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LIQUID CHROMATOGRAPHY ON MACROPOROUS SILICA MODIFIED BY LAYERS OF CARBON AND WEAKLY POLAR POLYMERS*

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SUMMARY

The surface of the macroporous silica adsorbent Silochrom has been modified with layers of carbon and an organosilicon polymer and the modified adsorbents have been used in liquid chromatography with strongly and weakly polar eluents. Hydrocarbons and their derivatives with a large hydrocarbon nucleus are eluted from the column in order of increasing molecular weight and in order of increasing size of the hydrocarbon nucleus of the derivatives. This effect is caused by the increase in adsorption energy with increase in molecular size and corresponds with measurements of heats of wetting and adsorption isotherms for fatty alcohols and acids from alcoholic and aqueous solutions on charcoal and carbon black. When the polar eluent is replaced with a non-polar eluent, the same weakly specific adsorbent separates substances according to its capability of effecting specific intermolecular interactions with the functional groups. The term "reversed-phase chromatography" is not an accurate description of the adsorption process in this separation.

INTRODUCTION

In adsorption liquid (liquid-solid) chromatography, different specific¹ adsorbents, namely silica gels and aluminas, are usually used. Separations on such specific adsorbents are determined mainly by specific intermolecular interactions of the functional groups of the molecules of the components to be separated with hydroxyl groups and other sites of specific adsorption on the surface of the adsorbent. It is also customary to assume² that adsorption liquid chromatography cannot be used for the separation of homologues and substances that differ in molecular weight. True, alkanes and other compounds that differ in the number of alkyl groups in the molecule are not separated on polar adsorbents when eluted by non-polar or weakly polar solvents.

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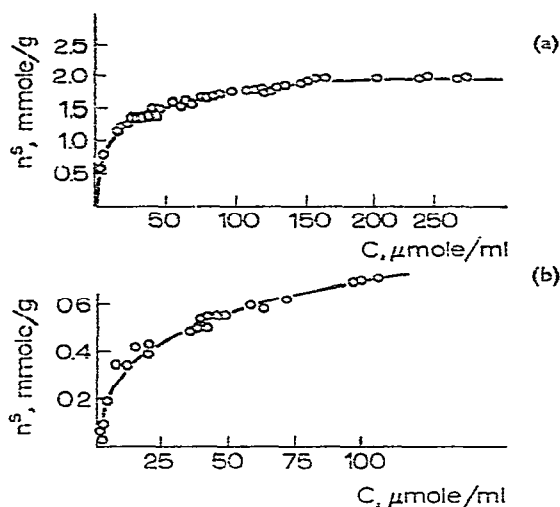


Fig. 1. Isotherms for adsorption of phenol: (a) from *n*-heptane solutions on wide-pore silica gel; (b) from aqueous solutions on channel carbon black.

The study of heats of wetting with solutions³ and adsorption from solutions³⁻⁶ on which the adsorption liquid chromatography is based shows the following, however. (1) Adsorption of a particular substance occur on both specific and non-specific adsorbents if an appropriate solvent is used (non-polar in the former and polar in the latter instance). Fig. 1 shows a comparison of such adsorption isotherms (plotted according to the data of refs. 4 and 5). (2) An increase in the number of carbon atoms in an organic molecule that contains a polar functional group sharply increases the adsorption from solutions in polar solvents on non-porous and porous non-specific adsorbents. This is shown in Fig. 2 (data taken from ref. 6). (3) An increase in the size of the hydrocarbon nucleus of a molecule that contains a functional group results in

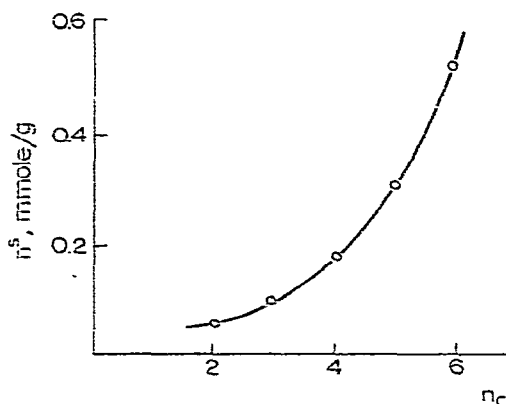


Fig. 2. Dependence of Gibbs' adsorption values, n^s , for fatty acids on Spheron-6 carbon black from aqueous solutions at an equilibrium concentration of $c \approx 0.015$ mmole/ml on the number of carbon atoms in the molecule, n_c .

