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THE ROLE OF MOLECULAR INTERACTIONS IN CHROMATOGRAPHY

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SUMMARY

A dynamic equation for the distribution coefficient is proposed that accounts for the different solute-phase interactions in chromatography. The validity of the basic form of the equation is experimentally verified and the effect of phase composition on retention is shown to be dependent on the probability of solute-phase interactions. Using the equation, the change in retention volume with phase composition can be accurately predicted in liquid-solid chromatography, thin-layer chromatography, ion-exchange chromatography and gas chromatography.

Where polar forces between the solute and each phase are weak, it is shown that dispersive interactions in the mobile phase are proportional to the density of the dispersing solvent. The magnitude of the polar interactive effects of both solute and solvent are shown to be proportional to the exponent of the polarizability per milliliter of the respective solute or solvent for a limited number of substances having monofunctional groups only.

INTRODUCTION

Chromatography has been classically defined as a separation that is achieved by the distribution of substances between two phases, a mobile phase and a stationary phase. Those substances distributed preferentially in the mobile phase will move through the system more rapidly than those distributed preferentially in the stationary phase.

Although scientifically correct, this definition is rather vague and while it introduces the essential concepts of a mobile and a stationary phase, it does obscure the basic phenomena by which a separation is effected in the term "distribution". The distribution of a solute between two phases results from the forces that exist between solute molecules and the molecules of each phase. In liquid chromatography these forces can be polar in nature, arising from permanent or induced electric fields associated with both solute and solvent molecules, or non-polar, due to London's dispersion forces (Van der Waal's forces), which depend on the relative masses of the solute and solvent molecules. In ion-exchange chromatography the major forces between the solute molecules and the two phases arise from the permanent electric charges on the solute ions and the ions in the two phases. Separations in chromatography are therefore achieved by exploiting the different molecular forces that can occur between each

solute and the two phases. It follows that if the nature of the interactions between the solute molecule and the two phases were understood, the behavior of a particular solute in a given phase system could be predicted and, as a result, chromatographic separation could be made more effective, more rapid and the chromatographic conditions could be chosen on a rational basis.

There have been several approaches made to provide a theoretical explanation of these interactions in order to aid in phase selection. The most common approach¹ is to use the thermodynamic relationship that describes the distribution coefficient as a function of the exponent of the partial molar excess free energy associated with the removal of the solute from one phase to another. However, the thermodynamic approach has severe limitations from the practical point of view.

Thermodynamic properties are bulk properties and are a measure of the net interactive effects and, although thermodynamic functions can be assigned to the different interactions², there is usually more than one effect present and the individual interactions cannot easily be separated or identified. Another disadvantage of this approach results from the fact that there is very limited thermodynamic data available on even a few solutes in the multiplicity of solute-phase systems that one could choose from. Further, it is difficult and time consuming to obtain such data.

In this paper a dynamic equation for the distribution coefficient is developed that provides separate functions for the different types of molecular interactive forces and takes into account the probability that any specific interaction may occur. The argument is based on that given in a previous publication by the same author³, but a somewhat different approach is used and further experimental evidence is provided that supports the equation given.

THEORY

The distribution of a solute between two phases results from the balance of forces between solute molecules and the molecules of each phase, as stated in the Introduction. These forces can be polar in nature, arising from permanent or induced electric fields associated with both solute and solvent molecules, or be due to London's dispersion forces (Van der Waal's forces). The exact nature of these forces is not, at present, known and the so-called polar and dispersive forces may be essentially the same in character, but of different magnitude. For the purpose of this paper, however, it will be assumed that two forces are different and that each requires to be accounted for separately. This approach will also simplify the theoretical arguments necessary to describe interactions in mobile phases that consist of polar solvents such as ethyl acetate in dispersive solvents such as heptane. In ion-exchange chromatography the forces on the solute molecules will be substantially ionic in nature, but may include the so-called polar and non-polar forces as well.

It is, therefore, possible to define the distribution coefficient (K) of a solute between the two phases in a chromatographic system in the following way

$$K = \frac{\text{The magnitude of the total forces acting on the solute in the stationary phase}}{\text{The magnitude of the total forces acting on the solute in the mobile phase}}$$

The above word equation, however, is rather loose in that it conjures up an impression that there is a group of molecules permanently held in the stationary phase and another

group permanently held in the mobile phase. This, of course, is not so, as molecular interactions in fluids are transient. Association will depend on the probability of a particular molecule coming in contact with another molecule of a specific type, on the position of contact and on its thermal energy relative to its potential energy of association. Thus the magnitude of the total forces acting on a solute in a given phase will be dependent on the following: the magnitude and the nature of the force between each solute molecule and a given molecule of that phase, their probability of interaction, their probability of position of contact, and their thermal energy.

For a series of n different types of interaction the word equation given above can be put in the following form

$$K = \frac{\left[\sum_{j=1}^{j=n} \Phi_j F_j P_j f_j(T) \right]_S}{\left[\sum_{j=1}^{j=n} \Phi_j F_j P_j f_j(T) \right]_M}$$

where Φ is a constant and incorporates the probability of position of contact as this will be decided by the size and geometry of the molecules concerned, F is the magnitude of the respective force between the solute molecule and the phase molecule, P is the probability of molecular interaction, and $f(T)$ is the thermal energy of the molecule at the time of contact and will contain another thermal probability factor but will be constant at constant temperature, and the subscripts S and M denote the stationary and mobile phases, respectively. If at any instant a molecule is in phase 1 at the interface between phase 1 and phase 2 and conditions favor strong interactions with a molecule in phase 1, it will remain in phase 1. If, however, instantaneous conditions favor strong interactions with a molecule in phase 2 but weak interactions with its neighbor in phase 1, the solute molecule will migrate from phase 1 into phase 2 and this will be the mechanism by which equilibrium is achieved and maintained.

Three types of interaction can be considered, namely ionic, polar, and dispersive, and then the above equation can be put in the form

$$K = \frac{(\Phi_i F_i P_i f_i(T) + \Phi_p F_p P_p f_p(T) + \Phi_D F_D P_D f_D(T))_S}{(\Phi_i F_i P_i f_i(T) + \Phi_p F_p P_p f_p(T) + \Phi_D F_D P_D f_D(T))_M}$$

where the subscripts i , p , and D denote ionic, polar, and dispersive interactions, respectively.

In the special case of liquid-solid chromatography where the adsorbent is silica gel and non-ionic mobile phases are employed, there will be no ionic forces effective in the separation.

