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ION-PAIR CHROMATOGRAPHY OF ORGANIC COMPOUNDS

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SUMMARY

Ionized organic compounds can be extracted into organic phases as ion-pairs with counter ions of suitable hydrophobic character. The distribution ratio depends on the structure of the ion-pair and the concentration of the counter ion.

Systems for liquid–liquid ion-pair partition chromatography are easy to adapt to different types of compounds and have a high selectivity. The counter ion is selected on the basis of batch extraction procedures or chromatographic tests. A photometric detector can be used for samples of <0.1 nmole of non-absorbing compounds if a counter ion with high absorbance is used.

Straight-phase systems with a high counter-ion concentration on a cellulose support have a high sample capacity and good separating efficiency ($H \leqslant 0.5$ mm at a mobile phase speed of $\leqslant 3$ mm/sec when $k' \geqslant 5$).

Examples are given of straight-phase separations of carboxylic acids with a quaternary ammonium ion as counter ion in the stationary phase and amines and quaternary ammonium ions with picrate or β-naphthalene sulphonate as counter ions.

Aminophenols have been separated by reversed-phase systems containing bis-(2-ethylhexyl)phosphoric acid in chloroform as the stationary phase.

INTRODUCTION

Ion-pair chromatography is based on a liquid–liquid partition technique usually called ion-pair extraction or ion-pair partition.

Ion-pair partition is well known from the inorganic field, and during the last 10 years it has also found use in many procedures for the isolation and determination of organic compounds, in ion-selective electrodes, in photometric and fluorimetric procedures[1,2], in extraction processes[3,4], etc. It can be applied to all kinds of ionizable organic compounds, aprotic ions such as quaternary ammonium ions, sulphonates and organic sulphates as well as weak protolytes such as carboxylic acids, phenols, amines and amino acids.

The general principle of ion-pair extraction is illustrated by the equation

$$Q^+_{aq} + X^-_{aq} = QX_{org} \quad (1)$$

The extraction of the cation, Q^+, as an ion-pair with the counter ion, X^-, can be expressed quantitatively by the distribution ratio, D_{QX},

$$D_{QX} = E_{QX} \cdot [X^-] \quad (2)$$

where E_{QX} is the extraction constant defined by

$$E_{QX} = [QX]_{org} \cdot [Q^+]^{-1} \cdot [X^-]^{-1} \quad (3)$$

The distribution ratio of the cation can be varied within wide limits by the concentration and the nature of the counter ion, X^-.

The influence of the nature of the ions is demonstrated in Tables I and II. Extraction constants of ion-pairs between tetrabutylammonium and different anions are given in Table I, and between chloride and different organic ammonium ions in Table II. The organic phase is chloroform in both cases.

TABLE I

EXTRACTION CONSTANTS OF TETRABUTYLAMMONIUM ION-PAIRS

Organic phase: chloroform.

Anionic component	*log E_{QX}*	*Ref.*
Cl^-	−0.11	5
Br^-	1.29	5
ClO_4^-	3.48	5
Benzoic acid	0.39	6
Salicylic acid	2.42	6
Toluene-4-sulphonic acid	2.33	1
Naphthalene-2-sulphonic acid	3.45	1
Picric acid	5.91	5
Dipicrylamine	9.6	1

TABLE II

EXTRACTION CONSTANTS OF ORGANIC AMMONIUM ION-PAIRS WITH CHLORIDE

Organic phase: chloroform.

Class	*Cationic component*	*log E_{QX}*	*No. of alkyl or aryl carbon atoms*	*Ref.*
Secondary amine	Protriptyline	0.32	19	7
	Desipramine	0.34	18	7
	Nortriptyline	0.46	19	7
Tertiary amine	Lidocaine	−1.47	13	8
	Codeine	−1.12	18	8
	Papaverine	1.96	20	9
	Imipramine	2.14	19	7
	Methadone	2.24	20	9
	Amitriptyline	2.33	20	7
	Chloropromazine	2.45	19	7
Quaternary ammonium ion	N,N-Dimethylprotriptyline	−0.30	21	18

