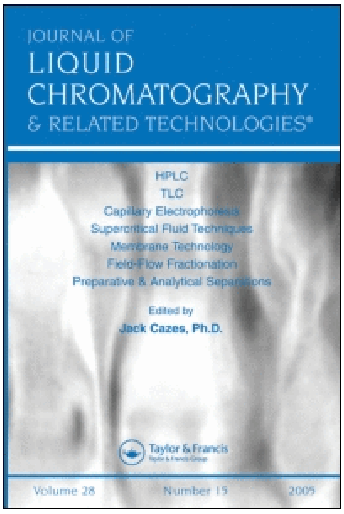

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

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To cite this Article Rey, M. E. Del and Vera-avila, L. E.(1988) 'A Simple Two-Membered Model for Retention in RP-IPC with Hydrophobic Counterions', Journal of Liquid Chromatography & Related Technologies, 11: 14, 2885 — 2905
To link to this Article: DOI: 10.1080/01483918808076767
URL: http://dx.doi.org/10.1080/01483918808076767

JOURNAL OF LIQUID CHROMATOGRAPHY, 11(14),2885-2905 (1988)

A SIMPLE TWO-MEMBERED MODEL FOR RETENTION IN RP-IPC WITH HYDROPHOBIC COUNTERIONS

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ABSTRACT

The retention behavior of some organic anions in the presence of tetralkyl ammonium ions in the mobile phase is analized under conditions where these hydrophobic counterions do adsorb on the surface of a reversed-phase packing. The study comprises the effects of the counterion concentration in the mobile and in the stationary phase, the salt concentration in the eluent and the salt type. Experimental results are discussed in view of previously proposed mechanisms and a simple two-member expression relating corrected capacity factor to adsorbed counterion concentration is deduced. The retention model indicates that solute is present in the stationary phase at two different levels; first into a diffuse ionic cloud associated electrically to the ionic surface and, second, at the surface of the packing where it forms ion pairs with the adsorbed counterions.

INTRODUCTION

Ion pair chromatography (IPC) with non polar stationary phases has had, since the beginning of its development, a great success, displacing the polymeric ion exchange resins in some of their typical applications. The numerous studies dealing with solute retention in IPC have permitted to find out the most influent parameters to adjust retention times and selectivities for the best separation of ionic compounds.

Actually it is well known that retention in IPC depends mainly on the following variables:

Counterion concentration in the mobile phase.
Hydrocarbon surface of counterion and solute molecules.
Nature and concentration of salts in the eluent.
Concentration of organic modifier in the mobile phase.
pH (if solutes and/or counterions are weak acids or weak bases).
Carbon content of the stationary phase.

In order to predict and quantify the effect of changes in these variables on separation, investigators in the field have proposed several retention models. The first proposals were based on a mechanism of ion pair formation in the mobile phase or at the interface (1-6) or on a dynamic ion exchange process (7-10). Other models, resulting from different combinations of both types of mechanisms, appeared some time later and the relationship between the capacity factor and the mobile phase composition parameters became more complicated (12, 15-17). Nevertheless, none of these models adjust perfectly to all the results obtained using a great variety of experimental conditions.

The problem seems to be more complex when the counterions dissolved in the mobile phase have an hydrophobic character and do adsorb on the surface of the stationary phase. It has been demonstrated that this adsorption depends on the mobile phase composition and specially the organic modifier concentration and the counterion concentration and hydrophobicity (9-14, 18). As we mentionned before, these variables affect also solute retention.

In this work we have studied the retention of some aromatic acids in their anionic form using tetralkyl ammonium ions as counterions. In the conditions employed these hydrophobic ions do adsorb on the stationary phase (18). We hope that the conclusions derived from our results will contribute to a better understanding of retention in these systems.

EXPERIMENTAL

Instrumentation and Chromatographic Procedures.

The liquid chromatograph was a Perkin-Elmer 3B series pump system equipped with a model 7105 Rheodyne injector, a LC-75 variable wavelenght Perkin-Elmer UV-Vis detector, a LC-100 Perkin-Elmer oven and a Perkin-Elmer 023 recorder. In all the experiments the column temperature was fixed at 40·C. All the mobile phases employed were degassed before use by means of an ultrasonic bath (Branson Instruments model D-50).

Empty columns of Lichroma tubing (15cm x 4.6mm i.d.) purchased from Alltech were homepacked with a 5 μ C-18 irregular shaped packing, Supelcosil LC-7 (13.6% carbon content from elementary analysis and 250 m^2/g, supplier data). These columns were employed in all the experiments except the ones for the study of the effect of different salts on solute retention where we used columns packed with 5 μ Rsil C-18 HL from Alltech (16% carbon content, supplier data).

Before each experiment the column was washed with 40 mL of pure methanol in order to remove all the counterion adsorbed on the stationary phase remaining from previous experiments. Then, a mobile phase containing all components (water, acetonitrile and salts), except the tetralkyl ammonium ion, was passed through the column to pre-equilibrate it and a $NaNO_3$ solution was injected to measure the void volume of the chromatographic system. At this stage the test solutes were also injected and their capacity factors, k_0, were evaluated. Finally the column was equilibrated with the eluent containing the counterion and the solutes were injected at least twice; their capacity factors in the presence of the tetralkyl ammonium ion, k', were then calculated.

