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THE SEPARATION AND DETERMINATION OF LOW-MOLECULAR-WEIGHT (C_2–C_{10}) STRAIGHT-CHAIN CARBOXYLIC ACIDS IN DILUTE AQUEOUS SOLUTION BY THE GAS CHROMATOGRAPHY OF THEIR *p*-BROMOPHENACYL AND *p*-PHENYLPHENACYL ESTERS

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SUMMARY

The quantitative and qualitative analysis of acetic, propionic, *n*-butyric, *n*-valeric, *n*-hexoic, *n*-heptoic, *n*-octoic, *n*-nonoic and *n*-decanoic acids present in dilute aqueous media as free acids or salts, in the presence of mineral acids or bases, is described. The *p*-bromophenacyl or *p*-phenylphenacyl esters of the acids were formed and injected into 2.5 % sodium dodecylbenzene sulphonate or 1 % Apiezon L or a 1:1 mixture of the two (depending on which acids were present) coated on acid-washed 60/80 mesh Chromosorb G packed in a 6-ft. glass column.

INTRODUCTION

Low-molecular-weight carboxylic acids are often encountered in electro-organic synthesis. They are formed as the main or byproducts in aqueous media in the presence of supporting electrolytes which may be mineral acids and salts or organic bases and salts. Most of the conventional methods of esterification and silylation of this class of carboxylic acids for gas chromatography (GC) utilise nonaqueous conditions. The recovery of the acids in aqueous mixtures by solvent extraction or distillation for direct injection or derivative formation was not only irreproducible but unsuitable for some of the aqueous situations of interest. It was therefore found necessary to investigate the various derivatives of carboxylic acids which could be quantitatively made in aqueous conditions.

The author has reported a method for the determination of formic acid or its salts in dilute aqueous solution by the gas chromatography of its anilide or toluidide derivatives[1]. Even though the anilide derivatives of C_2–C_8 straight-chain carboxylic acids were also found suitable for GC analysis, they could not be quantitatively prepared in aqueous conditions[1]. Malic, glycolic and oxalic acids in aqueous mixtures have been analysed by GC of their methyl esters. The alkaline salt solution of the acids was acidified with conc. hydrochloric acid and then taken nearly to dryness under reduced pressure at 35–40°. The resulting wet solid was treated with large excess of diazomethane in ether, and the ether extract injected into the instrument[2]. This

method was found tedious and in any case did not apply to the more volatile acids like acetic and propionic.

p-Bromophenacyl and *p*-phenylphenacyl esters of C_2–C_{10} acids were easy to prepare in aqueous conditions. Some stationary phases and inert supports were tried for the quantitative separation of these derivatives. Apiezon L and sodium dodecylbenzene sulphonate coated on acid-washed 60/80 mesh Chromosorb G were found suitable under various oven temperatures and carrier gas inlet pressures for the analysis of the C_2–C_{10} carboxylic acids.

EXPERIMENTAL

A Perkin-Elmer Model F11 gas chromatograph equipped with dual flame ionisation detector was used for the analysis and nitrogen served as the carrier gas.

Preparation of the columns

(i) 2.5 % sodium dodecylbenzene sulphonate on AW Chromosorb G was prepared and packed as described earlier[1].

(ii) 0.5 g of Apiezon L was dissolved in benzene–methylene chloride (1 : 1) solvent mixture. 49.5 g of AW 60/80 mesh Chromosorb G were added and the mixture was made into a slurry. The solvent was removed with stirring at 50° until the solid appeared dry. It was then cooled and packed by suction into a 6 ft. × 3 mm I.D. glass column.

(iii) 10 g of the preparation in (i) and 10 g of (ii) were mixed thoroughly by stirring and then packed into a 6 ft. × 3 mm I.D. glass column. These columns were conditioned at 230° for 18 h while nitrogen flowed through at an inlet pressure of 8 lb./in.^{2}.

Chemicals

The chemicals were used as obtained from B.D.H. The derivatives were prepared as described by VOGEL[3] for retention time determinations.