Further, if the separations are carried out at constant temperature, $f(T)$ will be constant and can be incorporated in Φ , therefore

$$K = \frac{(\Phi_p F_p P_p + \Phi_D F_D P_D)_S}{(\Phi_p F_p P_p + \Phi_D F_D P_D)_M}$$

It is now necessary to expand the individual functions in the above equation to contain variables that are pertinent to liquid chromatography. In so doing the change in K that will result from a change in the magnitude of those variables should

be quantitatively predictable. The probability of interaction of a solute with one of the phases will be proportional to the concentration of the interacting moieties in each of the respective phases. Thus

$$K = \frac{(\Phi_p F_p c_p + \Phi_D F_D c_D)_S}{(\Phi_p F_p c_p + \Phi_D F_D c_D)_M}$$

where c_p and c_D are the concentrations of polar moieties and dispersive moieties in the respective phase.

If the hypothesis is made that the dispersive forces result from mass interaction (which will be subsequently substantiated), then c_D will be proportional to the density of the dispersion medium, which can be expressed as a concentration in terms of the mass/unit volume. Thus

$$c_D = Ad$$

where A is a constant and d is the density of the dispersion solvent in the mobile phase. It follows that

$$K = \frac{(\Phi_p F_p c_p + \Phi_D F_D c_D)_S}{(\Phi_p F_p c_p + \Phi_D F_D d)_M}$$

Now the corrected retention volume V' is equal to KA_s , where A_s can be the volume of the stationary phase or the total number of active moieties on the surface of a solid, depending on the chromatographic system and the manner in which the concentration of solute in or on the stationary phase is defined. Thus

$$V' = KA_s = \frac{(\Phi_p F_p c_p + \Phi_D F_D c_D)_S A_s}{(\Phi_p F_p c_p + \Phi_D F_D d)_M} \quad (1)$$

Eqn. 1 is a novel form of describing the corrected retention volume of a solute and has considerable practical significance. For example, the use of such an equation would allow the prediction of retention volumes and separation ratios for specific concentrations of a polar solvent in the mobile phase and for non-polar dispersion solvents of specific density, provided suitable reference data were available.

For practical convenience, eqn. 1 can be put in the inverse form

$$\frac{1}{V'} = \frac{(\Phi_p F_p c_p + \Phi_D F_D d)_M}{(\Phi_p F_p c_p + \Phi_D F_D c_D)_S A_s} \quad (2)$$

DISCUSSION AND EXPERIMENTAL VERIFICATION OF THE DYNAMIC EQUATION FOR THE DISTRIBUTION COEFFICIENT

Polar interactions

If different concentrations of polar or semi-polar solvents in a dispersion medium such as heptane are employed, then provided the concentration of polar solvent is kept above 2% w/v^{4,5}, the activity of the silica gel will be constant, the denominator in eqn. 2 becomes constant, and thus

$$\frac{1}{V'} = A + Bc_p \quad (3)$$

where A and B are constants.

Scott and Kucera³ examined the reciprocal relationship of the corrected retention volume V' and the concentration of polar solvent in the mobile phase and an example of their curves are shown in Fig. 1. Experimental details are given in their original paper. It is seen that the relationship predicted by eqn. 3 is completely verified and that the concentration of polar solvent in the mobile phase given in terms of mass/unit volume of mobile phase precisely controls the probability of interaction. It should also be noted that the linearity of the curve extends to a mobile phase composition of 90% v/v of polar solvent in heptane.

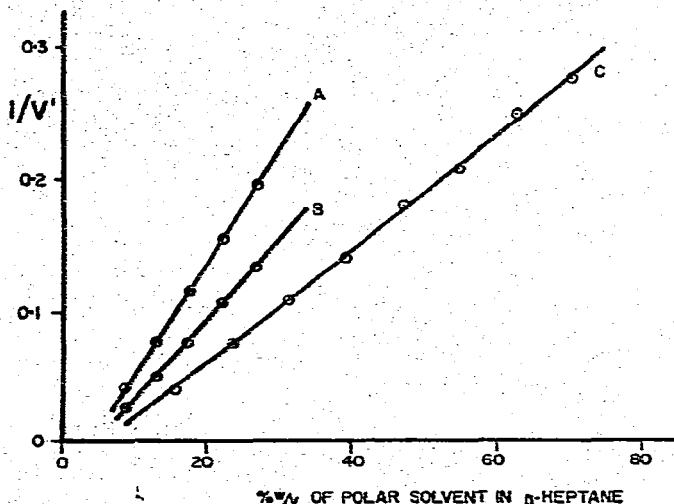


Fig. 1. Graphs relating the reciprocal of the corrected retention volume V' to the % w/v of polar solvent in *n*-heptane for different solutes. Column, 25 cm \times 4.6 mm I.D., packed with Partisil 10. Solutes: (A) benzyl alcohol; (B) 3-phenyl-1-propanol; (C) desoxycorticosterone alcohol. Polar solvents: (A and B) tetrahydrofuran; (C) isopropanol.

It has been previously shown^{4,5} that small concentrations of a polar solvent in a non-polar dispersive medium behave quite differently due to the silica gel being incompletely deactivated until the polar solvent is present at concentrations of greater than 1 or 2% w/v. It was, therefore, of interest to examine the change in the function given by eqn. 2 over that range of solvent concentrations where the silica gel was being deactivated. In Fig. 2 the results obtained from chromatographing a series of different solutes using different polar solvents in heptane are shown where the concentrations ranged from about 0.2–7.0% w/v. It is seen that where the concentration of polar solvent is insufficient to deactivate the silica gel, the linear relationship between $1/V'$ and polar solvent concentration breaks down. For solutes that have relatively low retention, that is, where the forces in both phases are of the same order, activation of the adsorbent as the concentration of polar solvent is reduced and the silanol groups of the silica gel become exposed, produces a rapid increase in retention volume and this increase occurs between 0.8% and about 1.2% w/v of polar solvent in heptane.

* Some of the figures have been re-published from *J. Chromatogr.*, 112 (1975) 425–442 for convenience of the readers.