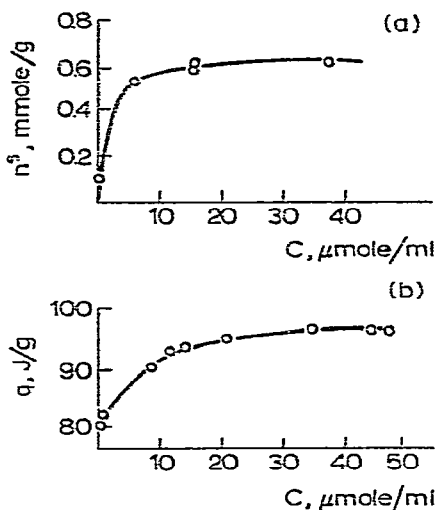


Fig. 3. (a) Adsorption isotherm for octadecanol from solutions in methanol on wide-pore charcoal; (b) corresponding dependence of heat of wetting of this charcoal by solutions of octadecanol in methanol on concentration.

positive adsorption and a larger energy of adsorption of higher homologues from their solutions in lower homologues. This effect can be seen in Fig. 3 (comparing data from ref. 3).

Hence the extent of adsorption (at low concentrations) and the heat of adsorption from solutions in polar solvents increase with increasing molecular weight on non-specific and weakly specific adsorbents. Therefore, the so-called "reversed-phase chromatography" technique is based, from the point of view of the theory of adsorption from solutions, simply on the positive adsorption of less polar organic substances from a more polar solvent on a non-specific or weakly specific adsorbent, or on the positive adsorption of a substance that has the same polar functional group as the solvent, but a larger hydrocarbon nucleus. Hence there is no reason to introduce a special term such as "reversed-phase chromatography". Adsorption liquid chromatography of any type is based on intermolecular interactions of the components of the solution with the adsorbent and the eluent. In "reversed-phase chromatography", non-specific intermolecular interactions of the components with the adsorbent mainly take place.

In 1948, Boldingh⁷ first used this choice of adsorbent and eluent for liquid chromatography for the separation of the methyl esters of fatty acids. Recently, this choice has been used to separate condensed aromatic substances⁸⁻¹¹, vitamins^{10,12}, insecticides¹³, antibiotics¹⁴, steroids⁸ and many other substances. In this connection, the study of the possible application of adsorbent supports (*i.e.*, supports with a high specific surface area) modified with layers of non-polar or weakly polar substances is of great interest. In this work, we modified an adsorbent support (macroporous silica) with layers of pyrolytic carbon and layers of an organosilicon polymer.

SILLOCHROM COVERED WITH A LAYER OF CARBON

The method of modification of silica adsorbent surfaces by carbon layers¹⁵ is based on the catalytic conversion of benzene accompanied by the formation of carbon deposits on the support surface heated to 700–850°. The modified adsorbents are mechanically strong and, as far as their surface chemistry is concerned, they are similar to carbon adsorbents.

The macroporous silica Silochrom C-280 was used as the adsorbent support¹⁶. Its surface was covered with a layer of pyrolytic carbon at a level of *ca.* 5 mg/m² at 850°. The adsorbent obtained was named Carbosilochrom. Liquid chromatography was carried out with a dielectric capacity detector, using a 350 × 3 mm I.D. column filled with adsorbent (particle size 80–160 μm). Diethyl ether, *n*-hexane and isopropanol were used as eluents. Capacity factors, K_c (the ratio of the corrected retention time of a substance being investigated to the retention time of a virtually non-adsorbing substance² characterizing the distribution of the adsorbate in the adsorbed layer and in the liquid mobile phase of the column) were determined. Adsorption processes in liquid chromatography are competitive¹⁷, and the retention of organic molecules therefore depends greatly on the nature of eluent used. The capacity factors for the adsorption of some substances from different eluents on to modified Silochrom are given in Table I.