TABLE I

INSTRUMENTAL CONDITIONS FOR ANALYTICAL SEPARATIONS

The oxygen and hydrogen inlet pressures and the injection port temperature were the same for all the separations, *viz.* 26/18 lb./in.^{2} and 250°, respectively.

Acids	*Derivative*	*Oven temperature (°C)/Nitrogen inlet pressure (lb./in.^{2})*			*Figs.*
		Column A[a]	*Column B*[b]	*Column C*[c]	
C_2–C_5	(a) *p*-bromophenacyl	—	195/10	195/10	5, 9
	(b) *p*-phenylphenacyl	—	220/15	220/15	4, 7
C_6–C_{10}	(a) *p*-bromophenacyl	220/15	220/15	—	—
	(b) *p*-phenylphenacyl	—	—	—	—
C_2–C_{10}	(a) *p*-bromophenacyl	210/20	210/15	210/15	2, 3, 6
	(b) *p*-phenylphenacyl (C_2–C_8)	—	—	230/20	8

[a] Column A = 2.5% (w/w) sodium dodecylbenzene sulphonate on acid-washed Chromosorb G.
[b] Column B = 2.5% DBS on AW Chromosorb G–1 % Apiezon L on AW Chromosorb G (1 : 1).
[c] Column C = 1% (w/w) Apiezon L on AW Chromosorb G.

Preparation of derivatives for chromatography

C_2–C_5. 5 ml of an ethanolic solution of acetic, propionic, *n*-butyric and *n*-valeric acids (0.1 *M* in each acid) were taken in a 50-ml stoppered flask, followed by 5 ml of distilled water and one drop of 0.5 % solution of phenolphthalein. The solution was neutralised with a few drops of 5 *N* potassium hydroxide solution, and then made just acid with 3 drops of 1 *N* hydrochloric acid. 0.56 g of *p*-bromophenacyl bromide or *p*-phenylphenacyl bromide (depending on which ester was being prepared) were weighed into the flask. The mixture was boiled under reflux for 10 min. Any solid reagent remaining soon after the mixture started boiling was dissolved with a few milliliters of ethanol. The solution was then cooled under running tap water and treated with 20 ml of ethyl acetate. After all the brown solid at the bottom of the flask was dissolved, cold distilled water was run in until the ethyl acetate layer separated clearly on top of the aqueous phase. A convenient volume of the organic layer was transferred with a dropping pipette into a dry stoppered tube containing a few grams of anhydrous sodium sulphate. The clear solution was then injected into the instrument.

C_6–C_{10}. 5 ml of ethanolic solution of *n*-hexoic, *n*-heptoic, *n*-octoic, *n*-nonoic and *n*-decanoic acids (0.1 *M* in each acid) were treated exactly as in (C_2–C_5) except that 0.70 g of the reagent were used and 25 ml of ethyl acetate were employed for extraction.

C_2–C_{10}. 2.5 ml of the ethanolic solution of the nine acids (0.1 *M* in each acid) were treated as above except that 0.63 g of the reagent were used and 25 ml of ethyl acetate were employed for extraction.

RESULTS AND DISCUSSION

The C_2–C_{10} carboxylic acids present in dilute aqueous solution as free acids or salts and in the presence of mineral acids may be quantitatively determined by the methods described. The formation of any of the derivatives is quantitative; under identical conditions the amount of each derivative formed is directly proportional to the amount of acid present (Fig. 1).

$$p\text{-}C_6H_5\cdot C_6H_4\cdot CO\cdot CH_2Br + RCOOK = \underset{\text{phenylphenacyl ester}}{p\text{-}C_6H_5\cdot C_6H_4\cdot CO\cdot CH_2\cdot OOCR} + KBr$$

$$p\text{-}Br\cdot C_6H_4\cdot CO\cdot CH_2Br + RCOOK = \underset{\text{bromophenacyl ester}}{p\text{-}Br\cdot C_6H_4\cdot CO\cdot CH_2\cdot OOCR} + KBr$$

The volume of ethyl acetate employed for the extraction of either of the derivatives formed in the water–ethanol medium as described is critical. To effect a quantitative analytical recovery 1 mmole total acids present in any aliquot of solution taken for derivative formation requires a minimum of 10 ml of ethyl acetate.

