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ION-EXCHANGE–DESOLVATION MECHANISM ON OCTADECYL SILICA USING ANIONIC HYDROPHOBIC PAIRING IONS

C. T. HUNG and R. B. TAYLOR*
School of Pharmacy, Robert Gordon's Institute of Technology, Schoolhill, Aberdeen AB9 1FR (Great Britain)

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SUMMARY

The adsorption isotherms for several anionic hydrophobic pairing ions in water on ODS Hypersil are reported. The previously proposed retention model involving ion exchange and desolvation is assessed for various ionised basic solutes by comparing experimental capacity factors with those calculated on the basis of the proposed model as a function of pairing-ion concentration. Relative ion-exchange constants are calculated and shown to be constant for all pairing ions and solutes. Addition of acetonitrile as organic modifier in aqueous solution is shown to alter both the loading and the isotherm shape for both sodium laurylsulphate and cetrimide. Using appropriate solutes, it is shown that the ion-exchange–desolvation model applies quantitatively to the mixed solvents studied.

INTRODUCTION

Although successful chromatographic separations of ionogenic species have been achieved since the inception of paired-ion liquid chromatography, the retention and separation mechanisms involved remain unclear. The variety of nomenclature applied to the technique[1] is indicative of this uncertainty.

Previous attempts to explain the mechanism of paired-ion chromatography using hydrophobic pairing ions have been on the basis of ion pairs formed on the C_{18} surface[2,3] or in the eluent[4,5]. The equation relating eluent pairing-ion concentration to experimentally observed capacity factors do not predict the parabolic behaviour experimentally observed by various workers[2,6,7]. Neither do these approaches consider the adsorption of pairing ion on to the C_{18} surface other than in competition with the solute species[2,3]. Increasing evidence that the pairing ion is adsorbed by the stationary phase to a considerable degree has supported an alternative retention mechanism involving ion exchange between adsorbed ionised pairing ions and ionised solutes[8–11]. Quantitative treatment of this model has been restricted to relating capacity factors to measured adsorbed-pairing-ion concentrations over very limited concentration ranges and for unique pairing ions[8,10,11]. Also, in this ion-exchange model no consideration is given to the hydrophobicity of the solute determining desolvation on the C_{18} surface. Other mechanisms describing the retention of ionogenic solutes have not been treated quantitatively[1].

In a recent communication a retention model incorporating ion exchange and desolvation was proposed[12] and the dependence of capacity factor on adsorbed-pairing-ion concentration was derived. Experimental data involving several different cationic pairing ions and acidic solutes were found to be in broad agreement with the proposed model over a very wide range of pairing-ion concentrations, and the parabolic nature of the dependence of capacity factor on pairing-ion concentration was demonstrated.

The present investigation extends this work in two ways. It verifies that anionic pairing ions, which are perhaps more generally used for chromatography of bases, show similar adsorption isotherms to those obtained for cationic species, and that the ion-exchange–desolvation model also accounts for the chromatographic behaviour of basic solutes on such phases. It also examines the effect of organic modifier on adsorption isotherms with subsequent chromatography. In the present work a series of variously substituted amines have been used as solutes in order to verify the general application of the model and treatment.

THEORY

The ion-exchange–desolvation model may be represented as follows. Pairing ion is adsorbed from an eluent to an extent governed by its hydrophobicity and in some cases largely independently of its eluent concentration. Solutes, oppositely charged with respect to the pairing ion, interact electrostatically with the adsorbed pairing ion. Retention of the solute by the C_{18} stationary phase is largely as a result of desolvation consequent on the neutralisation of coulombic charges. Occupation of the C_{18} surface by adsorbed pairing ions modifies the surface area available for desolvation of the solute.

This model embodies previously suggested ion-exchange ideas and also includes an attempt to allow for the conventional desolvation processes normally thought to be operative in reversed-phase liquid chromatography. An equation of the form:

$$k' = \frac{1}{V_m}(A_s - [P_n^{\mp} C^n \quad]_{org} A_p)\left\{K_1 + \left[K_2 \frac{K_{IE}[P_n^{\mp} C^{n\mp}]_{org}}{[C^{n\mp}\quad]_{aq}}\right]\right\}^{\frac{1}{n}} \qquad (1)$$

has been derived on the basis of this model for the case of cationic pairing ions.

The parameter k' is the capacity factor for a given solute and V_m the void volume of the column. The factor A_p is the area occupied by the adsorbed pairing ion per mole. K_1 and K_2 are the desolvation constants of the solute in absence and presence of pairing ion, respectively, and K_{IE} is the ion-exchange equilibrium constant. $P^{\mp}$ represents a univalent pairing ion of either charge and $C^{n}\pm$ a possibly multivalent counter-ion. A_s represents the area of the stationary phase available for desolvation in the absence of pairing ion. In the case of a monovalent counter ion, eqn. 1 may be simplified and expanded to

$$k' = \frac{1}{V_m}(A_s K_1 - K_1 [P^{\mp} C^{\mp}]_{org} A_p + \frac{K_2 K_{IE} A_s [P^{\pm} C^{\mp}]_{org}}{[C^{\mp}]_{aq}} - \frac{K_2 K_{IE} A_p [P^{\pm} C^{\mp}]_{org}^2}{[C^{\mp}]_{aq}} \qquad (2)$$

Agreement between the parabolic behaviour of acid solutes using cationic pairing ions predicted by an equation of this form and experiment has already been established[12]. In the present work it is intended to show that the agreement extends to widely differing basic solutes with different anionic pairing ions.

EXPERIMENTAL

Apparatus and materials

The apparatus used has been described previously[12]. Columns for isotherm measurements were 250 × 5 mm I.D. and those used for the determination of capacity factors 50 × 5 mm I.D.