TABLE I

CAPACITY FACTORS OF DIFFERENT ADSORBATES ON A COLUMN PACKED WITH SILLOCHROM MODIFIED WITH A CARBON LAYER USING THREE ELUENTS

<i>Adsorbate</i>	K_c		
	<i>n-Hexane</i>	<i>Diethyl ether</i>	<i>Isopropanol</i>
<i>n</i> -Nonane	0	0	0.2
<i>n</i> -Tetradecane	—	0	0.5
<i>n</i> -Hexadecane	—	0	1.0
<i>n</i> -Heptadecane	—	—	2.1
Benzene	0	0	0
Di- <i>n</i> -propyl ether	0.8	—	—
Acetone	3.5	0	0
Isobutanol	4.7	—	—

With the non-polar eluent *n*-hexane, strong retentions of acetone, ester and alcohol molecules were observed, due either to insufficient modification of the adsorbent support surface with the carbon layer or to polar groups on the surface of the carbon film itself interacting specifically with polar molecules.

When the more polar eluents diethyl ether and isopropanol are used, their adsorption energy is near to that of acetone and acetone is therefore eluted from the column with almost the same retention time as that of the eluent (*i.e.*, $K_c = 0$). The influence of the polarity of the eluent on the retention of alkanes is opposite to that on acetone: when using isopropanol as the eluent, the adsorption of alkanes is positive. In this instance, an increase in the number of carbon atoms in the molecule causes an increase in the non-specific intermolecular interaction of alkanes with the

carbon surface and selectivity of the adsorbent towards hydrocarbon homologues is observed.

Fig. 4 shows that the retention of *n*-alkanes in a column filled with Carbosilochrom when they are eluted with isopropanol increases very rapidly with an increase in the number of carbon atoms, n_C , in the *n*-alkane molecule. For a more rapid elution of higher alkanes ($n_C \geq 17$) from the column with this adsorbent it is necessary to decrease the polarity of the eluent, isopropanol, by adding to it a less polar component such as diethyl ether or *n*-hexane.

In order to separate wide fractions of high-boiling *n*-alkanes, gradient elution using isopropanol and a non-polar component can be used.

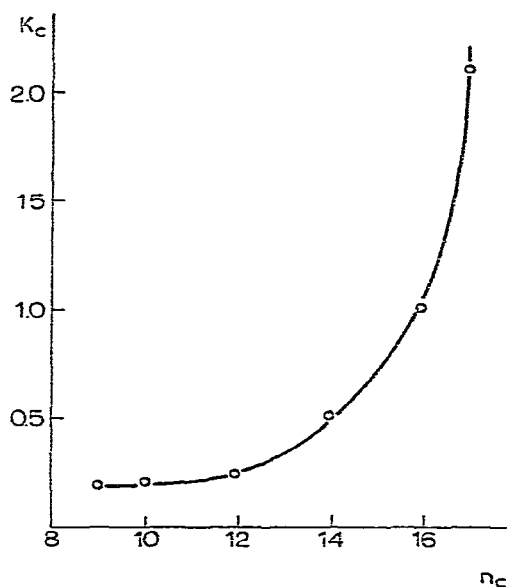


Fig. 4. Dependence of capacity factors, K_c , on Carbosilochrom on the number of carbon atoms in *n*-alkane molecules. Eluent, isopropanol.