The choice of which esters to form and which column to use for qualitative or quantitative analysis depends mainly on which acids are present in mixture because the *p*-bromophenacyl and *p*-phenylphenacyl esters of these acids behave differently on the various columns. This choice is especially important when the C_1–C_3 acids are present.

The *p*-bromophenacyl esters of formic, acetic and propionic acids are not quantitatively resolved on any of the three columns. On column A acetic and propionic

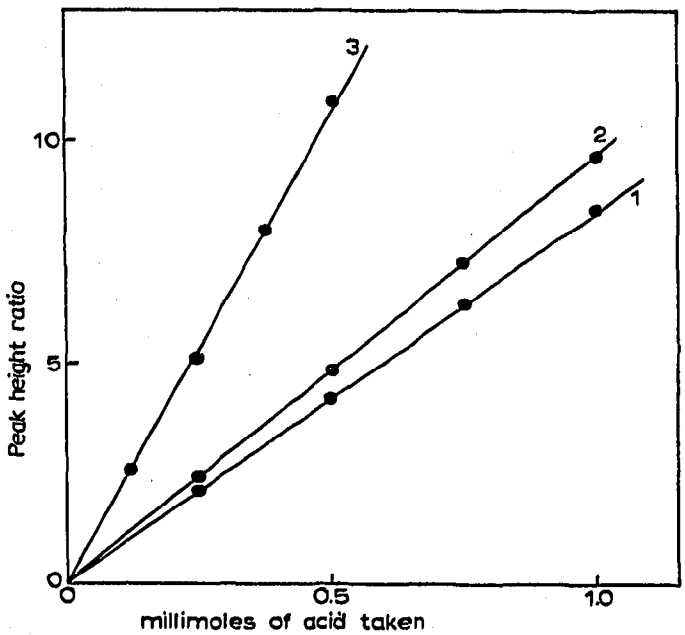

Fig. 1. Calibration curve for the formation of *p*-bromophenacyl esters of (1) acetic, (2) propionic and (3) *n*-butyric acids using *n*-valeric acid ester as the internal standard.