All the data concerning the adsorbed counterion concentration were taken from a previous work (18).

Standard and Reagents.

The bromide salts of the counterions tetrabutyl ammonium (TBA) and tetrapropyl ammonium (TPA) were purchased from Eastman

Chemicals. The test solutes were the organic acids p.hydroxybenzoic, sorbic, o.toluic, and p.toluic, all from Eastman Chemicals Other salts added to the mobile phase, sodium sulphate (Baker), mono and disodium phosphates (Merck), citric acid (Merck), crotonic acid (Eastman Chemicals) and ammonium acetate (Merck), were analytical grade reagents.

Analytical grade acetonitrile and methanol were from Merck. Water was bidistilled and degassed thoroughly by vacuum. Sodium hydroxide, used to adjust the pH, was from Reactivos Químicos Monterrey.

Concentrated aqueous solutions of the tetralkyl ammonium bromides and of the other salts were prepared in volumetric flasks, adjusting if necessary the pH to 7. The ammonium acetate solution was titrated using the formaldehyde method (19).

Mobile phases were prepared in volumetric flasks just before use, taking the appropriate aliquots of the concentrated solutions mentionned before and adding the required volume of acetonitrile and water.

Solutes were dissolved in a water-acetonitrile 90:10 v/v mixture and pH was adjusted to 7. Their concentration was 10^{-2} M except for p.toluate which was 10^{-3} M. In all the experiments a 5 microliter volume of each solute was injected.

RESULTS

Effect of Counterion Concentration.

TABLES 1 and 2 summarize the capacity factors of the 4 test solutes deduced from the experimental retention times at different concentrations of the counterions TPA or TBA in the mobile phase. The concentration of adsorbed counterion in the stationary phase is also marked.

The relation between retention and counterion concentration in the mobile or in the stationary phase is better appreciated in Figures 1 and 2. For all the studied solutes there is a linear increment in retention with the adsorbed tetralkyl ammonium concentration. This relation can be represented by equation (1).

TABLE 1

Effect of TPA Concentration on Solute Retention.

Conditions: column (15cm x 4.6mm i.d.) packed with 5 μ Supelcosil LC-7; mobile phase acetonitrile-water (pH=7) 10:90 v/v containing TPA and 0.31 M ammonium acetate. Temp. 40°C.

$[TPA]_m x10^2$ (mmol/mL)	$[TPA]_{st} x10^2$ (mmol/g)	solutes (K') 1*	2*	3*	4*
0.00	0.00	0.05#	0.69#	0.58#	1.35#
0.01	0.35	0.03	0.69	0.58	1.35
0.10	1.11	0.07	0.79	0.69	1.65
0.40	2.89	0.09	0.88	0.79	1.88
1.00	5.10	0.14	1.11	1.02	2.58
4.00	9.70	0.26	1.43	1.42	3.31

* 1 = p.hydroxybenzoate 2 = sorbate
3 = o.toluate 4 = p.toluate
\# experimental K_o values

TABLE 2

Effect of TBA Concentration on Solute Retention.

Conditions: column (15cm x 4.6mm i.d.) packed with 5 μ Supelcosil LC-7; mobile phase acetonitrile-water (pH=7) 10:90 v/v containing TBA and 0.25 M ammonium acetate. Temp. 40°C.

$[TBA]_m x10^2$ (mmol/mL)	$[TBA]_{st} x10^2$ (mmol/g)	solutes (K') 1*	2*	3*	4*
0.00	0.00	0.05#	0.69#	0.58#	1.35#
0.01	2.07	0.14	0.84	0.73	1.65
0.04	3.70	0.19	0.99	0.90	2.13
0.06	4.15	0.23	1.06	0.97	2.25
0.10	6.47	0.28	1.46	1.39	3.30
0.20	8.11	0.34	1.73	1.74	4.04
0.40	9.63	0.44	1.92	1.97	4.75

* solutes as in TABLE 1. # experimental K_o values.