SILOCHROM COVERED WITH AN ORGANOSILICON POLYMER CHEMICALLY BOUND TO THE SURFACE

Another means of modifying the surface of Silochrom C-80 is the chemical reaction of the silica surface with a weakly polar organosilicon polymer. We used a Perkin-Elmer 1220 liquid chromatograph with a UV detector and a stainless-steel column, 50 cm \times 2.6 mm I.D. A mixture of water and methanol in different proportions was used as the eluent.

Figs. 5-7 show separation of some alkyl derivatives of benzene, naphthalene and phenol. It can be seen that the order of elution of the alkyl derivatives is determined by the number of methyl groups in the molecule. It is noteworthy (Fig. 7a) that with the same number of methyl groups, the dimethyl ether of phloroglucinol is eluted from the column earlier than *p*-xylenol (2,5-dimethyl-1-hydroxybenzene) in spite of the higher molecular weight of the former. The lower retention of this ether

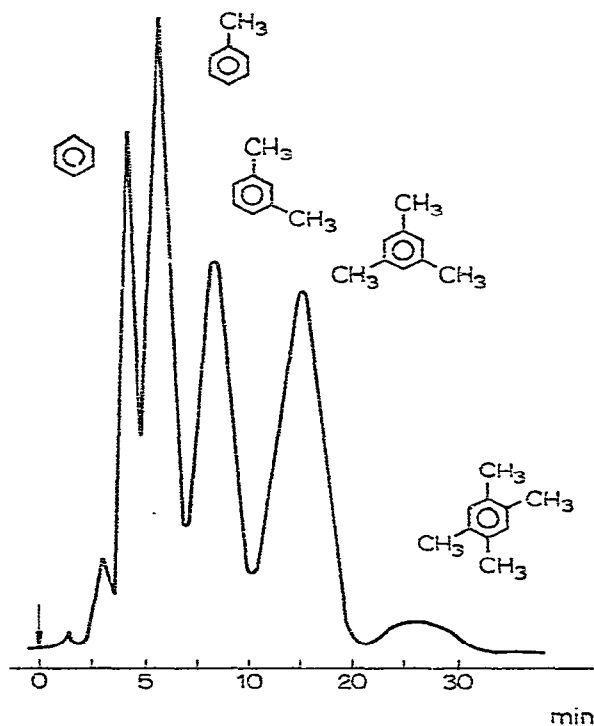


Fig. 5. Separation of polymethylbenzenes on an organosilicon polymer film deposited on Silochrom C-80. Eluent, methanol-water (1:1), flow-rate 1.1 ml/min; 50°; UV detector (254 nm).

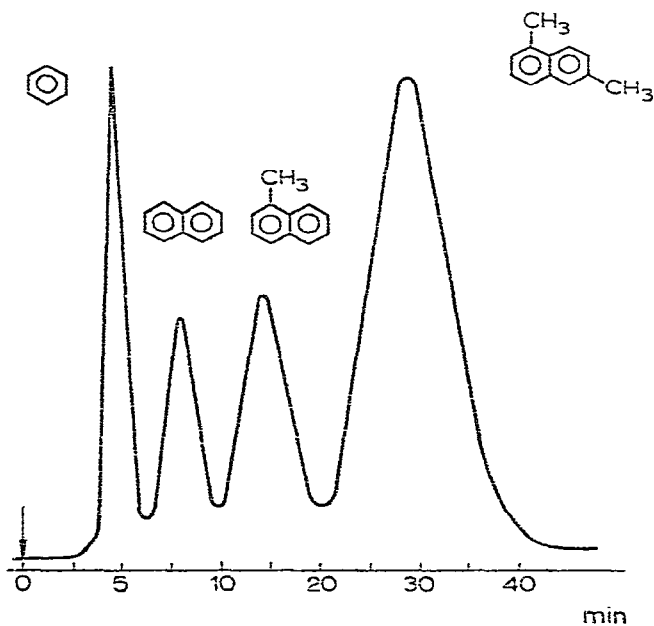


Fig. 6. Separation of methylnaphthalenes on organosilicon polymer film deposited on Silochrom C-80. Conditions are as in Fig. 5.