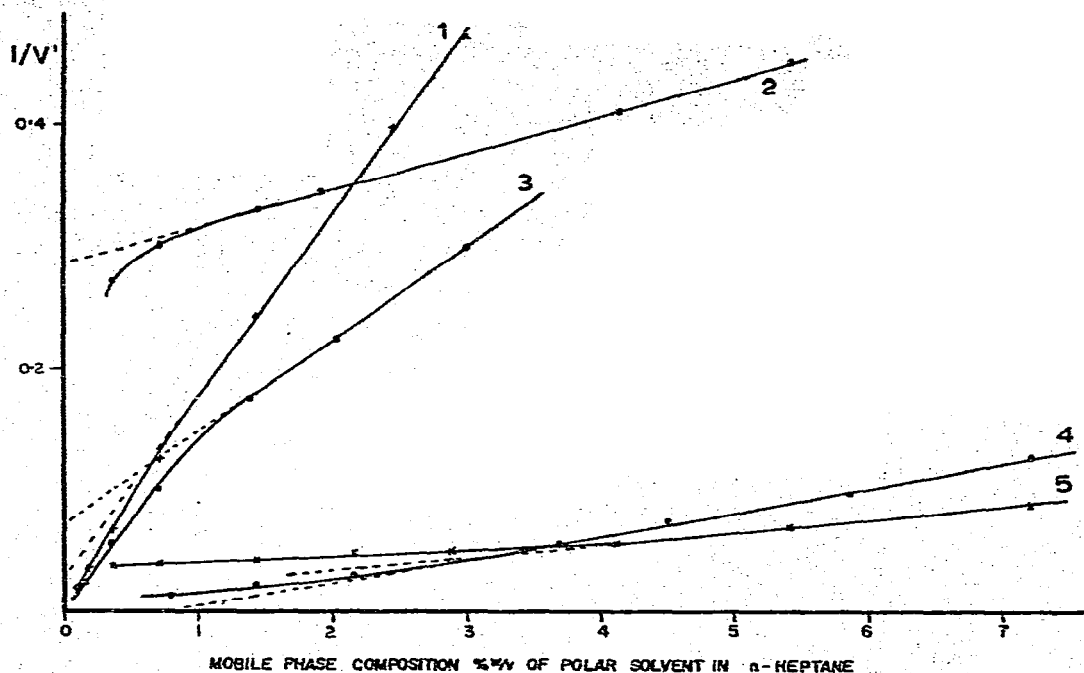


Fig. 2. Graphs relating the reciprocal of the corrected retention volume V' of different solutes to the % w/v composition of the mobile phase. Column, 10 cm \times 4.6 mm I.D., packed with Partisil 10. Solute: (1) phenyl methyl carbinol; (2) phenanthrene; (3) benzyl acetate; (4) phenyl methyl carbinol; (5) nitrobenzene. Polar solvents: (1) isopropanol; (2 and 5) chloroform; (3 and 4) ethyl acetate.

Under these circumstances, solute molecules have an opportunity to interact with silanol groups as opposed to adsorbed polar solvent molecules which normally cover the surface when the adsorbent is deactivated. As the interactive forces between the solute molecules and the hydroxyl groups of the silanol moieties are greater than the forces between solute molecules and the firmly adsorbed polar solvent molecules, V' rapidly increases and consequently $1/V'$ rapidly decreases.

However, if the solutes are well retained, the forces between solute molecules and the silanol groups are of the same order as those between the polar solvent in the mobile phase and the silanol groups. Under these circumstances, as the solute molecules are already interacting with the silanol groups of the silica gel, exposing more silanol groups produces no new situation. Thus, the shape of the curves relating $1/V'$ with the concentration of polar solvent in the mobile phase exhibits little change at low concentrations of polar solvent.

The relationship given in eqn. 2 can also be substantiated in ion-exchange chromatography. In Fig. 3 linear curves are shown relating $1/V'$ with buffer concentration in the separation of a number of nucleotides using a strong anion-exchange bonded phase column. Solute interactions in the mobile phase occur with the buffer moiety, which in this case is the dihydrogen phosphate ion and again it is demonstrated that the probability of interaction is conditioned by the concentration of that moiety in the mobile phase. Knox and Pryde⁶ also confirmed this relationship in their work on the separation of organic acids again using a bonded-phase anion ex-

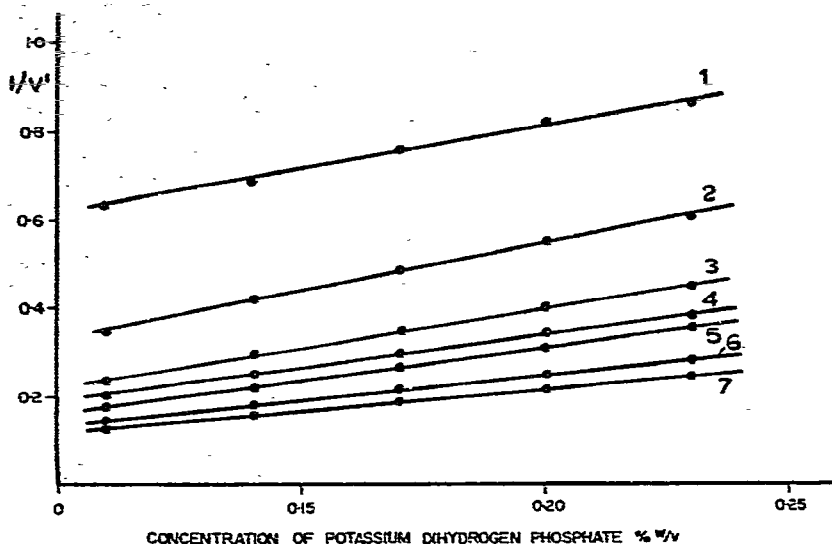


Fig. 3. Graph of $1/V'$ against buffer concentration for different solutes. Column, 25 cm \times 4.6 mm I.D., packed with Partisil 10 SAX, pH 4.80. Solutes: (1) adenosine 3',5'-cyclic monophosphate; (2) uridine 5'-monophosphate; (3) cytidine 5'-monophosphate; (4) adenosine 5'-monophosphate; (5) uridine 2',3'-diphosphate; (6) adenosine 2',3'-diphosphate; (7) guanosine 5'-monophosphate.

changer. In their work, however, they plotted k' against the reciprocal of the ionic strength of the buffer solution, which is algebraically equivalent to eqn. 2. These authors also obtained excellent linear correlation.

Eqn. 2 should be applicable to all distribution systems including thin-layer chromatography. However, if low concentrations of a polar solvent are employed in the mobile phase, a significant proportion of the solvent is adsorbed by the silica gel in the deactivation process and thus the concentration of polar solvent in the solvent front is significantly reduced, producing a quasi gradient elution effect. This, however, only occurs for low concentration of polar solvent and its effect is only significant on fast moving peaks which follow closely the solvent front for a significant proportion of the development. In thin-layer chromatography, the k' value of a given solute can be determined from the R_F factor and the reciprocal of k' is calculated by means of the following equation⁷

$$\frac{1}{k'} = \frac{R_F}{(1 - R_F)}$$

In Fig. 4 curves relating $1/k'$ against concentration of ethyl acetate in *n*-heptane are shown for the separation of three dyes. It is seen that excellent linear correlation is obtained except for the fast moving dye at low concentrations of ethyl acetate, which is explained in the discussion above. Thus in thin-layer chromatography solute retention is dependent on the probability of interaction of the solute with the polar molecules contained in the mobile phase, which in turn is dependent on the concentration of polar solvent in the mobile phase.