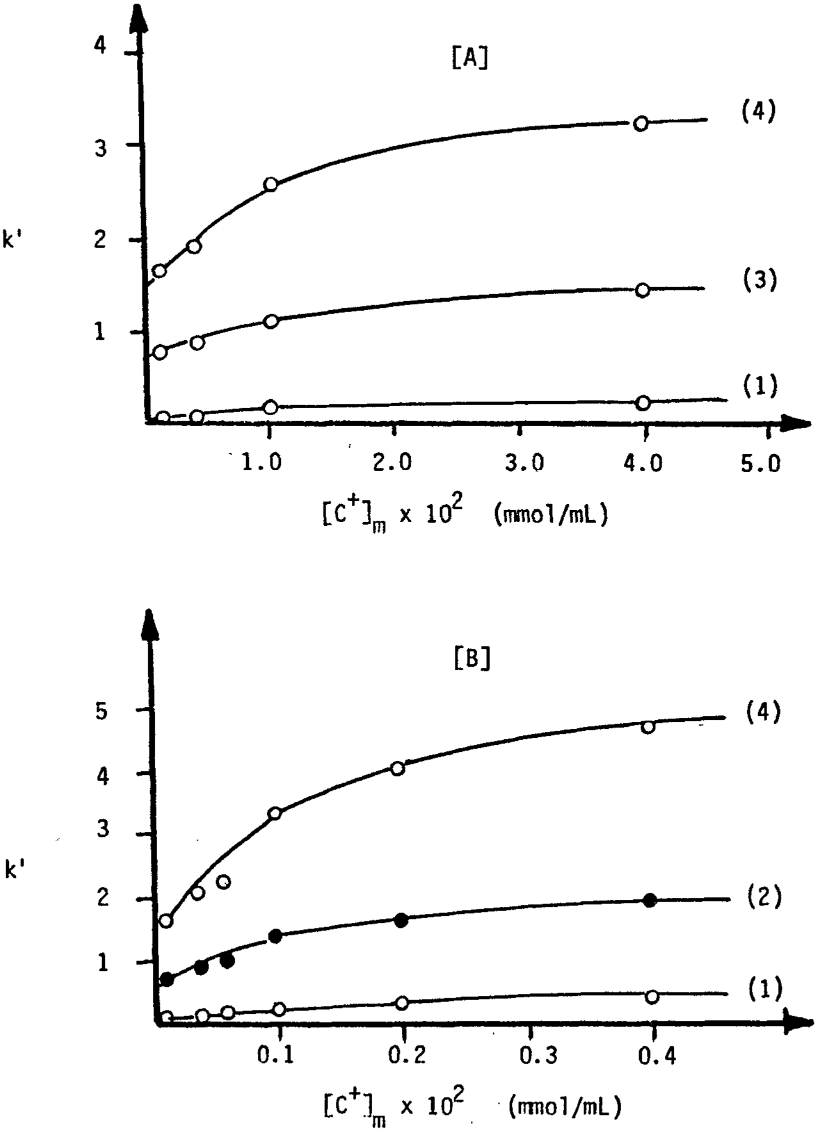

Figure 1. Effect of Counterion Concentration in the Mobile Phase on Solute Retention.

Conditions as in TABLES 1 and 2

Counterions:	[A] TPA	[B] TBA
Solutes:	(1) p. hydroxybenzoate	(2) sorbate
	(3) o. toluate	(4) p. toluate

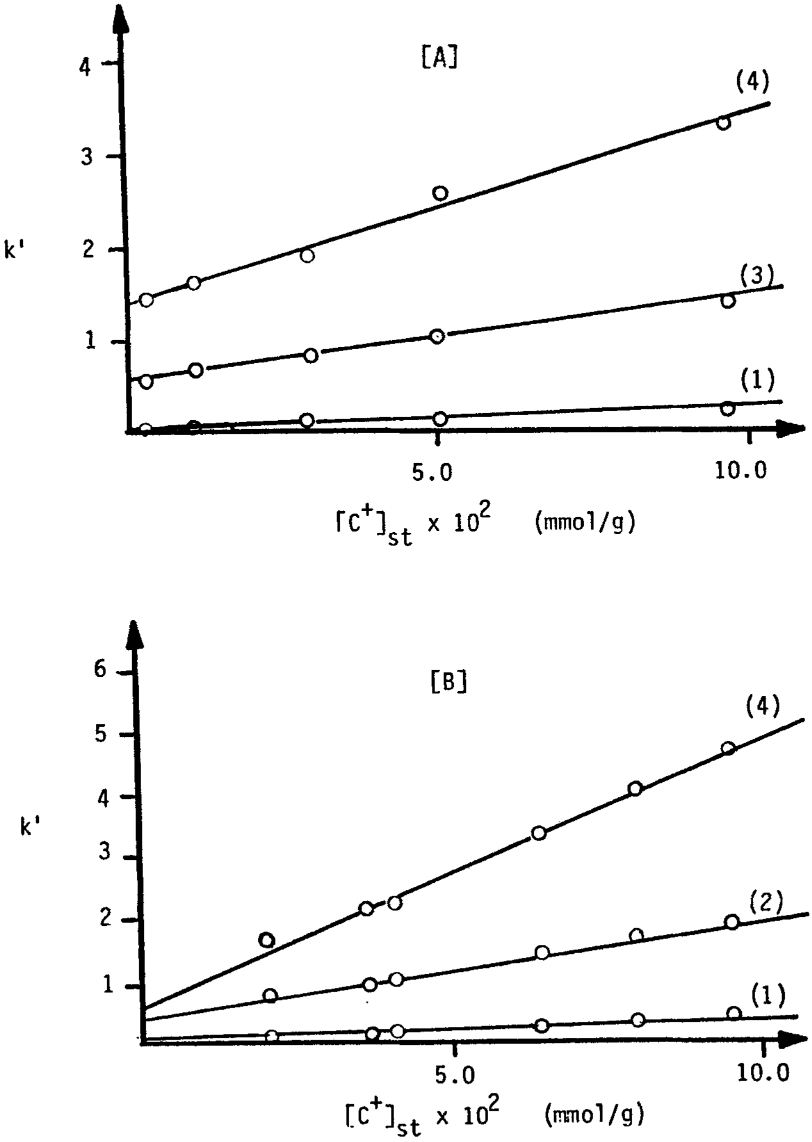

Figure 2. Effect of Counterion Concentration in the Stationary Phase on Solute Retention

Conditions as in TABLES 1 and 2
Counterions: [A] TPA [B] TBA
Solutes: (1) p. hydroxybenzoate (2) sorbate
(3) o. toluate (4) p. toluate

TABLE 3

Regression Parameters.