1-Pentanesulphonic acid (PANS) and 1-octanesulphonic acid (OCTS) were obtained as their sodium salts from Eastman-Kodak (Rochester, NY, U.S.A.). Sodium laurylsulphate (SLS), aniline hydrochloride (AN) and benzylamine (BZ) were obtained from BDH (Poole, Great Britain). Dopamine hydrochloride (DOP) was obtained from Sigma (St. Louis, MO, U.S.A.) and *dl*-10-camphorsulphonic acid (CAMS) as the sodium salt and L-adrenaline-D-hydrogentartrate (AD) from Koch-Light Labs. (Colnbrook, Great Britain). The stationary phase ODS-Hypersil, cetrimide (CTAB) and the various carboxylic acids, benzoic acid (BA), 2,4-dimethylbenzoic acid (2,4-DMA), 4-ethylbenzoic acid (4-EBA) and naphthoic acid (NA) together with the test neutral compound 4-hydroxymethyl furfuraldehyde (5HMF) were obtained as described previously. Acetonitrile was obtained from Rathburn (Walkerburn, Great Britain). Water was double distilled and all other reagents were of AnalaR or comparable quality.

Procedures

Adsorption isotherms for the four anionic pairing ions PANS, OCTS, CAMS and SLS on ODS-Hypersil were determined as before by adsorbing from a fixed concentration of pairing in aqueous solution, desorbing using ethanol and weighing the total amount recovered. This method was adopted after practically assessing other techniques of isotherm determination[3,9,13]. A similar procedure was adopted for isotherms measured in the presence of organic modifier.

Chromatographic measurements were carried out in a standard phosphate buffer, pH 1.5 for the cationic solutes and pH 7.4 for the anionic solutes measured on CTAB in mixed solvents. A flow-rate of 3 $cm^3 min^{-1}$ was employed throughout, and the wavelength of measurement of all solutes was at 254 nm except when CAMS was used as pairing ion. In this case 235 mm was chosen as the wavelength of minimal background absorbance.

RESULTS AND DISCUSSION

Isotherm results

Adsorption isotherms for the four anionic pairing ions used in pure water are shown in Fig. 1 over an aqueous concentration range of 2–400 mol m^{-3}. With the exception of SLS, the isotherms are L type according to the Giles classification[14] and indicate a smooth rise to a plateau level characteristic of the particular pairing ion. At low concentrations these isotherms approach linearity. SLS, on the other hand, is

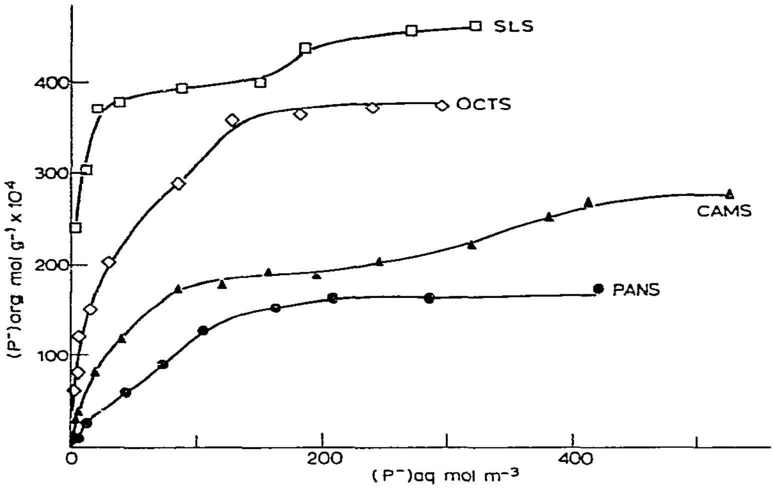

Fig. 1. Adsorption isotherms for various anionic pairing ions for the ODS Hypersil–water system. ●, PANS; ▲, CAMS; ◇, OCTS; □, SLS.

markedly H type, *i.e.* even at low aqueous concentrations very large concentrations of pairing ion are adsorbed at equilibrium. These results confirm the findings of our previous investigation in that the loading capacity of different pairing ions on ODS-Hypersil appears to be related to the hydrophobicity of the particular pairing ion. In this case, however, no attempt is made to correlate the loading capacity with any hydrophobic parameter in view of the difficulty of estimating the hydrophobicity of CAMS. The H-type behaviour of SLS observed after long equilibration times is analogous to that of the correspondingly surface-active cationic species (CTAB). Since most chromatographic separations involve the use of an organic modifier, it was thought useful to examine the effect of acetonitrile concentration on the adsorption properties of the two most highly adsorbed pairing ions used in this and the previous study, namely SLS and CTAB. The results are shown in Figs. 2a and b, respectively, for three concentrations of acetonitrile (0, 30 and 60%, v/v). For both pairing ions the isotherm is markedly altered by the addition of acetonitrile. Firstly, the column loading is decreased, an effect which has been indicated by previous workers[1,11]. In addition, the shape of the isotherm is seen to be drastically altered. The H-type isotherms observed in aqueous eluent are L type in 30% and approximate to linearity in the highest acetonitrile concentration used.

These results demonstrate the inadequacy of arbitrarily fitting adsorption data to particular isotherms. They further support the present approach of utilising directly determined adsorbed-pairing-ion concentrations when attempting to quantity chromatographic behaviour.

Chromatographic results

The variation of the capacity factors (k') of the various ionised solutes with pairing-ion concentrations both in eluent and adsorbed on to the organic stationary phase are shown in Figs 3a–h and 4a–h, respectively. Included in these figures are the results obtained for mixed eluents containing different proportions of organic mod-