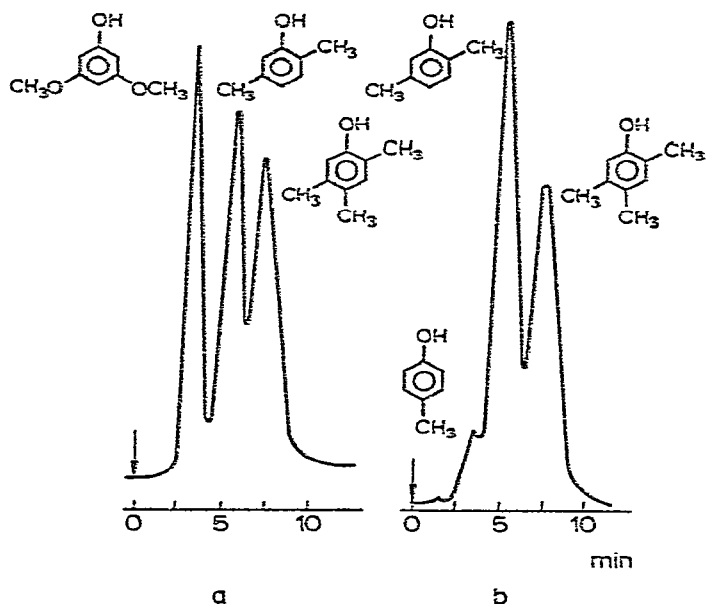


Fig. 7. Separation of methyl derivatives of phenols on organosilicon polymer film deposited on Silochrom C-80. Eluent, methanol-water (3:7), flow-rate 1.4 ml/min. Other conditions as in Fig. 5.

compared with that of *p*-xylenol is due to the larger number of oxygen atoms in its molecule, which causes an increase in the interaction of the molecules with the molecules of the polar eluent (methanol and water). With amino derivatives of benzene and diphenyl, an increase in the retention is observed with increasing molecular weight and number of methyl groups (Fig. 8).

The retention volumes of alkylbenzenes when separated on non-specific adsorbents depend strongly on the nature of the eluent. When the methanol content

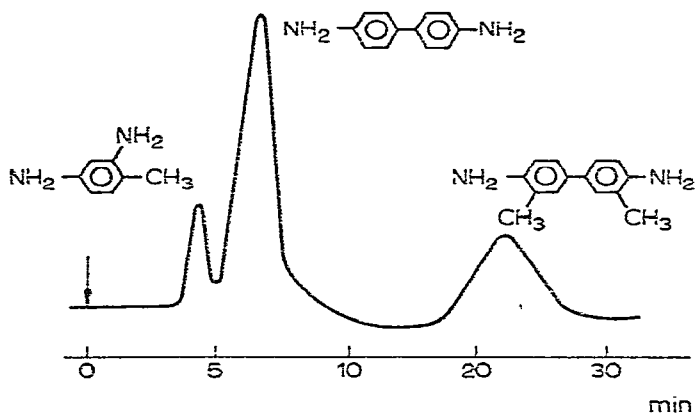


Fig. 8. Separation of aromatic amines on organosilicon polymer film deposited on Silochrom C-80. Eluent, methanol-water (15:85), flow-rate 1 ml/min. Other conditions as in Fig. 5.

of the methanol-water eluent is increased, the retention volume decreases (Fig. 9a), owing to the increase in the interaction between the alkylbenzenes and the eluent.

The selectivity of the separation of toluene and benzene and, correspondingly, *m*-xylene and benzene, also decreases for the above reasons when the methanol content in the eluent is increased (Fig. 9b). With an increase in the number of methyl groups in alkylbenzenes and alkylnaphthalenes, the selectivity of the separation of benzene and its alkyl derivatives (and correspondingly of naphthalene and its alkyl derivatives) increases. The same results are observed for *p*-cresol and other methyl-substituted phenols (Fig. 10).