Derived from application of the relation: $k' = A + B[C^+]_{st}$ to the retention data of TABLES 1 and 2.

SOLUTE*	INTERCEPT		SLOPE		Corr. Coef.	
	TPA	TBA	TPA	TBA	TPA	TBA
(1)	0.03	0.06	2.30	3.71	+0.995	+0.986
(2)	0.68	0.47	7.84	15.1	+0.996	+0.996
(3)	0.56	0.30	8.84	17.2	+0.998	+0.995
(4)	1.36	0.63	20.8	42.1	+0.991	+0.997

* Solutes as in TABLE 1.

$$k' = A + B\,[C^+]_{st} \qquad (1)$$

where, $[C^+]_{st}$ is the tetralkyl ammonium concentration in the stationary phase and A and B are constants.

TABLE 3 reports the regression parameters resulting from the application of equation (1) to the experimental values of TABLES 1 and 2.

Effect of Salt Concentration in the Mobile Phase.

In this study we measured the retention of the solutes at different concentrations of ammonium acetate in the eluent, first without counterion and then with a constant TBA concentration in the mobile phase.

In a previous work (18) it was demonstrated that the variation of salt concentration provokes slight changes in the adsorption of counterion. Therefore, in TABLE 4 and Figure 3 we report the value of the function : $\{(k' - k_0)/[C^+]_{st}\}$ for each concentration of ammonium acetate in the mobile phase.

The behavior of all the solutes in these experiments can be expressed by the relationship:

$$\{(k' - k_0)/[C^+]_{st}\} = C + D\,[P^-]_m^{-1} \qquad (2)$$

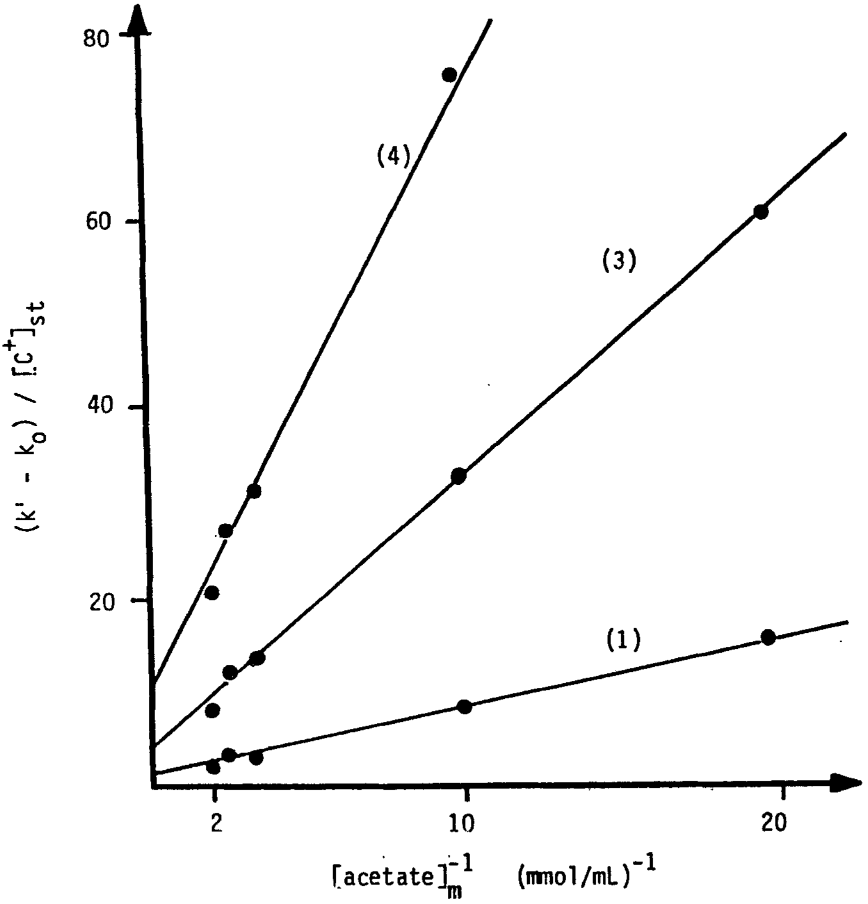

Figure 3. Effect of Salt Concentration in the Mobile Phase on Solute Retention

Conditions as in TABLE 4
Counterion: TBA
Solutes: (1) p. hydroxybenzoate (3) o. toluate
(4) p. toluate
Co-Ion: Acetate

TABLE 4

Effect of Salt Concentration on Solute Retention.

Conditions: column (15cm x 4.6mm i.d.) packed with 5 μ Supelcosil LC-7; mobile phase acetonitrile-water (pH=7) 10:90 v/v containing TBA 0.002 M and ammonium acetate. Temp 40°C.

$[\text{acetate}]_m$	$(k' - k_0)/[C^+]_{st}$			
(mmol/mL)	1*	2*	3*	4*
0.05	15.6	58.8	61.6	139
0.10	8.61	32.1	33.5	77.0
0.30	3.13	12.1	13.7	31.9
0.40	3.69	10.4	12.1	27.5
0.50	2.19	6.72	7.99	21.0

* Solutes as in TABLE 1.

TABLE 5

Regression Parameters

From application of the relation: $\{(k'-k_0)/[C^+]_{st}\} = C + D\,[P^-]_m^{-1}$ to the data of TABLE 4.

