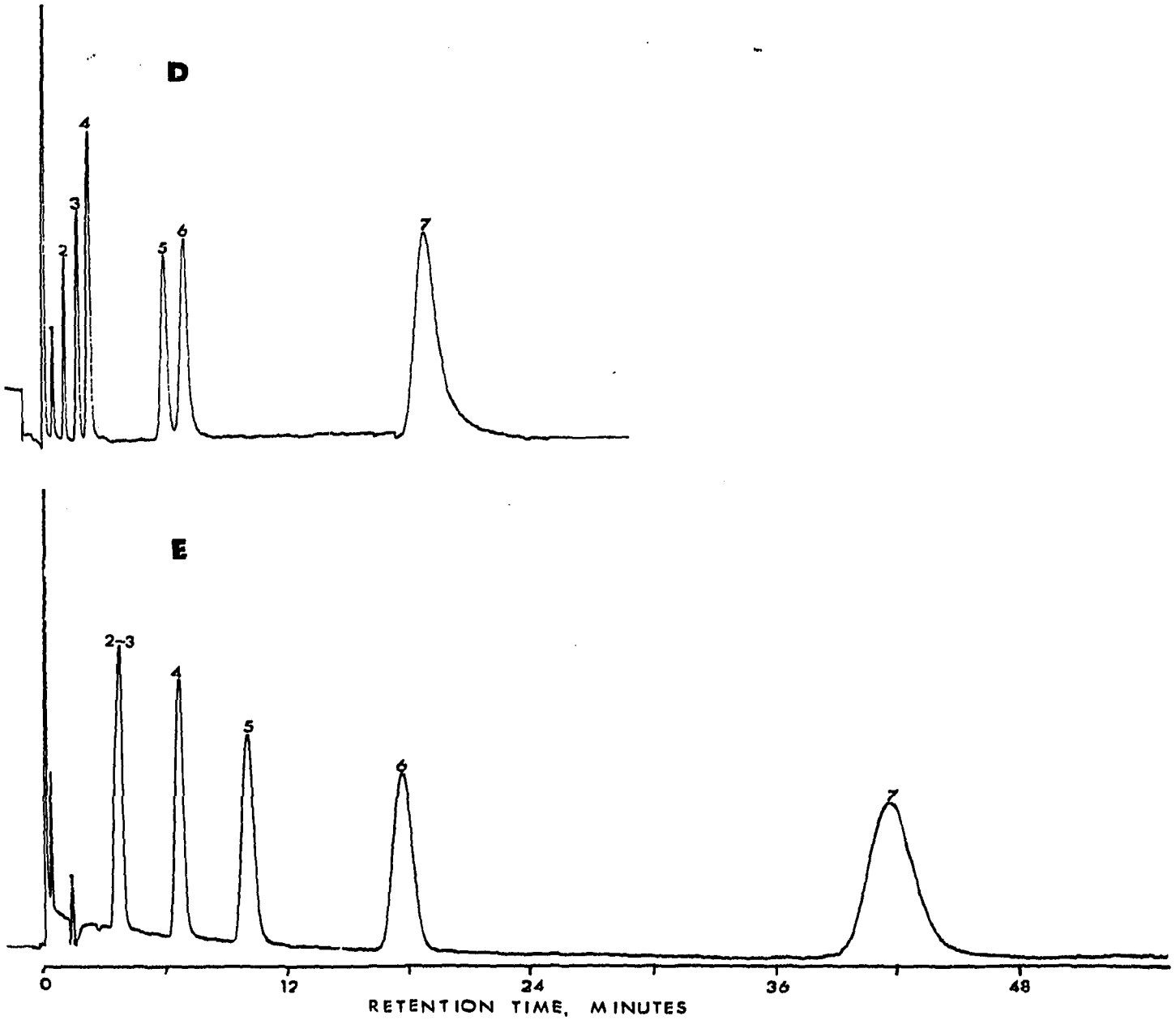

Fig. 2. Chromatograms of chlorophenol mixtures on 9 ft. × 4 mm I.D. glass coiled columns packed with liquid substrate and 2% H_3PO_4 on Gas-Chrom P, 60–80 mesh. Key: (D) 15% ethylene glycol malonate, (E) 15% ethylene glycol succinate. See Fig. 1 for identification of compounds.

to identify the OCP peak in the impure chlorophenol sample. The second peak observed in the sample was considered an electron-capturing impurity.

Chromatograms of the chlorophenol mixture on these columns are shown in Figs. 1 and 2. The criteria used in judging the overall performance of each column were resolution, response, and symmetry of the peaks. 2,4-DCP was resolved from 2,6-DCP on columns A, C, and D, but the two compounds eluted at the same time on columns B and E. Tailing of PCP was more evident on columns A and D but

was eliminated on columns B, C, and E. The greatest response for OCP was achieved on column B; however, absorption of PCP is quite evident. Column D gave the best total response for the chlorophenol mixture; although the column life was the shortest of the five tested.

Columns A, B, C, and E were in operation for four weeks and exhibited no sign of breakdown. The ghosting effect[8] was not noticed on any of the five columns.

No undue difficulties were encountered from H_3PO_4 column bleed-off with a tritium detector; however, the Teflon exit line from the detector did become clogged on several occasions. This problem was easily overcome by frequently changing the Teflon line. The best linearity of response was obtained with columns C and D, although a decrease in operating temperature for columns A, B, and E also resulted in good linearity. The best overall performance was obtained from column C.

By decreasing the temperature of the Carbowax 20M column by 30°, the less common 2,5-DCP isomer could be partially resolved from 2,4-DCP, but the separation was not complete enough for quantitative identification. KOLLOFF *et al.*[5] achieved the same results with a 20% Carbowax column and hydrogen flame detection.

These results show that the addition of 2% phosphoric acid to each of the five polar substrates reduced tailing of the chlorophenols; direct chromatography of these compounds was then possible.

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Received December 23rd, 1969

Editor's note. See also E. PILLION, *J. Gas Chromatog.*, 3 (1965) 238 and I. GERMAN AND M. CIOT, *Rev. Chim.*, 17 (1966) 177.