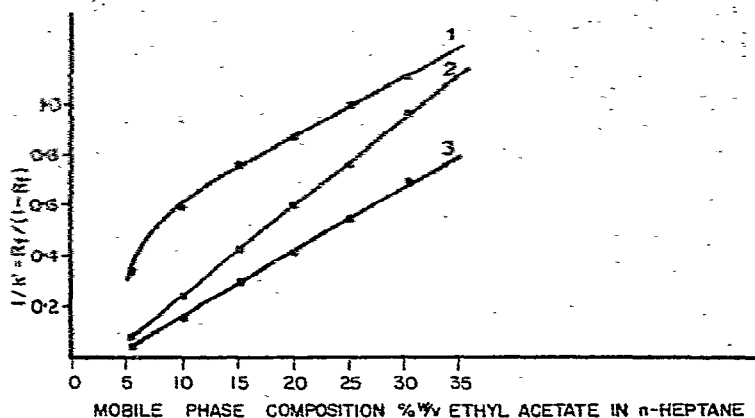


Fig. 4. Graph of $1/K'$ against concentration of the polar solvent in the mobile phase from the thin-layer separation of dyes. 1 = *p*-Dimethylaminoazobenzene; 2 = dihydroxyazobenzene; 3 = benzoquinone-*p*-hydroxyphenylimine.

Finally, the effect of solvent concentration on the probability of solute-solvent interaction should also be manifest in gas chromatographic separations. In gas chromatography, however, there are no significant interactions in the gas phase and thus the value of K will be directly related to interactions in the stationary phase only, thus the equation that describes the distribution coefficient will take the following form

$$V' = KA_s = (\Phi_p F_p c_p + \Phi_D F_D d) A_s$$

and eqn. 3 becomes

$$V' = A + Bc_p \quad (4)$$

Laub and Purnell⁸ examined the effect using mixed solvents in gas-liquid chromatographic separation. They measured the distribution coefficients of a number of solutes chromatographed on a series of stationary phase mixtures containing squalane and dinonyl phthalate. This system is directly comparable to a mobile phase of heptane and ethyl acetate used in liquid solid chromatography. The authors proved the following relationship

$$K_R = K_{R(A)}^0 V_A + K_{R(S)}^0 V_S$$

where K_R is the solute distribution coefficient for the mixture, $K_{R(A)}^0$ and $K_{R(S)}^0$ are the corresponding distribution coefficients of the solute in the pure phases A and S, respectively, and V_A and V_S are the volume fractions of phases A and S, respectively.

As V_A and V_S are volume fractions, the above equation can be rewritten in the form

$$K_R = K_{R(A)}^0 V + K_{R(S)}^0 (1 - V_A) = K_{R(S)}^0 + (K_{R(A)}^0 - K_{R(S)}^0) V_A$$

Further, if d is the density of A and c_A is the concentration of A in g/ml in the mixture, then

$$K_R = K_{R(S)}^0 + \frac{(K_{R(A)}^0 - K_{R(S)}^0) c_A}{d}$$

Hence, multiplying throughout by A_s ,

$$V' = K_R A_s = \left[K_{R(S)}^0 + \frac{(K_{R(A)}^0 - K_{R(S)}^0) c_A}{d} \right] A_s \quad (5)$$

Now eqn. 5 is the same form as eqn. 4 and thus the author's results also validate eqn. 4. The curves obtained are reproduced in Fig. 5. It is seen that excellent linear correlation is obtained substantiating the role played by the solvent concentration in controlling solute-solvent interactions.

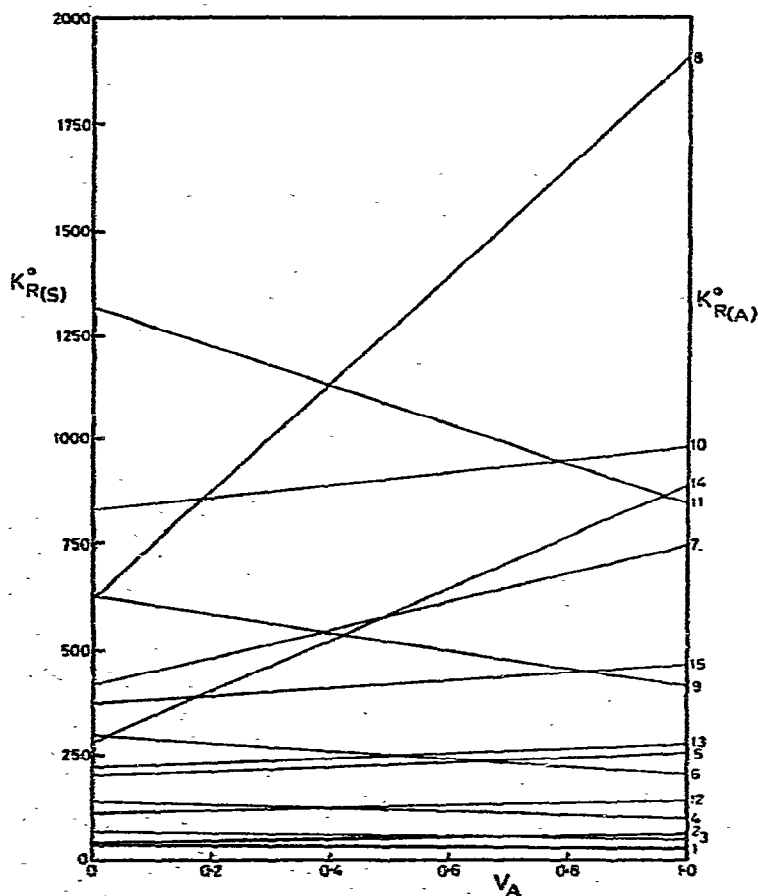


Fig. 5. Curves relating K_R , the solute distribution coefficient, to V_A , the volume fraction of dinonyl phthalate (A) in the stationary phase containing the dispersion solvent squalane (S). Column temperature, 100°.

From the results given so far it is clearly seen that the concentration of the polar solvent in the phase concerned governs the probability of interaction and for a given set of chromatographic conditions permits the corrected retention volume of a solute to be calculated by means of eqn. 3. Furthermore, the relationship between the probability of interaction and the concentration of interacting moiety has been confirmed for liquid-solid chromatography, ion-exchange chromatography, thin-layer chromatography, and gas chromatography.

Dispersive interaction

In eqn. 4 for liquid-solid chromatography, the constant A included dispersive interactions whereas the polar interactions were controlled and changed by varying c_p . To examine dispersive interactions, polar interactions must be kept constant and thus for a given phase system c_p must be kept constant. According to eqn. 2 dispersive interactions can be changed by varying the density of the dispersion solvent and thus eqn. 3 can be modified to the form

$$\frac{1}{V'} = A + Bd \quad (6)$$

Under conditions where both the solutes and the polar solvent are the same and the polar solvent is maintained at the same weight volume concentration in the mobile phase, the relationship between $1/V'$ and d , the density of the dispersing solute, indicate the degree and nature of the dispersive interactions between the solute and the diluting solvent and confirm or disprove eqn. 6. Scott and Kucera³ employed a 4.51% w/v of ethyl acetate in different hydrocarbons and determined the corrected retention volumes of the following solutes: 2-ethyl anthraquinone, 2-methyl anthraquinone, anthraquinone, phenyl methyl carbinol, benzyl alcohol, and 3-phenyl-1-propanol. The results are shown as curves relating $1/V'$ to the density of the dispersing solvent in Fig. 6. It is seen that excellent correlation is obtained.