SOLUTE*	INTERCEPT	SLOPE	Corr. Coef.
1	1.15	0.726	+0.996
2	2.53	2.84	+0.999
3	3.77	2.91	+0.999
4	10.2	6.48	+0.9995

* Solutes as in TABLE 1.

where, C and D are constants and $[P^-]_m$ represents the molar concentration of acetate in the mobile phase. We consider here that solute retention is affected because ions of the same charge (co-ions), dissolved in the mobile phase, compete with solutes for associating with the counterion.

Application of equation (2) to the data of TABLE 4 gives the regression parameters reported in TABLE 5.

Effect of Co-Ion Type.

Solute retention was measured for different anions dissolved in the mobile phase. Results are shown in Figure 4.

In all the experiments we worked with a 0.04 M concentration of the studied anion, a 0.002 M TBA concentration and a 0.01 M concentration of ammonium acetate, used to fix the pH of the mobile phase.

Figure 4 shows that, for all the solutes, retention increases in the following order:

{citrate^{3-}/citrate^{2-}} < crotonate$^-$ ≈ sulphate^{2-} < {phosphate^{2-}/ phosphate$^-$} < acetate$^-$.

this means that the competition effect increases with the charge and the hydrophobicity of the co-ion.

DISCUSSION

In order to deduce the mechanisms that take place in the retention of ionic solutes in IPC with hydrophobic counterions we will examine briefly the expressions derived from the two principal proposals and we will face them with our results.

A.- Ion Pair formation in the mobile phase, or at the interface, prior to adsorption of this species on the stationary phase.

Let C^+ be a cationic counterion and S^- an anoinic solute. The mobile phase contains also other cations and anions from buffers or other dissolved salts. The following equilibria are supposed to occur (1,2) :

Adsorption of the solute independent of the counterion.

$$S^-_m \overset{K_0}{\Longleftrightarrow} S^-_{st}$$

Adsorption of the solute with the counterion.

1) $$S^-_m + C^+_m \overset{K}{\Longleftrightarrow} [S^{-+}C]_{st}$$

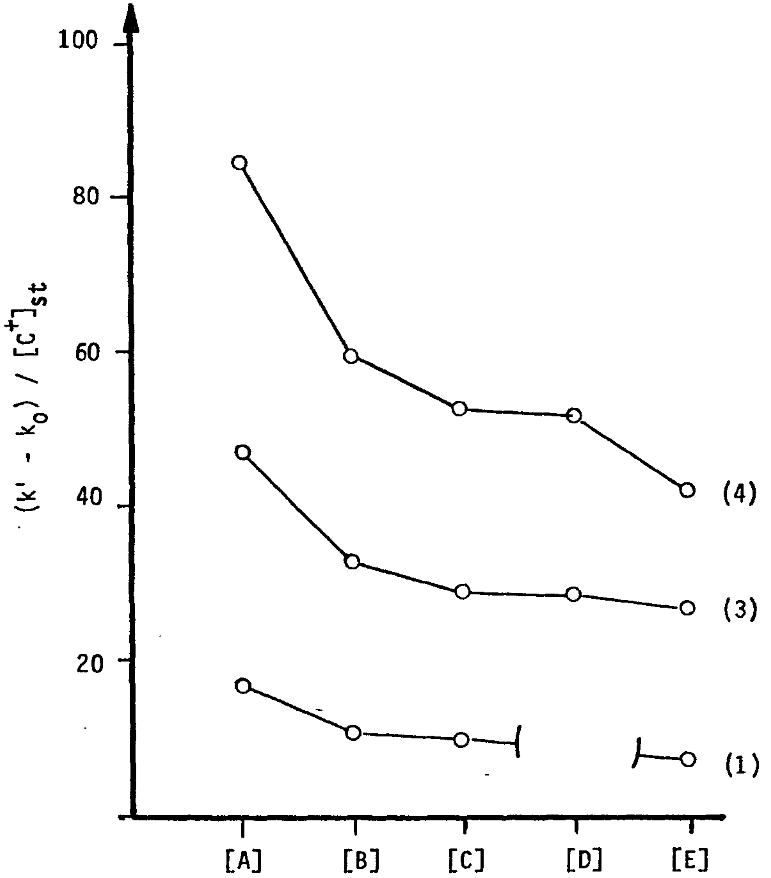

Figure 4. Effect of Co-ion Type on Solute Retention

Conditions: column (15cm x 4.6mm i.d.) packed with 5 µ Supelcosil LC-7; mobile phase acetonitrile-water (pH=7) 10:90 v/v containing TBA 0.002 M, ammonium acetate 0.01 M and studied co-ion 0.04 M.

Solutes: (1) p. hydroxybenzoate (3) o. toluate (4) p. toluate

Co-Ions: [A] CH_3COO^- ; [B] $HPO_4^{2-}/H_2PO_4^-$; [C] SO_4^{2-}

[D] $CH_3CH{:}CHCOO^-$; [E] $C_3H_4OH(COO^-)_3/C_3H_4OHCOOH(COO^-)_2$

or

2) $$S^-_m + C^+_m \overset{K_1}{<=====>} [S^{-+}C]_m \overset{K_2}{<=====>} [S^{-+}C]_{st}$$

The capacity factor of the solute in the first case is given by:

$$k' = k_0 + \beta K\,[C^+]_m \qquad (3)$$

If ion pair formation in the mobile phase occurs then:

$$k' = \frac{k_0 + \beta K_1 K_2\,[C^+]_m}{1 + K_1\,[C^+]_m} \qquad (4)$$

where, k_0 represents solute retention without counterion in the mobile phase and β is the phase relation.