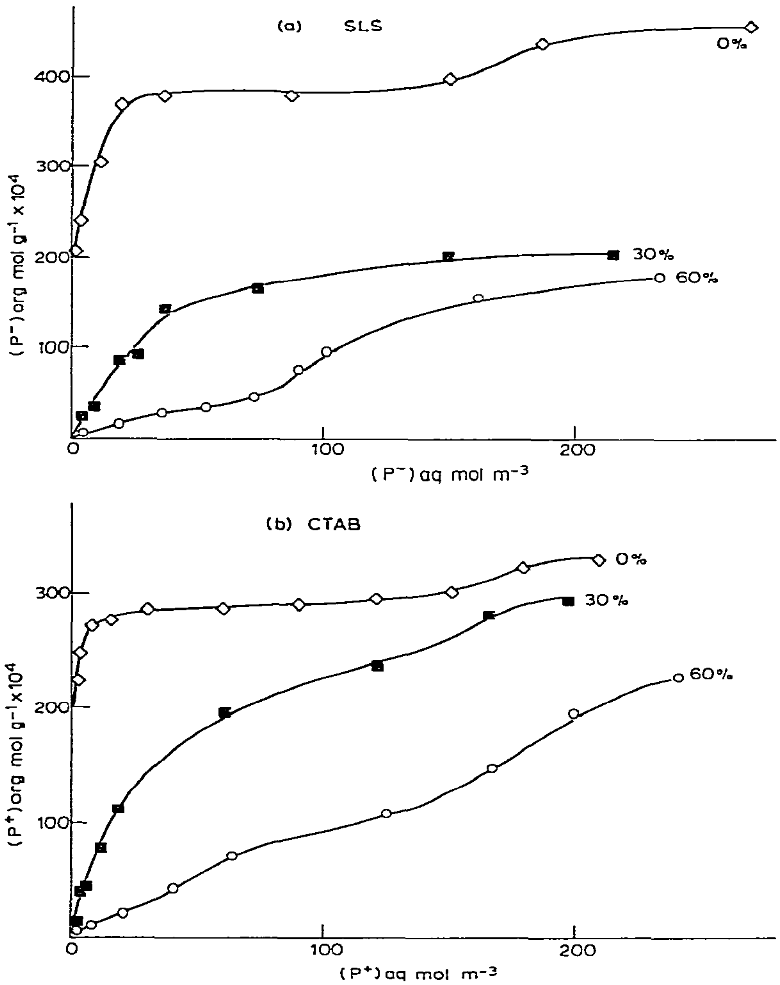

Fig. 2. Adsorption isotherms for (a) sodium laurylsulphate and (b) cetyltrimethylammonium bromide, for different concentrations of acetonitrile–water mixtures. ◇, Pure water; ■, 30% (v/v) acetonitrile; ○, 60% (v/v) acetonitrile.

ifier. Broken lines refer to the experimentally observed k' values and the solid lines represent the k' values calculated on the basis of the derived model.

As in the previous study of cationic pairing ions[12], K_1 values were determined from measurements of retention times in the absence of pairing ion. $[P_n^- C^{n+}]_{org}$ was obtained directly from the appropriate isotherm, and $[C^{n+}]_{aq}$ was estimated from aqueous pairing-ion concentration and buffer composition. The values of A_p were calculated as before from the dimensions of the individual solutes[15], and A_s was taken[16] as 142 m^2 cm^{-3}. The V_m value for the column was estimated using sodium nitrate as an unretained solute[17]. Since the K_2K_{IE} term could not be evaluated analytically, an iterative curve-fitting program (HOOKE) was employed to select the value of this composite constant which would yield the best least-squares fit of the equation with the experimental data.

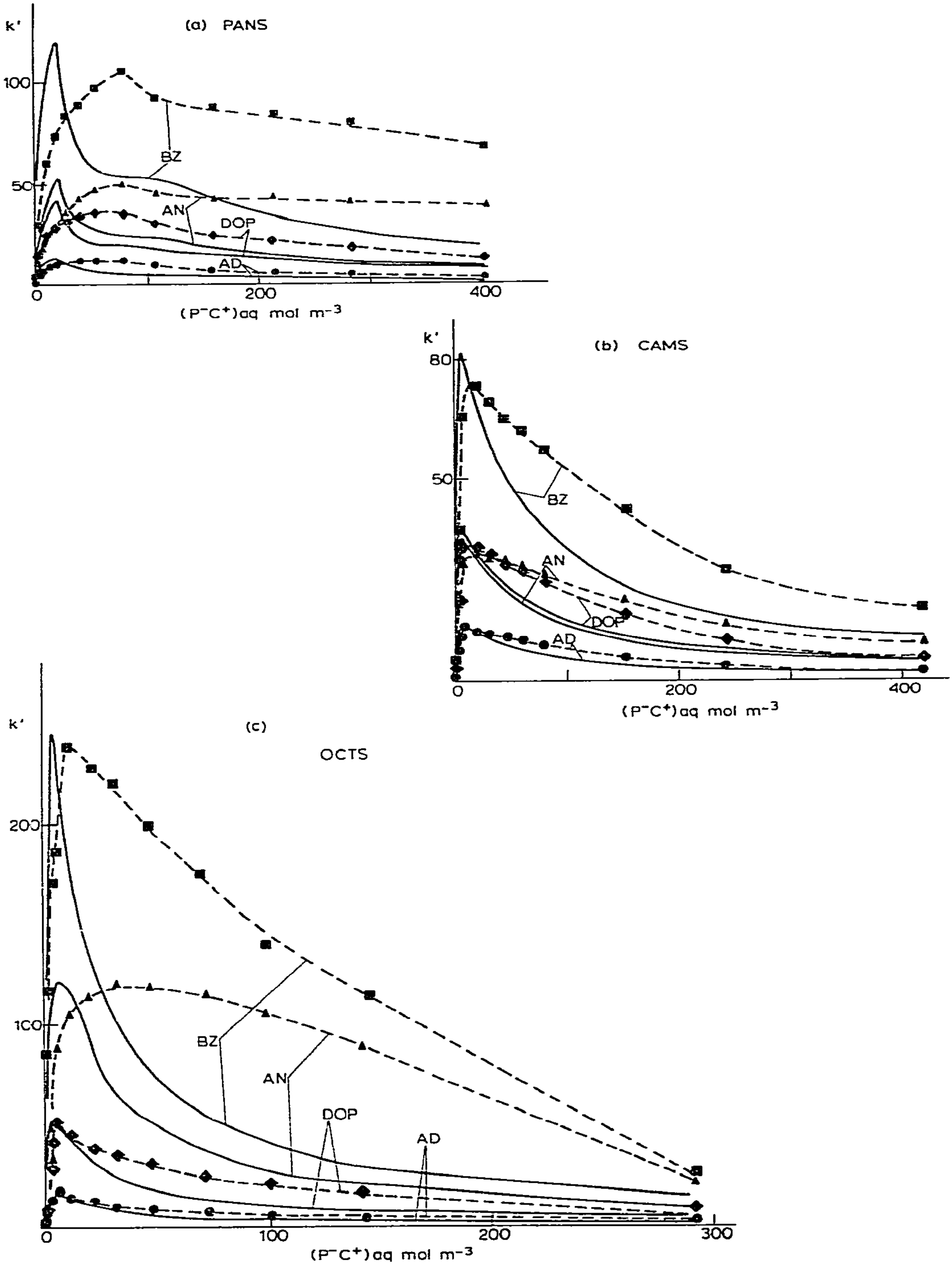
k'
(a) PANS
100
50
0
200
400
(P⁻C⁺)aq mol m⁻³
BZ
AN
DOP
AD
k'
(b) CAMS
80
50
0
200
400
(P⁻C⁺)aq mol m⁻³
BZ
AN
DOP
AD
k'
(c)
OCTS
200
100
0
100
200
300
(P⁻C⁺)aq mol m⁻³
BZ
AN
DOP
AD

Fig. 3.