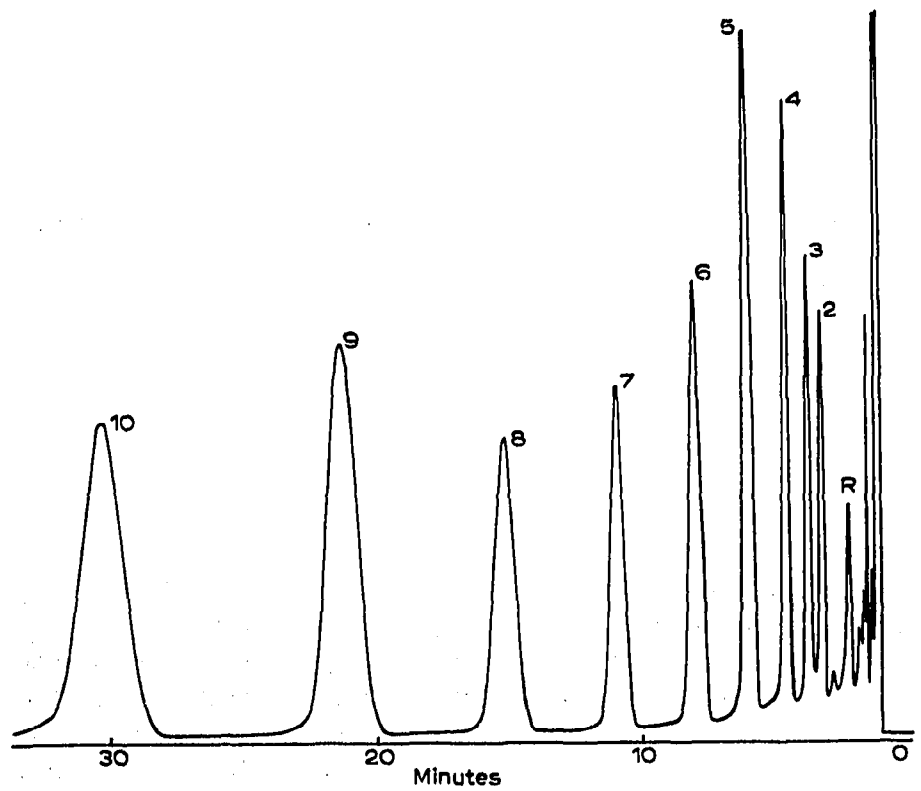

Fig. 2. C_2–C_{10} acids: *p*-bromophenacyl esters on 2.5% sodium dodecylbenzene sulphonate (column A). R = reagent; 2 = acetic; 3 = propionic; 4 = *n*-butyric; 5 = *n*-valeric; 6 = *n*-hexoic; 7 = *n*-heptoic; 8 = *n*-octoic; 9 = *n*-nonoic and 10 = n-decanoic.

acids are not resolved (Fig. 2); formic acid peak hardly appears but when it is in very large excess in the mixture it causes a distortion of the acetic and propionic acid peaks. On column B the acetic and propionic acid peaks are well resolved. Formic acid has the same retention time as the reagent on this column, both appearing with a single well-defined peak (Figs. 3 and 5). On column C the three acid derivatives behave as on B, but any excess reagent distorts the acetic and propionic acid peaks making quantitative measurements impossible. Thus acetic and propionic acids can be anal-

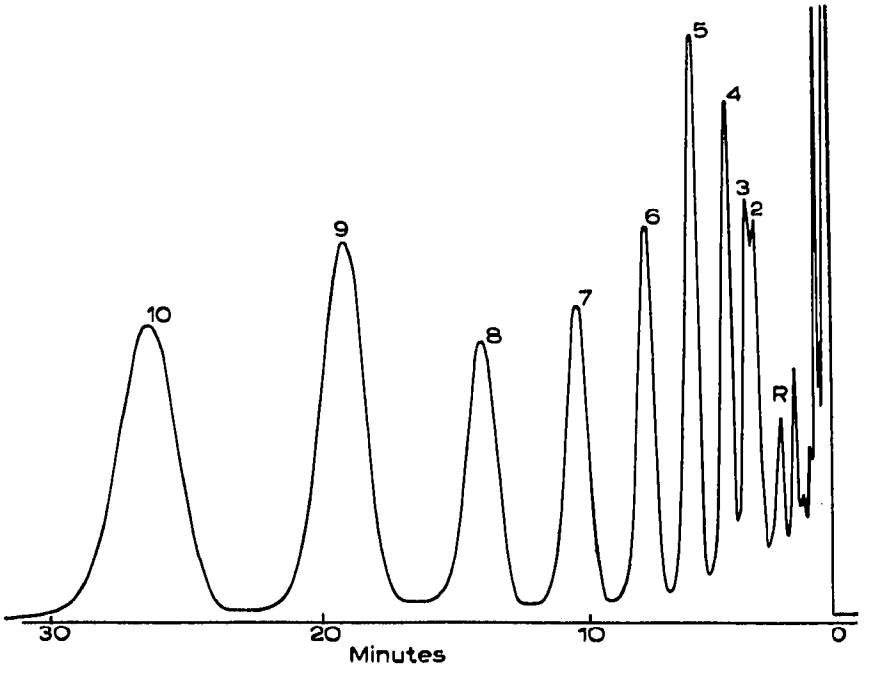

Fig. 3. C_2–C_{10} acids: *p*-bromophenacyl esters on 1:1 (w/w) 2.5% sodium dodecylbenzene sulphonate/1% Apiezon L (column B). Peak numbers as in Fig. 2.