Results in Figure 1 demonstrate that solute retention in the presence of TPA or TBA does not follow equation (3).

On the other hand, Figure 3 shows a remarkable dependance of retention on the concentration of salts in the mobile phase eventhough, in this case, the counterion concentration in the eluent was constant. The effect of salts on retention cannot be attributed to changes in the k_0 value, which are minor. So we conclude that equation (4) does not represent the retention mechanism in our conditions.

B.- Dynamic Ion Exchange.

This model proposes that a counterion present in the mobile phase, if sufficiently hydrophobic, will be adsorbed on the stationary phase associated to an ion of opposite charge (co-ion). The stationary phase is thus transformed into an ion exchanger where solute and co-ions compete for a place (7-10, 12). This process is represented by the equilibrium:

$$[P^{-+}C]_{st} + S^-_m \overset{K_3}{<=====>} [S^{-+}C]_{st} + P^-_m$$

Considering also the adsorption of the solute independent of the counterion, the capacity factor is given by:

$$k' = K_0 + \beta K_3 \frac{[P^{-+}C]_{st}}{[P^-]_m} \qquad (5)$$

From equation (5) it is evident that, at constant salt concentration, a plot of k' versus the counterion concentration in the stationary phase (as in Figure 2), must be linear. The intercept will be equal to K_0 and βK_3 will be given by the product {slope x $[P^-]_m$}.

If counterion concentration in the stationary phase and salt concentration in the mobile phase change, then a plot of $\{(k' - K_0)/[C^+]_{st}\}$ versus $\{[P^-]_m^{-1}\}$ (as in Figure 3) will be linear with a zero intercept and a slope equal to βK_3.

Figures 2 and 3 show that our results follow the linear behaviors mentionned above for all the solutes studied. Besides, we found that the affinity order of the stationary phase for different anions is similar to that existing in strong anion exchange resins. We must conclude from this that in IPC with hydrophobic counterions an ion exchange mechanism participates in solute retention.

Nevertheless, as we show in the following paragraphs, the analysis of intercepts and slopes suggests that ion exchange is not the only mechanism involved in solute retention. Another phenomenon whose importance increases as solute and counterion hydrophobicity augments, is affecting retention.

In Figure 2 we worked with constant salt concentration in the eluent and variable counterion concentration in the mobile and, thus, in the stationary phase. When we compare the intercepts (reported in TABLE 3) with the K_0 values of the four solutes (in TABLE 1 or 2) we find that with TPA as counterion both sets of values are similar but with TBA the intercepts are markedly smaller than the K_0 values and this difference is larger for the more retained solutes (more hydrophobic).

TABLE 6 reports the value of the product {slope x $[P^-]_m$} for every solute with both counterions, derived from Figure 2. Ideally this product represents the ion exchange constant (βK_3) which should be independent of the hydrocarbon area of a previously adsorbed counterion. But this is not the case and we observe that the larger counterion gives higher values and the relative difference between both sets of values increases with solute hydrophobicity.

The results in Figure 3 with TBA as counterion correspond to conditions where the salt concentration in the eluent is variable and the counterion concentration is constant in the mobile phase but slightly variable in the stationary phase. From equation (5) we would expect a zero value of the intercept for all the solutes but this is not the case and the extrapolated intercepts (TABLE 5) are significantly different from zero. This fact indicates the existance of an extra retention mechanism, independent of salt concentration, which superimposes the ion exchange process.

More evident is the fact that the slopes in Fig. 3 differ from the values of the product {slope x $[P^-]_m$} from Figure 2B (TABLE 6). Both sets of values refer to the same solutes and counterion (TBA) and they should represent the ion exchange constants if the

TABLE 6

Analysis of Slopes From Figures 2 and 3.

"A" - Product {slope x $[P^-]_m$} for TPA and TBA. Mobile phase: [salt] constant, [counterion] variable. Stationary phase: [counterion] variable.

"B" - Slope for TBA as counterion. Mobile phase: [salt] variable, [TBA] constant. Stationary phase: [TBA] slightly variable.

SOLUTE*	"A" (Figure 2)		"B" (Figure 3)
	TPA	TBA	
1	0.713	0.93	0.726
2	2.43	3.78	2.84
3	2.74	4.30	2.91
4	6.45	10.5	6.48

*Solutes as in TABLE 1.

retention mechanism were "pure" ion exchange. Surprisingly the set of values from Figure 3 is similar to the products {slope x $[P^-]_m$} for TPA.

Proposition of a Model.

The preceeding discussion leads us to the following proposals: In the presence of hydrophobic counterions in the eluent, the stationary phase is transformed into an electrical double layer formed by an ionic surface (hydrophobic chains + adsorbed counterions) and a diffuse ionic cloud composed mainly by co-ions. The solute enters into this phase by two different pathways:

- First, attracted by the ionic surface the solute can get into the diffuse ionic cloud displacing a co-ion.