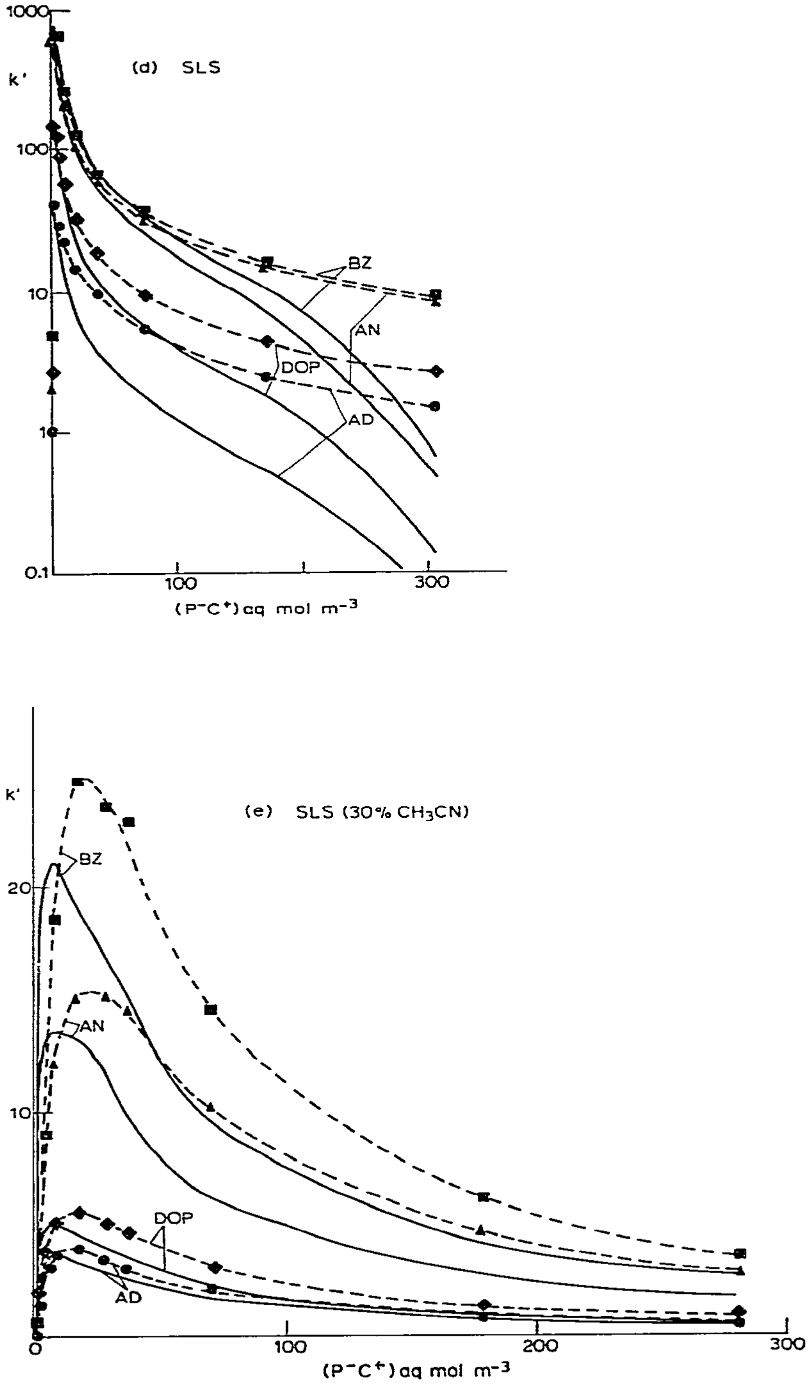

Fig. 3.