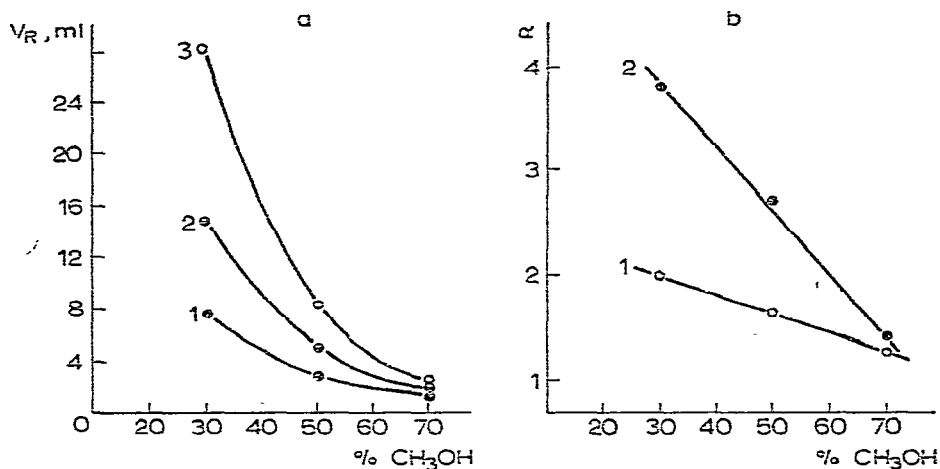


Fig. 9. Dependences on eluent composition (percentage of methanol in water) of: (a) retention volumes of benzene (1), toluene (2) and *m*-xylene (3) on organosilicon polymer film deposited on Silochrom C-80; (b) selectivity coefficients, α : toluene/benzene (1) and *m*-xylene/benzene (2) on organosilicon polymer film deposited on Silochrom C-80.

Fig. 11 shows that the selectivity of the separation of benzene and toluene and also that of benzene and *m*-xylene decreases with increase in temperature. However, the influence of temperature is not large compared with that in separations due to specific interactions¹⁸. The decrease in the retention volume of toluene with increase in temperature depends on the proportions of water and methanol in the eluent (Fig. 12). The greatest decrease is observed when the proportion of methanol is 30%. The logarithms of the retention volumes of alkylbenzenes and alkylnaphthalenes increase approximately linearly with the number of carbon atoms in the molecule (Fig. 13). It should be noted that if under these conditions toluene is retained more weakly than naphthalene, *m*-xylene is retained more strongly than naphthalene. In all instances, the separation occurred mainly due to the high energy of the non-specific intermolecular interaction of hydrocarbons and their derivatives with the weakly specific adsorbent. However, it is possible to separate polar molecules also owing to their specific intermolecular interactions with weakly specific adsorbents and with mixed specific and non-specific adsorbents if a non-polar eluent is used. For instance, nitroanilines are eluted by *n*-hexane on Silochrom modified with a weakly polar organosilicon polymer in the order *ortho*-, *meta*- and *para*-isomers. When polar iso-