TABLE III

EXTRACTION CONSTANTS OF PICRATE ION-PAIRS

Organic phase	*log E_{ox}*		
	*TBA^{+} [*5]*	*$TMEA^{+}$ [*10]*	*$Choline^{+}$ [*10]*
1-Pentanol	—	1.24	1.19
Methyl isobutyl ketone	—	0.64	0.98
Ethyl acetate	—	0.45	0.69
Methylene chloride	6.68	0.77	−0.19
Chloroform	5.91	0.04	−1.45
Benzene	3.59	—	—
Carbon tetrachloride	1.94	—	—

* TBA^{+} = tetrabutylammonium; $TMEA^{+}$ = trimethylethylammonium; $choline^{+}$ = 2-hydroxyethyltrimethylammonium.

The addition of one aryl or alkyl carbon usually increases log E_{ox} by 0.5–0.6. Hydrophilic substituents such as hydroxyl and carboxyl groups will give a decrease, but the size of the change depends strongly on the properties of the organic phase (*cf.*, $TMEA^{+}$ and $choline^{+}$ in Table III).

All ion pairs have a polar character, and they are extracted much better by slightly polar solvents such as chloroform and methylene chloride than by non-polar solvents such as aliphatic hydrocarbons and carbon tetrachloride (*cf.*, Table III).

Strongly hydrogen bonding solvents such as lipophilic alcohols have an even higher extracting power, particularly for ion-pairs with hydrophilic substituents. However, such extraction agents are often much less selective than solvents with a lower solvating capacity. (*cf.*, Table III).

Extraction constants for compounds of widely different structures have been given in a review of ion-pair partition methods[11].

Ion-pair partition in liquid chromatography

The ion-pair systems have several properties that are of particular value in liquid–liquid chromatography. They are very versatile and can easily be adapted to different kinds of samples by a proper choice of kind and concentration of counter ion. Another advantage is the high selectivity that can be obtained. It follows from the fact that most ion-pair extractions can be achieved with solvents of rather low polarity that even a highly hydrophilic compound, *e.g.* an amino acid, can be extracted with methylene chloride by the use of a sufficiently hydrophobic counter ion.

Straight-phase ion-pair chromatography can furthermore be used to obtain a high detection sensitivity with a UV detector: any sample, irrespective of its UV absorbance, will give a high response if a counter ion with high molar absorptivity is used as the stationary phase.

EXPERIMENTAL

Apparatus and materials

Detector. Chromatronix 200-L photometer (8-μl cell volume, path length 10 mm).

Pump. Chromatronix "Cheminert" metering pump, CMP-1 L.

Columns. Separation column and pre-column of borosilicate glass.

Supports. Cellulose, ethanolised (Munktell 410), extracted with ethanol to remove traces of pyridine; silicon-treated cellulose (Macherey, Nagel & Co.)

Chromatographic technique

The columns were packed with a rod by using the slurry technique. The samples were injected as ion-pairs dissolved in the mobile phase. The experimental temperature was 25.0° (ref. 12).

RESULTS AND DISCUSSION

The chromatographic separation of three acids with different polarities, namely phenylbutyric acid, salicylic acid and benzilic acid, shown in Fig. 1, illustrates the principle of ion-pair chromatography. The stationary phase is an aqueous solution of a quaternary ammonium ion, N,N-dimethylprotriptyline, on a support of ethanolised cellulose. The anions of the acids form ion-pairs with the quaternary ammonium ion and migrate in this form with the mobile organic phase.