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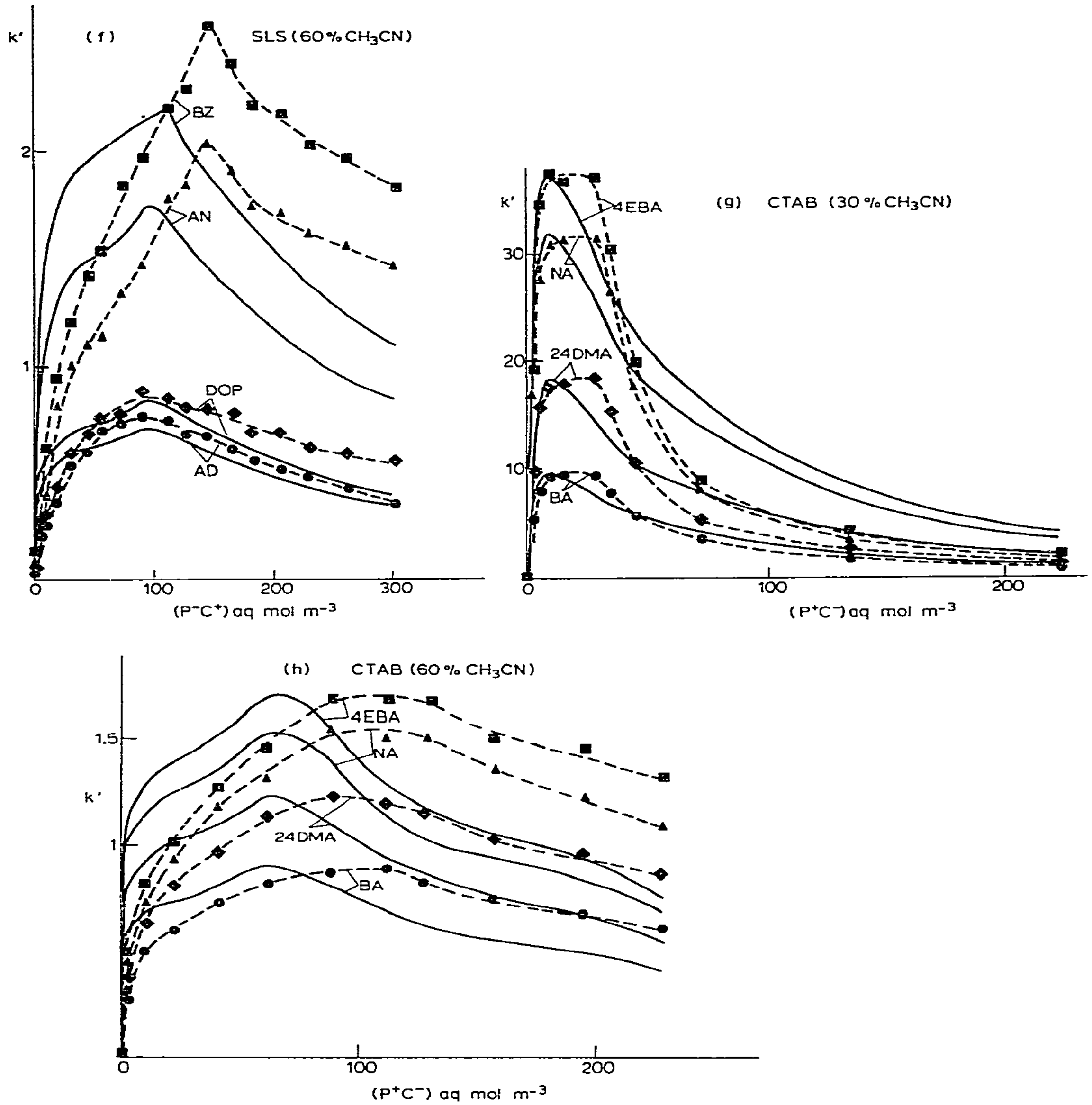

Fig. 3. Plots showing the variation of capacity factor (k') with aqueous pairing-ion concentration for the different solutes and pairing ions used. Broken lines represent experimental results. Solid lines represent calculated curves.

The experimental results clearly indicate a parabolic dependence of k' on pairing-ion concentration both in the mobile and stationary phases, and generally good agreement between the shapes of experimental and calculated curves is obtained. In the case of the surfactant pairing ion, SLS, only the decreasing part of the curve can be observed experimentally owing to the high adsorption at all concentrations exhibited for this pairing ion on the C_{18} surface, confirming the results previously obtained for the surfactant pairing ion, CTAB.

The increase of k' with aqueous pairing-ion concentration is interpreted as due to an increasing adsorption of pairing ion by the C_{18} surface, and the correlation of the magnitudes of k' for a given solute with different pairing ions supports the ion-exchange aspect of the present model. The decreasing section of the curve is accounted for in the mathematical treatment of the model by the sum of two effects, namely decreased C_{18} surface available for desolvation and increasing eluent counter-

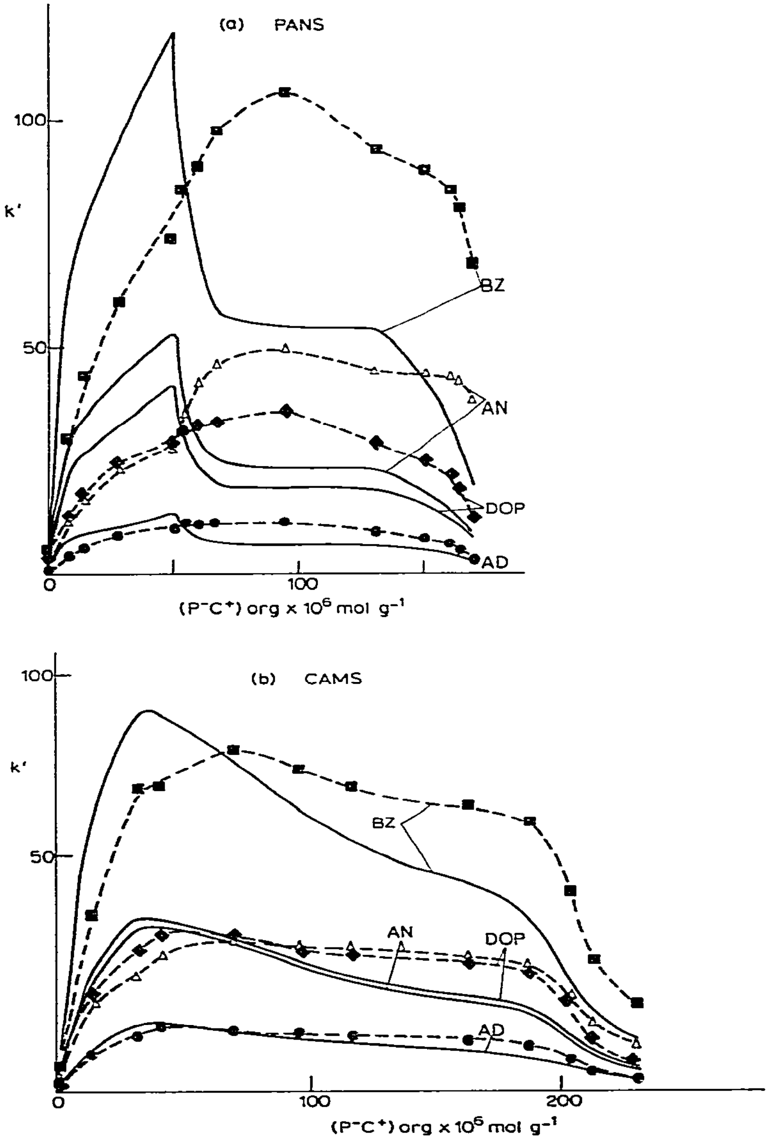

Fig. 4.