However, for the more polar aromatic alcohols, a straight line is produced which is horizontal to the density axis and indicates that no significant dispersive interactions occur with the different hydrocarbon solvents. If the net dispersive interactions between the two phases are weak, then, for them to significantly affect the retention characteristics of a solute, the net polar interactions must also be weak or at least of the same order as the dispersive interactions. In the case of the aromatic alcohols, the polar forces are relatively strong and so changes in dispersive interactions have little effect on the retention of the solutes. In the case of the anthraquinone solutes, however, the polar forces on the solute both in the stationary phase and in the mobile phase are relatively weak and, therefore, changes in the dispersive interaction have a strong effect on solute retention. It follows that where polar forces are weak and dispersive forces can be exploited, then changes in density of the dispersing media can result in considerable changes in the elution order of a series of solutes³.

From the results shown in Fig. 6 it would seem that if the polar forces acting on the aromatic alcohols in the two phases could be reduced, dispersive interactions could be made to influence their retention. The polar forces between the solutes and the silica gel stationary phase can be reduced by employing a more polar solvent in the mobile phase, for example isopropanol or ethanol, as these solvents will deactivate

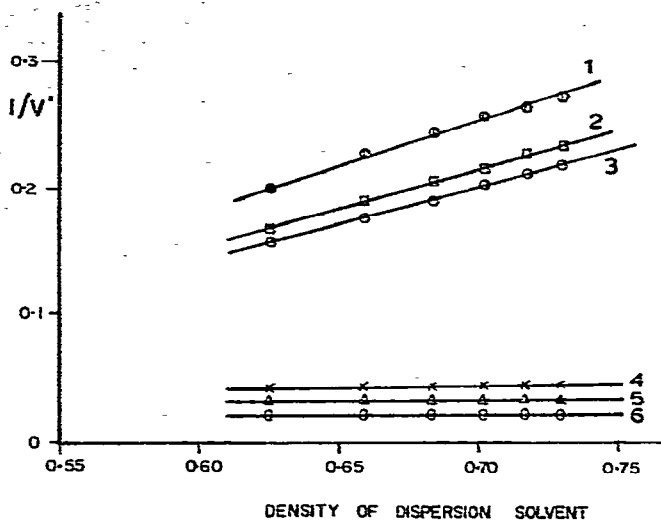


Fig. 6. Graphs of the reciprocal of the corrected retention volume against the density of the dispersion solvent. 1 = 2-Ethyl anthraquinone; 2 = 2-methyl anthraquinone; 3 = anthraquinone; 4 = phenyl methyl carbinol; 5 = benzyl alcohol; 6 = 3-phenyl-1-propanol.

the silica gel significantly more than ethyl acetate. However, this will also increase the polar interactions in the mobile phase. The polar interactions in the mobile phase, however, can be reduced by employing low concentrations of polar solvent, as has already been shown. In Fig. 7 curves displaying the effect of the density of dispersion solvent on solute retention are shown, where low concentration of ethanol and propanol are employed to reduce the level of polar interactions. Curves for mobile phases containing ethyl acetate as the polar solvent are included for reference purposes. It is seen that when the polar interactions are reduced all three aromatic alcohol solutes exhibit dispersive interactions and their retention is significantly affected by the density of the dispersion solvent. This again emphasizes that in liquid-solid chromatography polar interactions of the solute with the two phases must be relatively weak if dispersive interactions are to be exploited in separation procedures.

At this stage it should be pointed out that a specific value for K or the corrected retention volume V' can be realized in many ways. Assume that a given solute has a distribution coefficient of unity for a given chromatographic system. From eqn. 1 it is seen that for such a case the numerator and the denominator must take the same value, but this can be large, intermediate or small, depending on the nature of the interactions. Thus for two solutes eluting at the same k' one may have strong interactions with the two phases and the other weak interactions with both phases but, because the ratio of the forces in each phase is the same for each solute, they are both eluted at the same k' value. It follows that in order to exploit dispersive interactions it is not sufficient for a solute to be eluted at a low V' or k' value but that such low values of V' and k' result from relatively small polar interactive forces between the solute and the two phases. If low retention characteristics of a solute result from strong but relatively equivalent polar interactions with the two phases, then changes

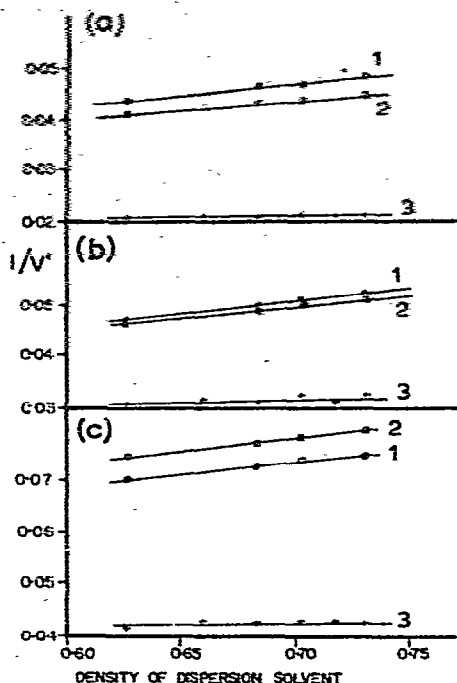


Fig. 7. Graph of the reciprocal of the corrected retention volume against the density of the solvent. Solutes: (a) 1-phenyl-3-propanol; (b) benzyl alcohol; (c) phenyl methyl carbinol. Solvents: (1) 0.8% w/v ethanol; (2) 1.0% w/v isopropanol; (3) 4.5% w/v ethyl acetate.

in the magnitude of any dispersive interactions may not change the retention of the solute significantly.