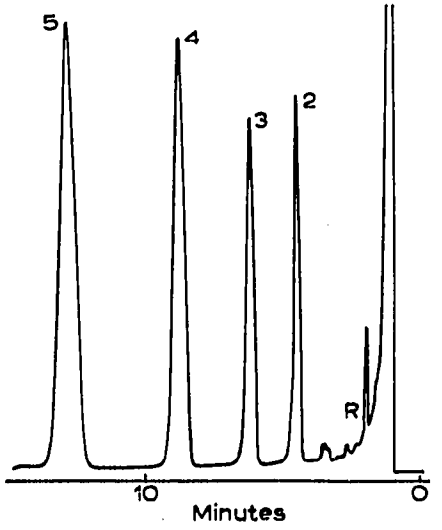

Fig. 4. C_2–C_5 acids: *p*-phenylphenacyl esters on 1:1 2.5% DBS/1% Apiezon L. Peak numbers as in Fig. 2.

ysed on column B whether or not formic acid is present. They may also be analysed on column C provided no excess reagent is present (Fig. 9).

The *p*-phenylphenacyl ester of acetic and propionic acids are quantitatively resolved on columns B and C. Formic acid if present comes out with the reagent and does not interfere with the C_2 and C_3 peaks. Hence acetic and propionic acids can also be analysed on either of the two columns as the *p*-phenylphenacyl esters.

When a mixture contains the three low-molecular-weight acids, formic, acetic and propionic, the latter two may be determined as described above on column B

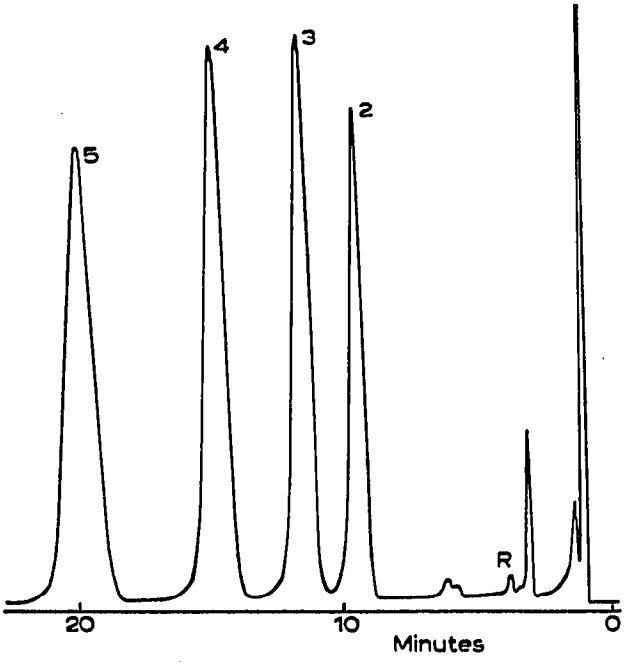

Fig. 5. C_2–C_5 acids: *p*-bromophenacyl esters on 1:1 2.5% DBS/1% Apiezon L (column B). Peak numbers as in Fig. 2.