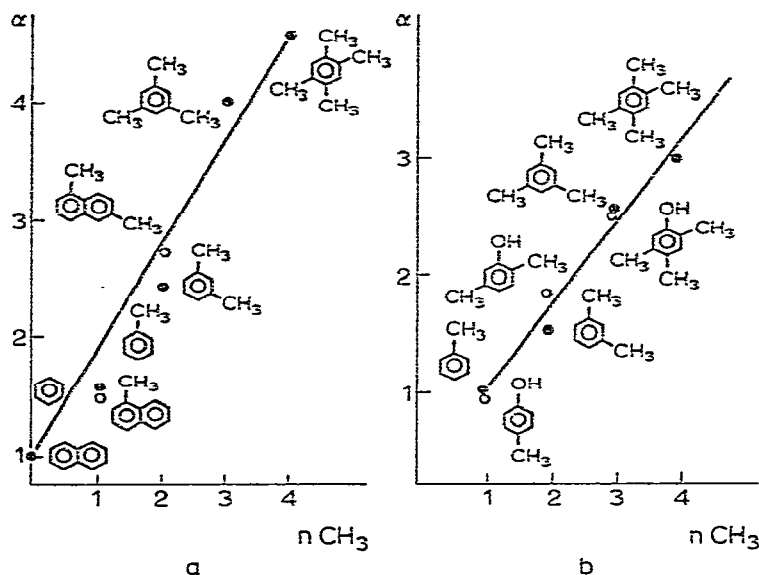


Fig. 10. Dependence of selectivity coefficient, α , on the number of methyl groups, n_{CH_3} , for aromatic hydrocarbons and their derivatives on organosilicon polymer film deposited on Silochrom C-80 [eluent, methanol-water (1:1); 50°]: (a) methylbenzenes relative to benzene (●) and methylnaphthalenes relative to naphthalene (○); (b) methyl-substituted phenols relative to *p*-cresol (○) and, for comparison, methylbenzenes relative to toluene (●).

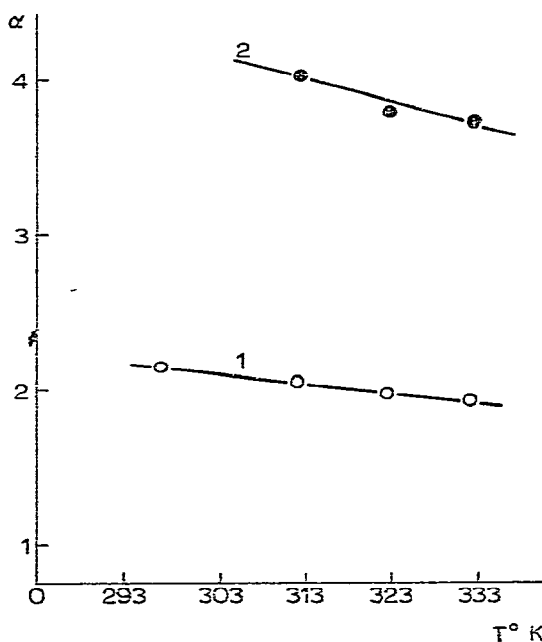


Fig. 11. Dependence of selectivity coefficient, α , on temperature for toluene (1) and *m*-xylene (2) relative to benzene on organosilicon polymer film deposited on Silochrom C-80. Eluent, methanol-water (3:7).

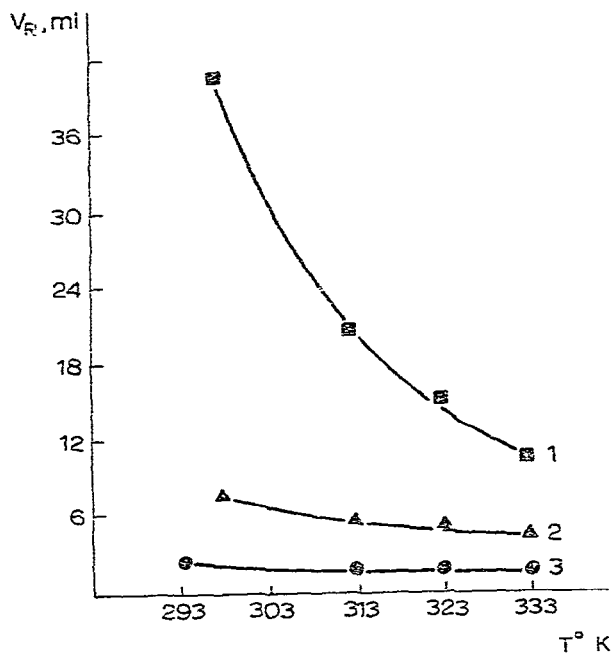


Fig. 12. Dependence of retention volume of toluene on organosilicon polymer film deposited on Silochrom C-80 on temperature with different methanol-water eluent compositions: 1, 3:7; 2, 1:1; 3, 7:3.