In the development of the dynamic equation for the distribution coefficient it was assumed that the different forms of molecular interactions, *viz.*, ionic, polar and dispersive, were independent and unrelated to each other. For this reason the individual interactions were made additive and not a function of one another. Thus in eqn. 6 the constant polar interactions represented in the equation by the constant A are added to the dispersive interactions represented by the product Bd . The validity of this assumption can also be tested. It has been shown that if a series of solutes are chromatographed in mobile phases containing a constant concentration of polar solvent and dispersion solvents of different density², then A remains constant and the value of B can be calculated from the curves relating $1/V'$ with d . If the experiment is repeated but at a different concentration of polar solvent, then the value of A will be changed and, provided the additive nature of the different interactions is valid, the same value for the slope B should be obtained for any given solute. In Fig. 8, curves relating $1/V'$ and the density of the dispersion solvent are shown for four different solvents chromatographed at two different concentrations of ethyl acetate. The change in ethyl acetate concentration resulted in a change in k' of approximately 50% between respective solutes in the two sets of curves. It is seen that linear correlation is obtained and the slopes of the curves (the value of the linearity constant B) are given in Table I. It is also seen from Table I that, within the confines of the experiment, the

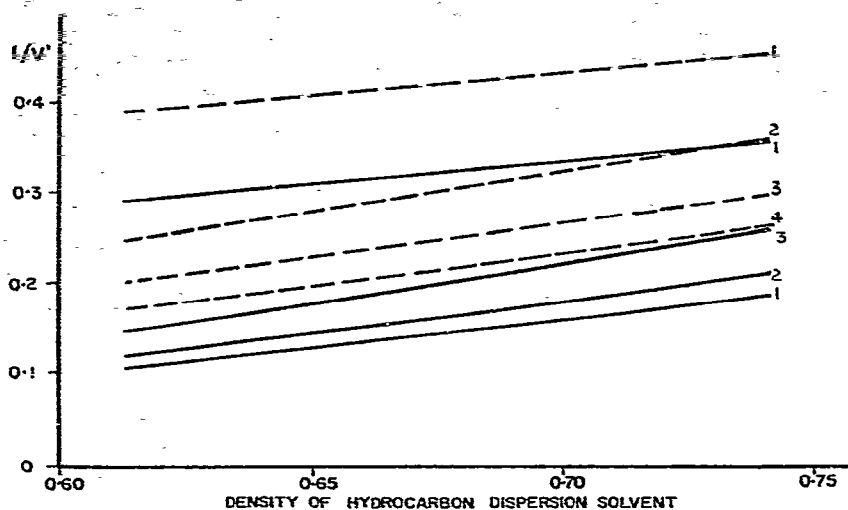


Fig. 8. Graphs of the reciprocal of the corrected retention volume against the density of the dispersion solvent at two different concentrations of ethyl acetate. Solvents: (1) 2-naphthyl benzoate; (2) 2-ethyl anthraquinone; (3) 2-methyl anthraquinone; (4) anthraquinone. Concentrations of ethyl acetate in heptane: ---, 7% w/v; —, 4.5% w/v.

values for the slope of the curves for each respective solute remain constant and independent of the concentration of the polar solvent ethyl acetate. This result confirms the assumption that individual polar and dispersive interactions are additive with respect to their total effect on the distribution coefficient or the retention of a given solute.

TABLE I

SLOPES OF CURVES RELATING $1/V'$ AND DENSITY OF DISPERSION SOLVENTS FOR DIFFERENT SOLVENTS EMPLOYING TWO DIFFERENT CONCENTRATIONS OF ETHYL ACETATE IN THE MOBILE PHASE

Polar solvent concentration	Solute	$d(1/V')/d(d)$
4.51% w/v of ethyl acetate	2-Naphthyl benzoate	0.47
	2-Ethyl anthraquinone	0.87
	2-Methyl anthraquinone	0.70
	Anthraquinone	0.62
7.00 w/v of ethyl acetate	2-Naphthyl benzoate	0.45
	2-Ethyl anthraquinone	0.90
	2-Methyl anthraquinone	0.71
	Anthraquinone	0.69

Magnitude of polar interaction

The magnitude of the polar forces of interaction as opposed to the probability of interaction will be dependent on some physical property of both the solute molecule and the molecule of the phase with which it is interacting. Such properties, among others, will be incorporated in the constant B in eqn. 3. It follows that if the relation-

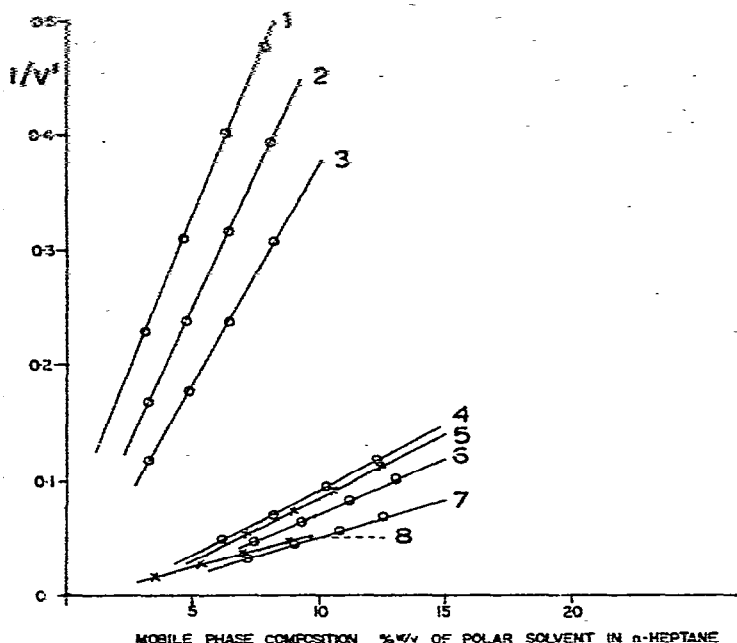


Fig. 9. Graphs relating the reciprocal of the corrected retention volume of phenyl methyl carbinol to the composition of the mobile phase containing different polar solvents in *n*-heptane. Column, 25 cm \times 4.6 mm I.D., packed with Partisil 10. Solvents: (1) isopropanol; (2) *n*-butanol; (3) *n*-pentanol; (4) dioxane; (5) tetrahydrofuran; (6) methyl acetate; (7) ethyl acetate; (8) butyl acetate.

ship between $1/V'$ and c_p is determined for a number of different polar solvents using one given solute, the physical properties of the solvent that condition the magnitude of the interacting polar forces will be reflected in the different values obtained for the slope B . In a similar manner values of B obtained by the same procedure will reflect the different properties of the solute that condition the magnitude of interaction, when a series of different solutes are employed with the same polar solvent. In Fig. 9, linear curves are shown relating $1/V'$ and c_p for different polar solvents using the same solute phenyl methyl carbinol. In Fig. 10 the logarithm of the slopes of the curves given in Fig. 9 for each respective solvent is plotted against the polarizability/ml calculated from the dielectric constants determined at the column temperature using the following equation

$$Z = \frac{e - 1}{e + 2}$$

where Z is the polarizability/ml of the substance and e is the dielectric constant of the substance.