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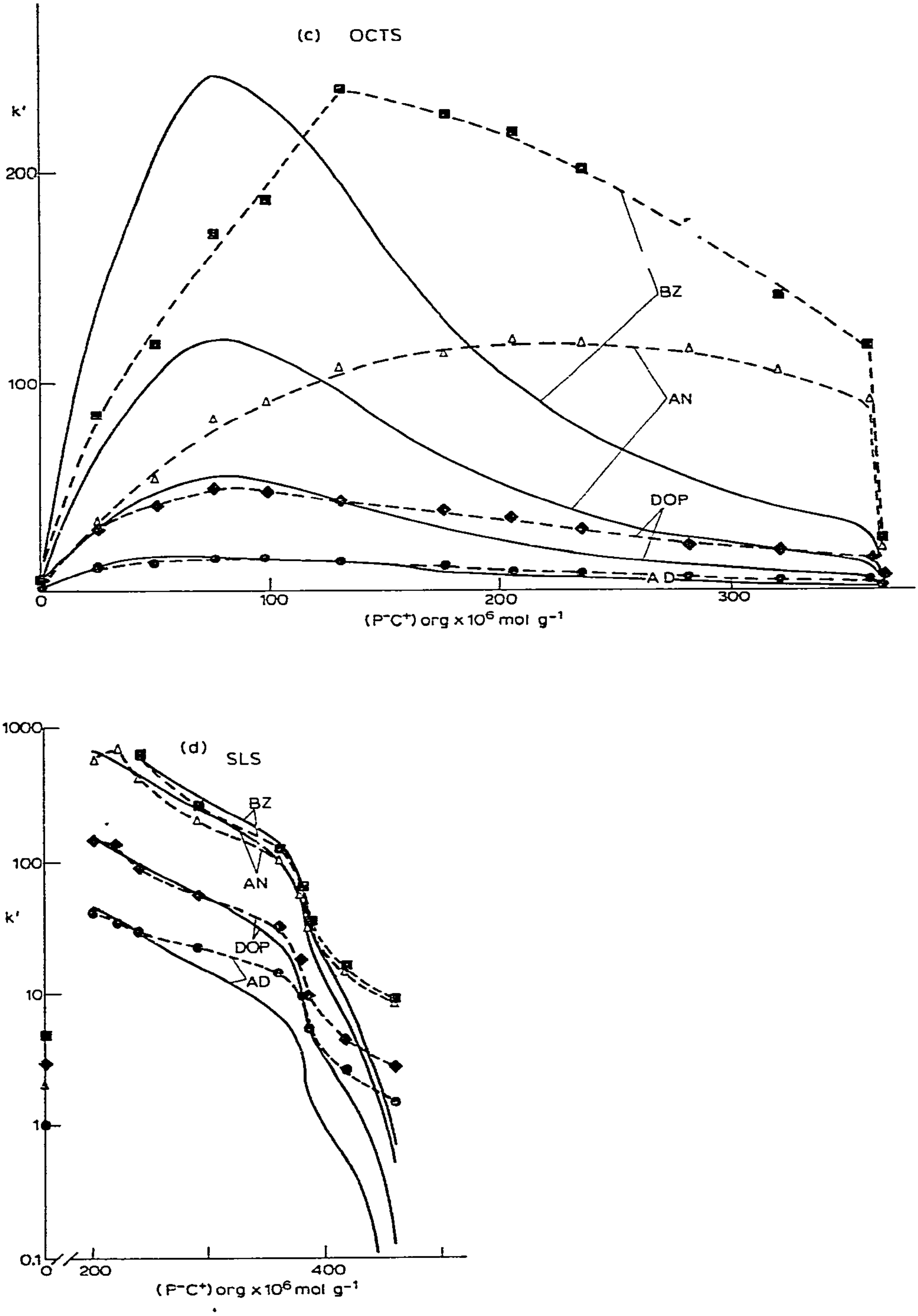
(c) OCTS
k'
200
100
0
100
200
300
BZ
AN
DOP
A D
$(P^-C^+)_{org} \times 10^6$ mol g^{-1}
(d) SLS
1000
100
10
1
0.1
k'
BZ
AN
DOP
AD
0
200
400
$(P^-C^+)_{org} \times 10^6$ mol g^{-1}

Fig. 4.

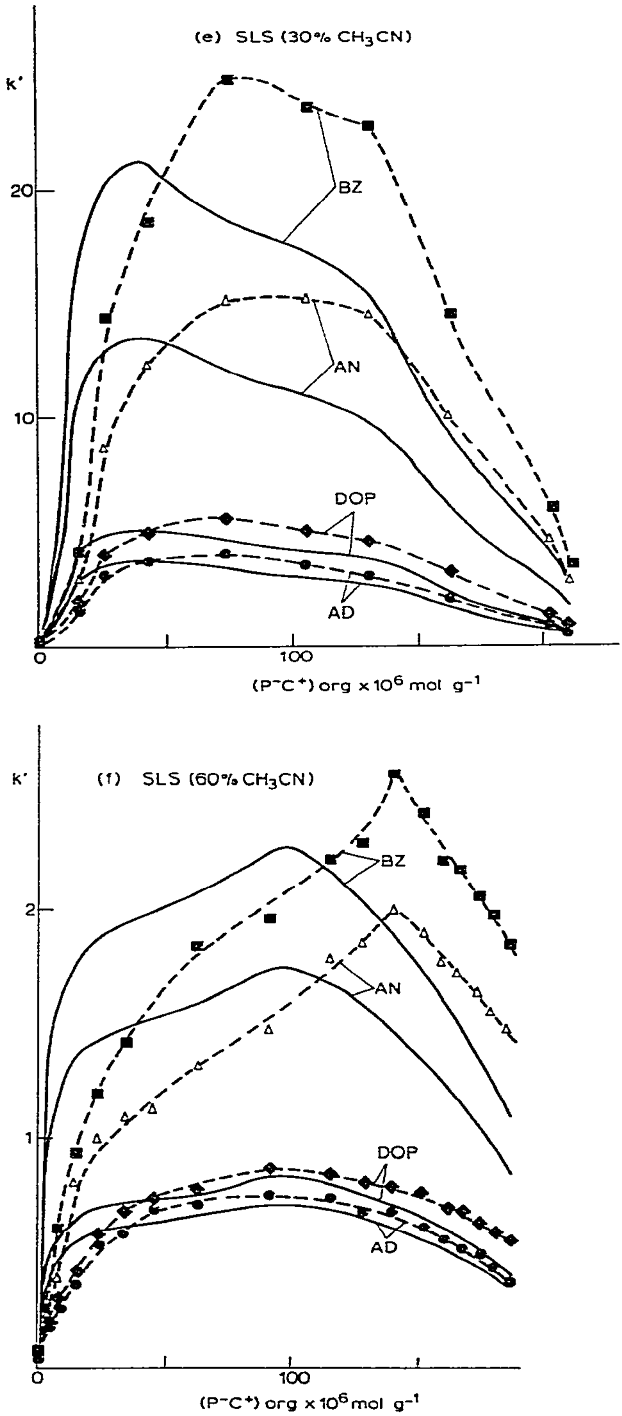

Fig. 4.