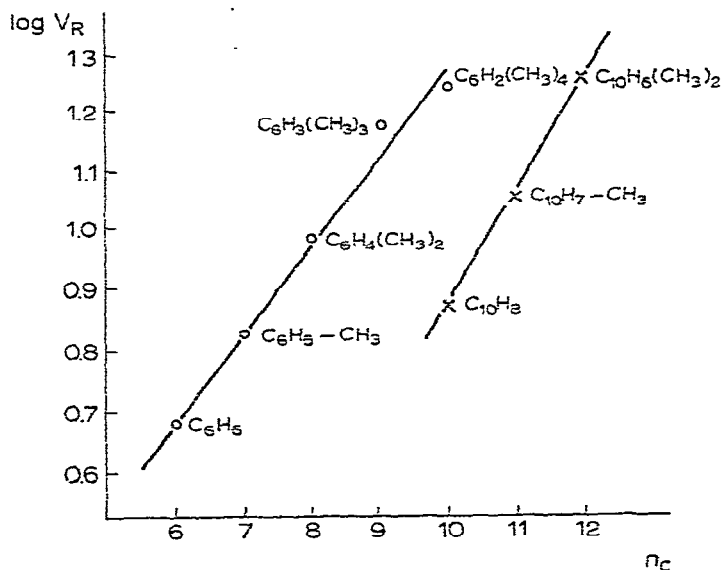


Fig. 13. Dependence of logarithm of retention volume on number of carbon atoms in molecule, n_C , for (left) methyl-substituted benzenes and (right) methyl-substituted naphthalenes.

propanol is added to the *n*-hexane, the retention volumes decrease. The best separation of nitroaniline isomers was obtained with *n*-hexane containing 0.2% of isopropanol as the eluent.

The results obtained are in agreement with the fact that retention volumes and separation selectivities in liquid chromatography are determined not only by interaction of the component with the adsorbent but also by its interaction with the eluent and by interaction of the eluent with the adsorbent¹⁷. By changing the relationship between the energies of these interactions, simply by altering the nature or composition of the eluent, one can achieve a transition from chromatography with a non-polar or weakly polar eluent to that described in this paper, *viz.*, chromatography with a polar eluent and the same weakly specific adsorbent. In fact, if the difference in the intermolecular interaction energies is due mainly to a specific interaction of polar functional groups of the component molecule with the adsorbent (the non-specific interaction of its alkyl groups is similar to that for corresponding non-polar eluents such as alkanes), then the process usually considered as a "common" variant of liquid chromatography takes place. Conversely, when this difference in intermolecular interaction energies is due mainly to a non-specific interaction of the hydrocarbon part of the molecule with the adsorbent (the specific interaction of polar functional groups of the component being similar to those for polar eluents), then the process that is called "reversed-phase chromatography" occurs. This indicates that the term "reversed-phase chromatography" is not an accurate description of the adsorption process in this separation.

One should take into account that adsorption chromatography on weakly specific and mixed adsorbents with elution with polar eluents can occur only in the separation of non-polar molecules or molecules with a large hydrocarbon nucleus. In the separation of polar molecules with a small hydrocarbon nucleus, a non-specific interaction between the component and the adsorbent and a specific interaction between the component and the eluent will mainly take place. For polar molecules of a component with a small hydrocarbon nucleus the latter interaction decreases the retention volumes of these components. The correlation between the intermolecular component-adsorbent, eluent-adsorbent and component-eluent interaction energies should always be taken into consideration and not simply such an indirect parameter as the solubility of the component in the eluent^{2,10}, which may give no information on the interaction of the component with the adsorbent. Attempts to explain the retention differences in series of substances in terms of their different solubilities in a particular eluent have repeatedly been made^{10,12}. Such an explanation, however, cannot be adequate as retention in adsorption liquid chromatography is caused not simply by interactions of the component and eluent molecules but mainly by the interactions of component and eluent molecules with the adsorbent.

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