It is seen from Fig. 10 that the slopes of the curves correlate well with the logarithm of the polarizability/ml for each respective solvent. However, it should be pointed out that the polarizability/ml for dioxane was not taken directly from the dielectric constant. The two dipoles in dioxane are in opposition and, therefore, cancel

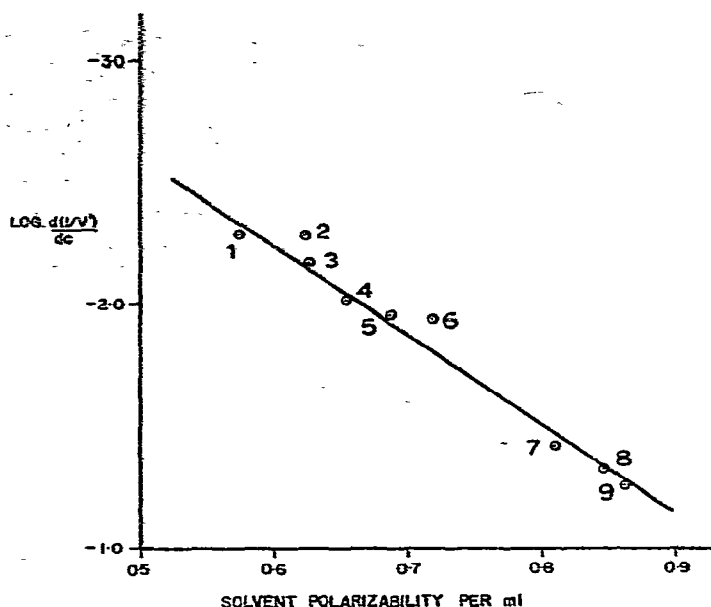


Fig. 10. Graph of $\log d(1/V)/dc_p$ against solvent polarizability per unit volume for phenyl methyl carbinol eluted from silica gel employing different solvents. Solvents: (1) Butyl acetate; (2) propyl acetate; (3) ethyl acetate; (4) methyl acetate; (5) tetrahydrofuran; (6) dioxane; (7) *n*-pentanol; (8) *n*-butanol; (9) isopropanol.

and provide dielectric constants that do not reflect the polarity of the solvent with respect to solute-solvent interaction. This is another example of the disadvantage of using bulk properties of the system to describe molecular effects. The dielectric constant of dioxane was, therefore, taken as twice that of diethyl ether. It is seen that using this value places the point for dioxane close to the regression line through the other polarizability values for the other solvents. It follows that the contribution to retention from the solvent-solute interactions would appear to be proportional to the exponent of the polarizability/ml and thus can be put in the following form

$$\text{Magnitude of solvent interaction} = A e^{\alpha Z_m} = A e^{\alpha \left(\frac{\epsilon-1}{\epsilon+2} \right)}$$

where A and α are constants and Z_m is the polarizability/ml of the solvent.

In Fig. 11 linear curves are shown relating $1/V'$ and c_p for different solutes using the same polar solvent ethyl acetate. In Fig. 12 the logarithm of the slopes of the curves given in Fig. 11 for each respective solute are plotted against the polarizability/ml for each respective solute. Thus

$$\text{The magnitude of solute interaction} = A' e^{\beta Z_s}$$

when A' and β are constant and Z_s is the polarizability/ml of the solute. It should be noted, however, that in this case β is negative. This is because the solute will interact with both the polar solvent in the mobile phase and the polar groups residing on the

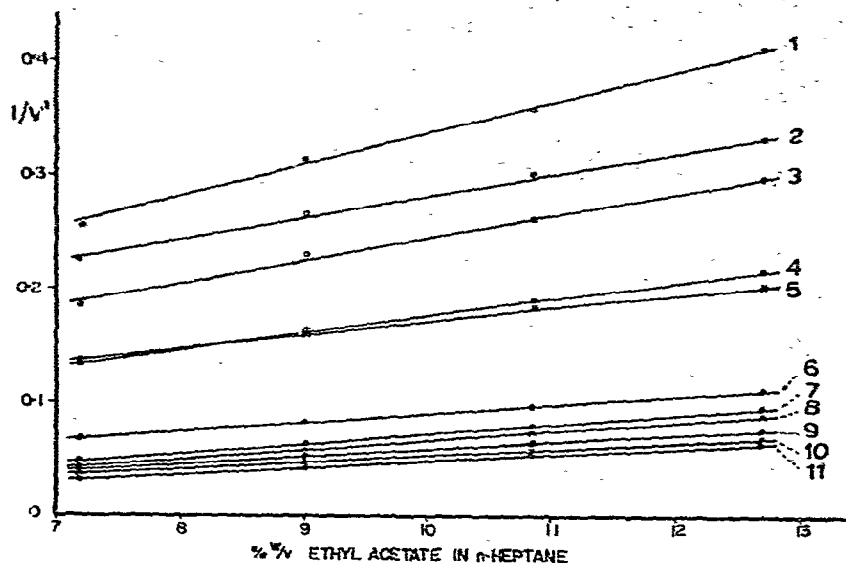


Fig. 11. Graphs relating the reciprocal of the corrected retention volume to the % w/v of ethyl acetate in *n*-heptane for different solutes. Solutes: (1) benzyl acetate; (2) methyl acetate; (3) acetophenone; (4) methyl ethyl ketone; (5) tetrahydrofuran; (6) acetone; (7) *n*-octanol; (8) phenyl ethanol; (9) *n*-pentanol; (10) benzyl alcohol; (11) *n*-butanol.

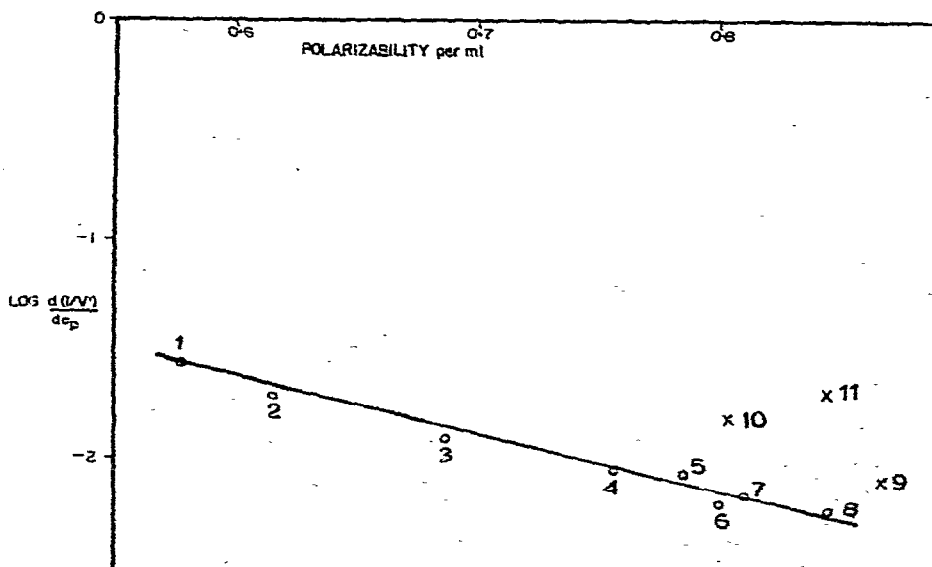


Fig. 12. Graph of $\log d(1/V')/dc_p$ against solute polarizability per unit volume. Solutes: (1) benzyl acetate; (2) methyl acetate; (3) tetrahydrofuran; (4) *n*-octanol; (5) phenyl ethanol; (6) benzyl alcohol; (7) *n*-pentanol; (8) *n*-butanol; (9) acetone; (10) methyl ethyl ketone; (11) acetophenone.