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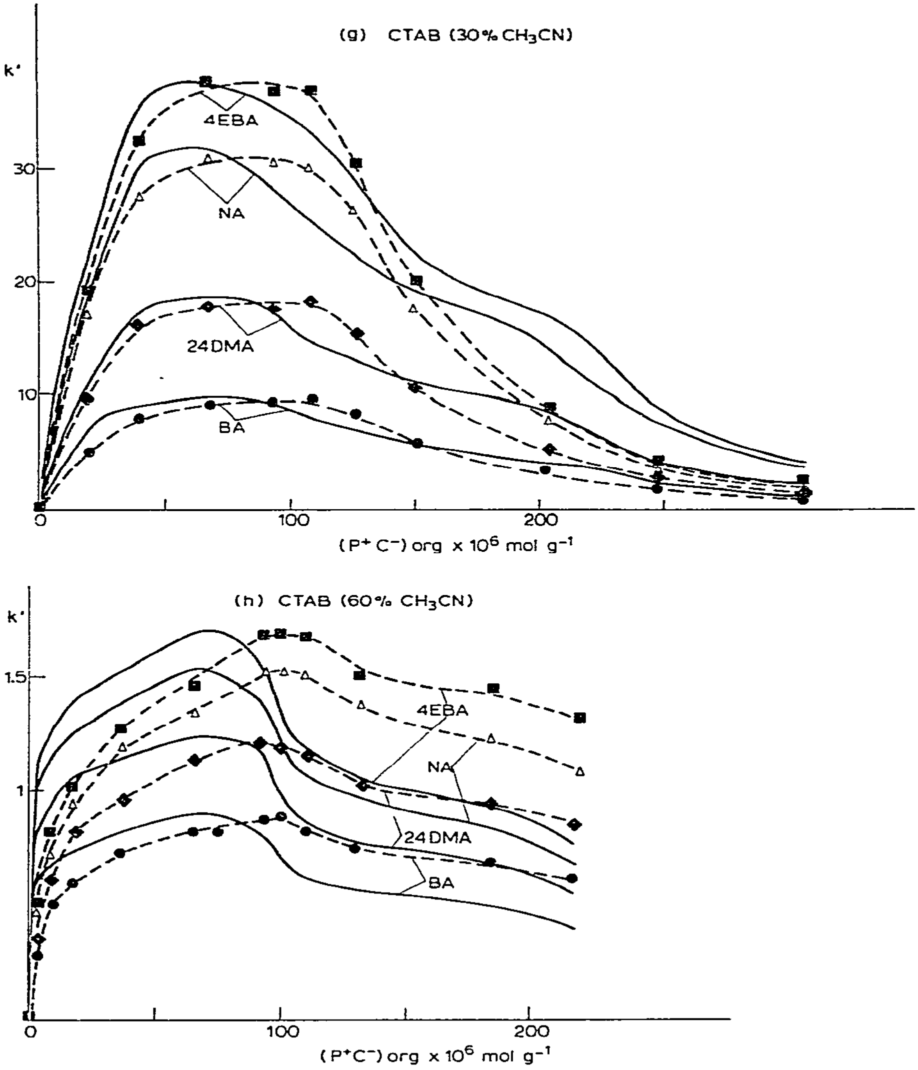

Fig. 4. Plots showing the variation of capacity factor (k') with organic-pairing-ion concentration for the different solutes used. Broken lines represent experimental results. Solid lines represent calculated curves.

ion concentration as a result of added pairing-ion salt. This latter effect is demonstrated in the case of SLS where the organic concentration of pairing ion is essentially constant, as evidenced by the plateau region of the isotherm. The effect of counter-ion in this model is similar to that of the original ion-exchange equilibrium model[11] but is more realistic in that the addition of pairing-ion salt to the eluent will firstly alter the C_{18} surface area of the phase ratio $(A_s - [P_n^{\mp} C^{n\mp}]\ A_p)/V_m$ and, if this is not possible owing to equilibrium loading of the surface having been achieved, act as a conventional counter-ion.

In our previous treatment of various carboxylic acids, the assumption was made that $K_1 = K_2$ which enabled the values of K_{IE} to be uniquely estimated. While this may be approximately true in the case of solutes which have appreciable lipophilicity in the ionised state, it will be less true in the case where the solute retention in the absence of pairing ion is very small. In this more general case, however, the assumption may be made that K_2/K_1 is constant for a given pairing ion, which allows relative values of K_{IE} to be estimated, *i.e.* K'_{IE}. This constant will still provide information on the coulombic interactions of solute and adsorbed pairing ion. This assumption appears to be justified in recent studies of the retention of carboxylic acids as a function of pH where the k' values of solutes have been measured in both non-ionised and fully ionised states[10,18]. Values of $K_2 K_{IE}$ and K_1 are shown in Table I and the calculated values of K'_{IE} involving the above assumption are shown in Table II. It can be seen from the latter results that the K'_{IE} values are remarkably constant, not only for a given pairing ion but also among different pairing ions. This lends support to the concept of a constant charge neutralising effect preceding a particular desolvation tendency which is characteristic of a given solute.