For a system with cationic monovalent counterions, C^+, monovalent solutes, S^-, and co-ions, P^-, this mechanism is represented by the equilibrium:

$$P^-_{ic} + S^-_m \Longleftrightarrow P^-_m + S^-_{ic}$$

$$K_{ie} = \frac{[P^-]_m\,[S^-]_{ic}}{[P^-]_{ic}\,[S^-]_m} \qquad (6)$$

where, the subscript "ic" represents the ionic cloud and K_{ie} is the ion exchange constant.

For an analytical injection of the solute we can state:

$$[S^-]_{ic} \ll [P^-]_{ic} \qquad \text{and} \qquad [P^-]_{ic} \approx [C^+]_{st}$$

then:

$$\frac{[S^-]_{ic}}{[S^-]_m} = K_{ie}\,\frac{[C^+]_{st}}{[P^-]_m} \qquad (7)$$

According to our results, when the counterion is not strongly hydrophobic, ion exchange is the only path for solute retention involving the participation of the counterion. The capacity factor for the solute is then given by:

$$k'_1 = k_0 + \beta K_{1e} \frac{[C^+]_{st}}{[P^-]_m} \qquad (8)$$

where, k_0 is the capacity factor of the solute in the absence of counterion in the system.

- Second, the hydrophobic effects experienced by the solute in the mobile phase and by the adsorbed counterions, specially if they have a large hydrocarbon area, can promote the <u>formation of an ion pair on the surface of the packing</u> .

This process can be represented by the equilibrium:

$$S^-_m + C^+_{st} \Longleftrightarrow [S^{-+}C]_{st}$$

$$K_{ip} = \frac{[S^{-+}C]_{st}}{[S^-]_m\ [C^+]_{st}} \qquad (9)$$

where the subscript "ip" stands for ion pair formation. The stability of the resulting ion pair depends on the structure of the solute and on the hydrophobicity of both entities.

Our experimental results indicate that the contribution of this mechanism to retention is expressed by a relation of the type:

$$k'_2 = -Z + \beta K_{ip} [C^+]_{st} \qquad (10)$$

where, Z is a constant term whose value depends on the nature of solute and counterion. We cannot give a physical interpretation of this parameter but we have observed that, in IPC with strongly hydrophobic counterions, a negative constant term is systematically present. In a previous work (14) with alkyl-sulphonates as counterions we also found a linear relation between k' and the counterion concentration in the stationary phase; the intercepts of the plots for the solutes studied in the presence of octylsulphonate corresponded to the k_0 values, but ,with dodecylsulphonate, the intercepts for the same solutes were plainly negative.

When ion pair formation on the surface of the packing occurs, the two mechanisms contribute simultaneously to retention, then:

$$k' = k'_1 + k'_2$$

$$k' = F + \{ \beta K_{1p} + \beta K_{1e} [P^-]_m^{-1} \} [C^+]_{st} \qquad (11)$$

or:

$$\frac{k' - F}{[C^+]_{st}} = \beta K_{1p} + \beta K_{1e} [P^-]_m^{-1} \qquad (11')$$

where, $F = k_0 - Z$

Test of the Model

From Figure 2B (for TBA) we obtained the value of "F" (intercepts) for each solute. This one was used in conjunction with the data of the experiments at different salt concentrations to calculate the function $\{(k' - F)/[C^+]_{st}\}$. Then by linear regression, applying equation (11'), we obtained the equilibrium constants of the proposed model (TABLE 7). Note that these values are deduced from experimental conditions where $[C^+]_m$ is constant and $[P^-]_m$ is variable.

The last column in TABLE 7 represents the slope in the relation:

$k' = A + B [C^+]_{st}$ {equation (1)}

TABLE 7

Equilibrium Constants of the Proposed Model.

SOLUTE*	βK_{ip}	βK_{ie}	$\{\beta K_{ip} + \beta K_{ie}[P^-]_m^{-1}\}$#
1	1.34	0.65	3.94
2	4.88	2.70	15.7
3	7.03	2.81	18.3
4	19.1	6.39	44.7

* Solutes as in TABLE 1
\# Calculated with $[P^-]_m = 0.25$ M

it was calculated for the 4 solutes at $[P^-]_m$ = 0.25 M using the equilibrium constants of the model and applying equation (11). These values can be compared with those reported in TABLE 3 (slopes) for TBA. There, the experimental conditions were $[C^+]_m$ variable and $[P^-]_m$ constant (0.25 M). The similarity between both sets of values demonstrates the coherence of the model.

Moreover, if we compare the βK_{ie} values in TABLE 7 deduced when using TBA as counterion, with the products (slope x $[P^-]_m$) for TPA, in TABLE 6, a strong similarity becomes evident. We mentionned before that for a "pure" ion exchanger this product is equal to the ion exchange constant mutiplied by the phase relation. Thus, we infer that with TPA as counterion the retention of the studied solutes is due only to an ion exchange mechanism. This means that TPA has a very weak tendency to form ion pairs with those solutes at the surface of the stationary phase.

We can thus reiterate that the value of the ion exchange constant, K_{ie}, depends only on the structure and charge of the solute and is independent of the hydrocarbon area of the adsorbed counterion.

CONCLUSIONS

In IPC, with hydrophobic counterions, ionic compounds can be retained by two different mechanisms involving the counterion.