silica gel, whether or not they are adsorbed polar solvent or free silanol groups. It follows that the constant β includes a ratio of exponents of the form

$$\frac{A' e^{\gamma z_s}}{e^{\delta z_s}}$$

when $e^{\gamma z_s}$ represents the magnitude of the forces between solute and mobile phase polar molecules and $e^{\delta z_s}$ represents the magnitude of the forces between the solute and the polar moieties on the silica gel surface. It follows that

$$\frac{A' e^{\gamma z_s}}{e^{\delta z_s}} = A' e^{\beta z_s}$$

and thus

$$\beta = \gamma - \delta$$

Now, if $\delta > \gamma$, β will be negative, as shown experimentally in Fig. 11.

It should be noted that although good correlation is obtained for the solutes containing ester, ether and alcohol groups, the solutes with ketonic moieties fall a significant distance from the regression curve for the other solutes. The cause of the anomalous behavior of the ketones is uncertain but it is likely that as the ketones are liable to enolize on the acidic surface of the silica gel this enolization could be conditioned by the concentration of ethyl acetate in the mobile phase. From the curve in Fig. 12, it is seen that the chromatographic behavior of the ketones appears to lie between that of the ethers and alcohols. This would be expected but the polarizability/ml, which is derived from the dielectric constant, indicated that they should behave more like alcohols. Therefore, an alternative explanation for the anomalous behavior of the ketone solutes would be that in the measurement of the dielectric constant and as a result of the applied electric field, the values obtained are reflecting the enolic properties of the ketone as opposed to the ketonic properties. It should also be pointed out that the correlation between the magnitude of polar interaction and the polarizability/ml of the polar substances concerned has only been demonstrated for molecules with one functional group. Simple dielectric constant measurement may not be suitable for determining the effective polarizability/ml of substances where more than one functional group is present. It may be necessary for poly-functional group substances to determine the effective polarizability/ml by summing various increments allotted to each functional group present in much the same way as the Parachor of a substance is calculated.

Magnitude of dispersive interactions

The property of the solute or solvent that conditions the magnitude of dispersive forces during interaction is difficult to determine from the data presently available. From the data of Laub and Purnell⁸, the dispersive interactions are shown to be exponentially related to the molecular weight for a series of *n*-paraffins chromatographed using squalane as the stationary phase. This relationship was also shown to

be true for the homologous series of alkyl benzenes with the same stationary phase although the slope of the curve differs from that of the *n*-paraffins. In the curves relating $1/V'$ to the density of the dispersion solvent shown in Figs. 6-8 it can be shown that if the slopes of the curves for each member of a respective homologous series are plotted against the molecular weights, closely linear correlations are obtained. However, as each of the derivative curves contains only three points, the evidence is not convincing. Furthermore, for a homologous series the molecular weight is linearly related to many other physical properties of each member of the series and so the apparent significance of molecular weight in dispersive interactions could well be misleading. From what data is presently available, the magnitude of the dispersive interacting forces (F) between two molecules of molecular weights M_1 and M_2 , respectively, could be given by the equation:

$$F = C e^{\psi M_1 + \sigma M_2}$$

where C , ψ and σ are constants.

However, at the present stage of development of the dynamic equation for the distribution coefficient, it is at this time appropriate to employ an arbitrary constant to describe the magnitude of the dispersion forces until such time that further experimental evidence suggests a more explicit function.

CONCLUSION

The general form of the dynamic equation for the distribution coefficient has been substantiated but more work is necessary to determine the precise nature of the individual functions that describe the different interactions.

There is also a strong indication that under the defined conditions given for the equation, the solute and solvent polar interactions are a function of the polarizability/ml of the respective solute and solvent. For liquid-solid chromatography, if the polarizability/ml of solute, polar solvent and the interactive moieties of the silica gel (probably simple hydroxyl groups³) are Z_s , Z_m and Z_o , respectively, then polar interactions in the mobile phase have been shown to be exponential functions of Z_s and Z_m . At this stage it can be reasonably assumed that the polar interactions with the silica gel will also be an exponential function of Z_o .

A possible expansion of eqn. 2 in the following form can be suggested for isothermal conditions

$$\frac{1}{V'} = \left(\frac{A e^{\alpha Z_m + \beta Z_s} c_p + B d}{(A e^{\gamma Z_o - \psi Z_m + \delta Z_s} + D)} \right) A_s$$

Irrespective of the complexity and the arbitrary nature of the constants in the above equation, the results discussed in this paper provide a number of valuable aids to the practicing chromatographer. They are as follows:

(1) In any liquid-solid chromatographic system utilizing silica gel as the stationary phase and a mobile phase consisting of a binary mixture of a dispersive and a polar solvent, the latter being in excess of 3% w/v, the retention of all solutes can

be predicted for any concentration of polar solvent from the retention of those solutes obtained at two known concentrations of polar solvent.

(2) In thin-layer chromatography, utilizing a binary mixture of a dispersive and polar solvent in excess of 3% w/v, the R_F factor of any solute can be predicted for any concentration of polar solvent from the R_F factor for those solutes obtained at two known concentrations of polar solvent. This rule does not apply to fast moving solutes chromatographed at low concentrations of polar solvent.

(3) In ion-exchange chromatography carried out at constant pH the retention of all solutes can be predicted at all buffer concentrations from retention data obtained for the solutes at two known buffer concentrations.

(4) In gas chromatography under isothermal conditions utilizing a binary mixture as a stationary phase, the retention of any solute can be predicted from retention data of the solutes on the individual stationary phases and the volume fraction of the two phases as described by Laub and Purnell⁸.

(5) For monofunctional polar solvents and solutes, the interactive forces are proportional to the exponent of the polarizability/ml of the solute or solvent as calculated from the dielectric constant.

(6) Where polar interactions are weak, dispersive interactions can be exploited to modify solute retention and under such circumstances it has been shown that the dispersive interactions are linearly related to the density of the dispersing solvent.

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