TABLE I

K_1 AND $K_2 K_{IE}$ VALUES CALCULATED TO PRODUCE THE BEST FIT OF EXPERIMENTAL DATA

Solute	$K_1 \times 10^8$	$K_2 K_{IE} \times 10^2$			
		PANS	*CAMS*	*OCTS*	*SLS*
AD	0.711	3.757	2.827	1.194	1.293
AN	1.41	15.12	7.295	8.718	20.153
DOP	1.99	11.49	7.484	3.825	4.405
BZ	3.52	34.093	16.656	17.429	27.35

TABLE II

K'_{IE} VALUES CALCULATED FOR SOLUTE-PAIRING SYSTEMS ASSUMING K_2/K_1 IS CONSTANT

Solute	$K'_{IE} \times 10^{-6}$			
	PANS	*CAMS*	*OCTS*	*SLS*
AD	5.284	3.976	1.679	1.819
AN	10.723	5.174	6.183	14.56
DOP	5.774	3.761	1.922	2.214
BZ	9.685	4.732	4.732	7.77

In the case of eluents incorporating acetonitrile, the values of k' in the absence of pairing ion are very small and for the higher concentrations of acetonitrile apparently negative[19]. For the mixed solvent measurements, the $K_2 K_{IE}$ values are shown as a function of acetonitrile concentration in Table III. As would be expected, their values are lower than in the pure aqueous solvent owing to adsorption of acetonitrile in addition to pairing ion by the surface.

TABLE III

K_2K_{IE} VALUES CALCULATED TO PRODUCE THE BEST FIT OF EXPERIMENTAL DATA IN MIXED ORGANIC/AQUEOUS SOLVENT

Solute	*$K_2K_{IE} \times 10^2$ (SLS)*		*Solute*	*$K_2K_{IE} \times 10^2$ (CTAB)*	
	30% (v/v) Acetonitrile	*60% (v/v) Acetonitrile*		*30% (v/v) Acetonitrile*	*60% (v/v) Acetonitrile*
AD	0.772	0.614	BA	1.228	0.748
AN	2.932	1.521	2,4-DMA	2.376	1.026
DOP	1.087	0.702	NA	4.139	1.270
BZ	4.582	1.947	4-EBA	4.876	1.415

As in our previous study, the behaviour of a single uncharged compound 5HMF agrees only qualitatively with the derived equation. In the absence of a K_{IE} term, eqn. 2 may be written:

$$k' = \frac{K_1}{V_m}(A_s - [P_n^{\pm}C^{n\mp}]_{org}\, A_p) \qquad (3)$$

The combined results of the behaviour of 5HMF on Hypersil ODS in the presence of an equilibrium concentration of adsorbed cationic and anionic pairing ion in aqueous solvents is shown in Fig. 5 as the variation of k' with $[P_n^{\mp}C^{n\pm}]_{org}A_p$.

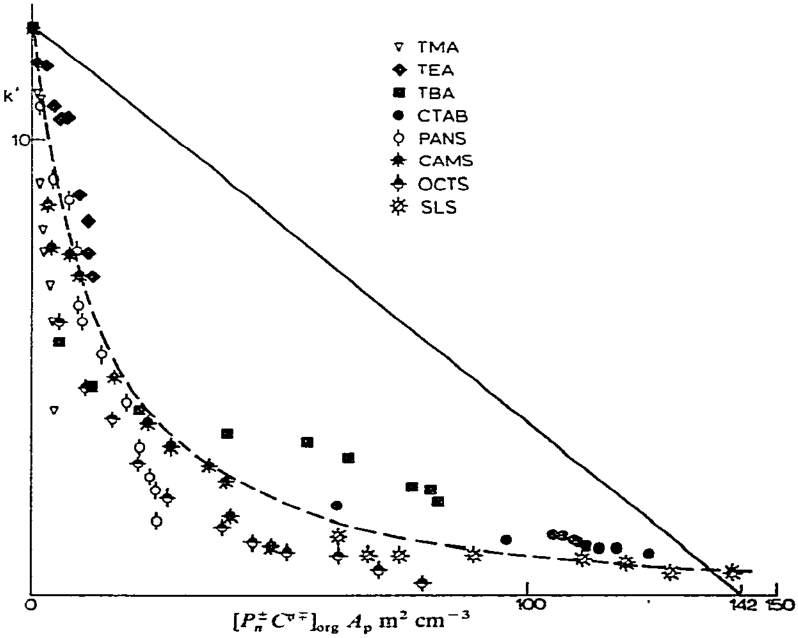

Fig. 5. Plot showing the variation of capacity factor for 5HMF as a function of the area of the C_{18} calculated as being covered by various pairing ions at the concentrations studied. Broken line represents experimental results. Solid line represents eqn. 3.

The theoretical line of this equation appears to underestimate the surface exclusion effect of the adsorbed pairing ions on a neutral species. For all pairing ions at all concentrations, there is a marked reduction of the 5HMF capacity factor, which tends to a small constant value. It is possible that the surface area aspect of the present model is quantitatively inadequate owing to our incomplete understanding of the physical nature of the C_{18} surface in the presence and absence of organic modifiers and the pairing ions[20,21]. This limitation does not invalidate the generality of the proposed model and any alteration in the A_s and A_p values used would only have the effect of altering the absolute magnitudes of the derived constants.

CONCLUSIONS

The present results for anionic pairing ions of different hydrophobicities confirm the findings of our previous study for cationic species, namely that the loading of the C_{18} phase with pairing ion in aqueous eluents is complex. This is especially true in the case of long-chain pairing ions having surfactant properties. These show high adsorption at very low eluent pairing-ion concentrations. The use of such pairing ions may produce unacceptably high k' values when used with solutes which have appreciable retention in the absence of pairing ion, unless appreciable concentrations of organic modifiers are used to change the H-type behaviour. Less strongly adsorbed pairing ions provide a more sensitive method of adjusting retention times by pairing-ion concentration.

The ion-exchange–desolvation model appears adequately to represent the behaviour of both anionic and cationic solutes in the presence of pairing ion. The magnitudes of the constants relating to the ion-exchange aspect of the proposed mechanism indicate that the electrostatic binding is approximately constant for all solutes and pairing ions. The model also predicts the chromatographic behaviour adequately in mixed eluents containing both water and organic modifier.

The behaviour of 5HMF as a fairly polar neutral solute shows a marked decrease in retention with increasing pairing-ion concentration for all pairing ions studied, which can be of use in the separation of charged and uncharged solute species by the ion-pairing technique[22]. The quantitative predictions of the model described do not adequately predict the behaviour of our neutral test substance and this, it is believed, is due to the difficulty of quantifying the phase ratio in the presence of pairing ion.

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