One is ion exchange, where the solute displaces co-ions from a diffuse ionic cloud associated electrically to the surface of the packing. This surface is composed by hydrocarbon chains and adsorbed counterions.

The other is based on an ion pair formation, between the solute and the adsorbed counterion, at the surface of the packing.

The first mechanism holds with all counterions if they are sufficiently hydrophobic and do adsorb on the stationary phase.

The second mechanism, which superimposes the ion exchange process, occurs only if the solute and/or the adsorbed counterion are strongly hydrophobic.

ACKNOWLEDGMENTS

Authors gratefully acknowledge "Consejo Nacional de Ciencia y Tecnología de México" for the financial support of this work.

REFERENCES

(1) Westerlund, D. and Theodorsen, A., Reversed-Phase Ion-Pair Chromatography of Naphtalene-Acetic Acid Derivatives with Water and Organic Modifier as the Mobile Phase, J.Chromatogr., 144, 27, 1977.

(2) Horváth, C., Melander, W., Molnár, I. and Molnár,P Enhancement of Retention by Ion-Pair Formation in Liquid Chromatography with Nonpolar Stationary Phases, Anal. Chem., 49, 2295, 1977.

(3) Tilly-Melin, A., Askemark, Y., Wahlund, K.-G. and Schill, G., Retention Behavior of Carboxylic Acids and Their Quaternary Ammonium Ion Pairs in Reversed Phase Chromatography with Acetonitrile as Organic Modifier, Anal. Chem., 51, 976, 1979.

(4) Sokolowski, A. and Wahlund, K.-G., Peak Tailing and Retention Behavior of Tricyclic Antidepressant Amines and Related Hydrophobic Ammonium Compounds in Reversed-Phase Ion-Pair Liquid Chromatography on Alkyl-Bonded Phases, J. Chromatogr., 189, 299, 1980.

(5) Eksborg, S. and Ekqvist, B., Reversed-Phase Ion- Pair Liquid Chromatography of Tetracyclines, J. Chromatogr., 209, 161, 1981.

(6) Tomlinson, E., Riley, C.M. and Jefferies, T.M., Ion-Pair High-Performance Liquid Chromatography. The Use of Low Concentrations of Long-Chain Alkyl- benzyldimethylammonium Chlorides for Resolving Anionic Solutes, J. Chromatogr., 173, 89, 1979.

(7) Kraak, J.C., Jonker, K.M. and Huber, J.F.K., Solvent-Generated Ion-Exchange Systems with Anionic Surfactants for Rapid Separations of Amino Acids, J. Chromatogr., 142, 671, 1977.

(8) Terweij-Groen, C.P., Heemstra, S. and Kraak, J.C., Distribution Mechanism of Ionizable Substances in Dynamic Anion-Exchange Systems Using Cationic Surfactants in High-Performance Liquid Chromatography, J. Chromatogr., 161, 69, 1978.

(9) Van de Venne, J.L.M., Hendrix, J.L.H. and Deelder, R.S., Retention Behavior of Some Carboxylic Acids in Reversed-Phase Column Liquid Chromatography, J.Chromatogr., 167, 1, 1978.

(10) Deelder, R.S., Linssen, H.A.J., Konijnendijk, A.P. and Van de Venne, J.L.M., Retention Mechanism in Reversed-Phase Ion-Pair Chromatography of Amines and Amino-Acids on Bonded Phases, J. Chromatogr., 185, 241, 1979.

(11) Knox, J.H. and Laird, G.R., Soap-Chromatography. A New High Performance Liquid Chromatography Technique for Separation of Ionizable Materials. Dyestuff Intermediates, J. Chromatogr., 122, 17, 1976.

(12) Knox, J.H. and Hartwick, R.H., Mechanism of Ion- Pair Liquid Chromatography of Amines, Neutrals, Zwitterions and Acids Using Anionic Hetaerons, J. Chromatogr., 204, 3, 1981.

(13) Knox, J.H. and Jurand, J., Determination of Para- cetamol and its metabolites in Urine by High-Performance Liquid Chromatography Using Ion-Pair Systems, J. Chromatogr., 149, 297, 1978.

(14) Vera-Avila, L.E., Caude, M. and Rosset, R., Chromatographie de Paires d'Ions. 1.- Fixation du Contreion et Mécanismes de Retention, Analusis, 10, 36, 1982.

(15) Bidlingmeyer, B.A., Deming, S.N., Price Jr., W.P., Sachok,B. and Petrusek, M., Retention Mechanism for Reversed-Phase Ion-Pair Liquid Chromatography, J. Chromatogr., 86, 419, 1979.

(16) Melander, W.R. and Horväth, C., Mechanistic Study on Ion-Pair Reversed-Phase Chromatography, J. Chromatogr., 144, 27, 1977.

(17) Stranahan, J.J. and Deming, S.N., Thermodynamic Model for Reversed-Phase Ion-Pair Liquid Chromatography, Anal. Chem., 54, 2251, 1982.

(18) Del Rey, M.E. and Vera-Avila, L.E., Adsorption of Tetralkyl Ammonium Ions on Reversed-Phase HPLC Columns, J. Liq. Chromatogr., 10, 2911, 1987.

(19) Charlot, G.,, Chimie Analytique Quantitative II, Masson, 6th edition, Paris, 1974, p. 342.