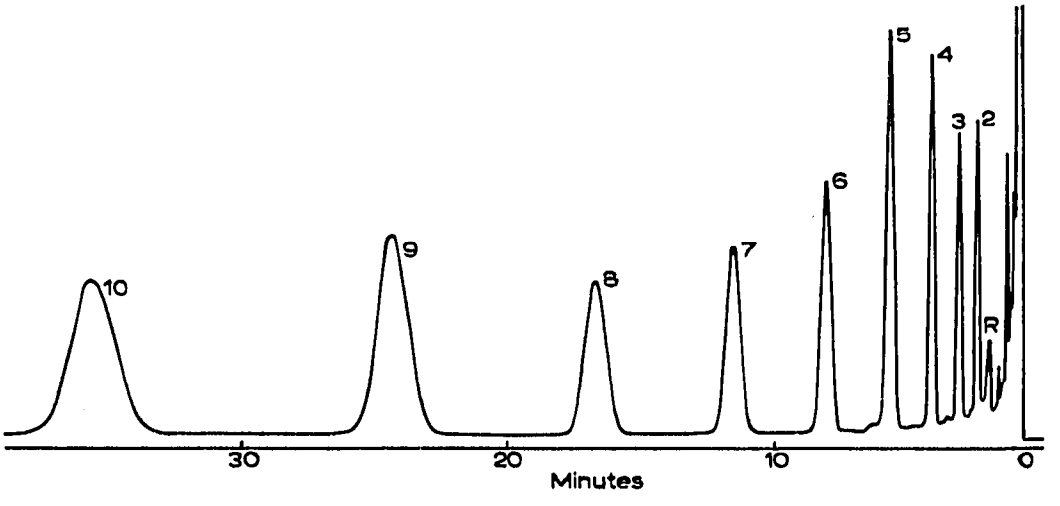

Fig. 6. C_2–C_{10} acids: *p*-bromophenacyl esters on 1% Apiezon L (column C). Peak numbers as in Fig. 2.

or C, while formic acid is determined as the anilide on column A at 200° oven temperature as earlier described[1].

The *p*-bromophenacyl esters of the rest of the acids, C_4–C_{10}, are quantitatively resolved on columns A, B and C, and the acids can be conveniently analysed (Figs. 2, 3, 5, 6 and 9). Only the C_2–C_8 acids can be conveniently chromatographed as their *p*-phenylphenacyl esters on columns B and C for quantitative purposes. Nonoic and decanoic acid esters take too long to come off the columns (Fig. 8).

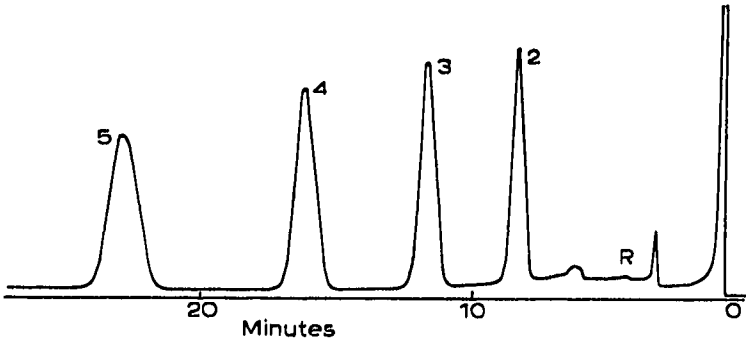

Fig. 7. C_2–C_5 acids: *p*-phenylphenacyl esters on 1% Apiezon L (column C). R, reagent; 2, acetic; 3, propionic; 4, *n*-butyric and 5, *n*-valeric.

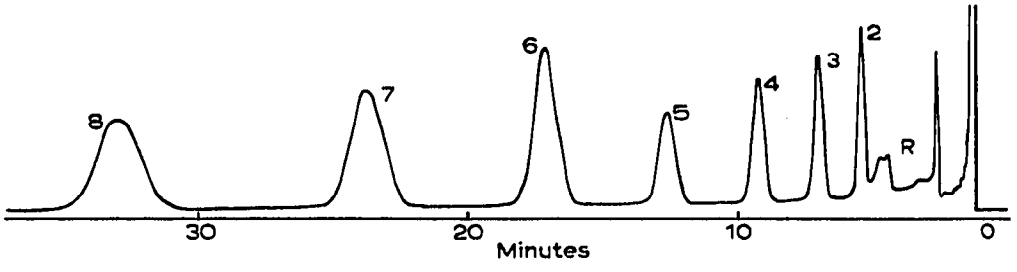

Fig. 8. C_2–C_8 acids: *p*-phenylphenacyl esters on 1% Apiezon L (column C). Peak numbers same as in Fig. 2.

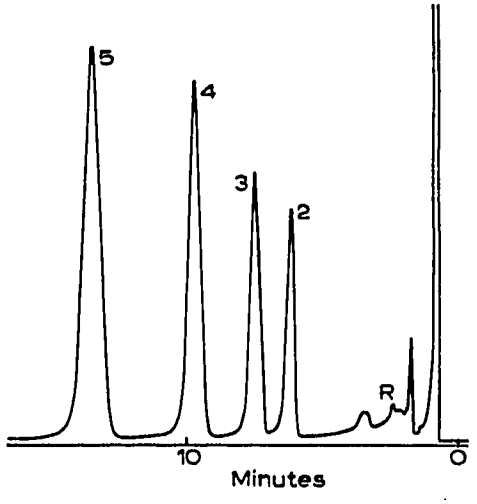

Fig. 9. C_2–C_5 acids: *p*-bromophenacyl esters on 1% Apiezon L (column C). The peak numbers as in Fig. 7.

For the analysis of mixtures containing formic, acetic and propionic acids, it is undesirable to use excess reagent in the ester formation. The reagent peaks often interfere with the peaks of these acids on all the columns. The degree of interference depends on the amount of reagent in excess. This interference is greatly eliminated or reduced by using an amount of reagent, calculated in millimoles, one half or more, but no more than, the estimated total number of millimoles of acids present. Whatever fraction of reagent used on the samples must also be used on the standard solutions. However, when dealing with mixtures containing C_4–C_{10} acids, a two-fold excess of reagent may be used without the risk of any interference with the acid peaks.

It is quite satisfactory to use any convenient ester of one of the C_2–C_{10} acids as an internal standard when dealing with a limited group of them. For example in the analysis of a mixture containing C_2–C_4 and C_6–C_{10} acids, valeric acid ester served as the internal standard.

The retention times of the bromophenacyl and phenylphenacyl esters of C_2–C_{10} and C_2–C_8 acids respectively on the various columns vary with the number of carbon atoms on the chain (Fig. 10). The esters of acids in an unknown aqueous mixture may be formed and injected into column A, B or C. By the examination of the retention times and comparison with an authentic mixture, the acids present may be qualitatively identified.

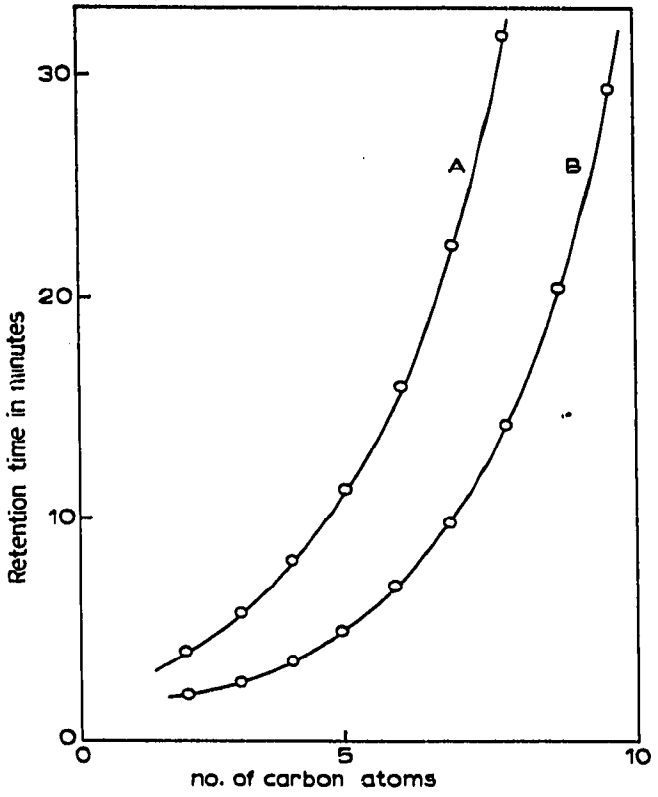

Fig. 10. Variation of the retention time with the number of carbon atoms on the carboxylic acid chains. (A) *p*-phenylphenacyl derivatives; (B) *p*-bromophenacyl derivatives.

REFERENCES

1 E. O. UMEH, *J. Chromatogr.*, 51 (1970) 139, 147.
2 E. O. UMEH, unpublished work.
3 A. I. VOGEL, *Practical Organic Chemistry*, 3rd Ed., Longmans, London, 1956, pp. 362, 363.