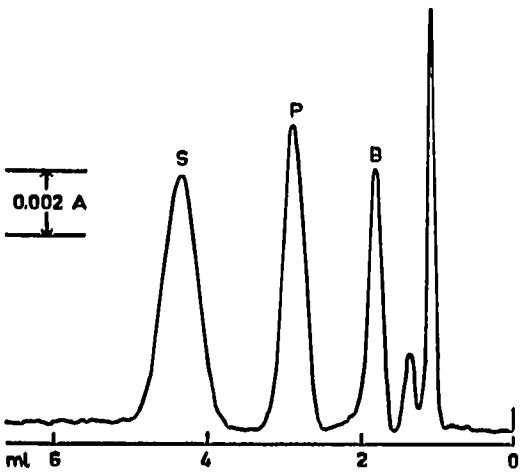


Fig. 1. Ion-pair chromatography of carboxylic acids. Sample: benzilic acid (B) 0.7 nmole; phenylbutyric acid (P) 1.1 nmoles; salicylic acid (S) 1.4 nmoles. Stationary phase: N,N-dimethylprotriptyline, 0.036 *M*, pH 9.0 (30% on cellulose). Mobile phase: cyclohexane–chloroform–1-pentanol (75:20:5). Mobile phase speed: 2 mm/sec. Column: I.D. 2.7 mm, length 300 mm.

The anions of the acids have a low absorbance at the wavelength of the detector (254 nm), but the cation contains a polycyclic aromatic ring system and gives ion-pairs with a molar absorptivity of about $4 \cdot 10^3$. The chromatogram was obtained with 0.7–1.4 nmoles of the acids, but the response of the detector to the ion-pairs is so high that sample levels at least ten times smaller can be detected.

Support

The specific properties of the ion-pair system cannot be fully utilized unless

TABLE IV

ION-PAIR CHROMATOGRAPHY OF CARBOXYLIC ACIDS

Sample concentration: $5 \cdot 10^{-5}$ *M* (30 μl). Stationary phase: N,N-dimethylprotriptyline, 0.036 M, pH 9.0 (30% on cellulose). Mobile phase: cyclohexane–chloroform–1-pentanol (75:20:5). Mobile phase speed: 1 mm/sec. Column: I.D. 2.7 mm, length 300 mm.

Sample	*Cellulose*			*Celite (37–74 μm)*		
	D_{found}*	*Assymmetry factor***	*H (mm)*	D_{found}*	*Asymmetry factor***	*H (mm)*
Phenylbutyric acid	0.095	1.15	0.6	0.091	1.25	2.2
Salicylic acid	0.053	1.12	0.7	0.067	1.65	2.2
Benzilic acid	0.23	1.39	0.7	0.2	>5	—

* $D_{found} = C_{org}/C_{aq} = \dfrac{V_s}{V_m \cdot k'_{found}}$.

** Asymmetry factor = $\dfrac{\text{Back part of } w_h}{\text{Front part of } w_h}$.

TABLE V

ION-PAIR CHROMATOGRAPHY ON DIFFERENT SUPPORTS

Sample: trimethylethylammonium picrate, $7 \cdot 10^{-5}$ *M* (10 μl). Stationary phase: picrate, 0.06 *M*, pH 11.2. Mobile phase: chloroform–1-pentanol (19:1). Column: I.D. 2.7 mm, length 300 mm.

Support	*Stationary phase loading (%)*	$\dfrac{k'_{found}}{k'_{calc}}$*	*Asymmetry factor*
Cellulose	25	1.0	0.83
	38	0.9	0.60
Celite	25	1.6	7.5
(37–74 μm)	50	1.3	6.4
Celite	25	2.2	>13
(15–37 μm)	50	1.0	7.4

* $k'_{calc} = \dfrac{V_s}{V_m \cdot E_{QX} \cdot [X^-]}$.

the composition of the stationary phase is well known and the support is so inert that its influence on the migration rate of the sample is negligible. Disturbances from the support have been one of the major problems in ion-pair chromatography, and it is probable that the strong tendency for interaction between sample and support to occur is due to the fact that the sample is present in a charged form, as an ion, in the stationary phase.

Some results obtained with two porous supports with low adsorptive properties, diatomaceous earth (Celite) and a brand of cellulose (ethanolised cellulose, Munktell 410) are presented in Table IV. The diatomaceous earth was thoroughly washed with acid, and the particles were fractionated according to their diameters by sieving. The cellulose particles had mean diameters of 30–65 μm (determined by the Coulter Counter technique).

The chromatograms were obtained with the same phase systems as those in Fig. 1. The migration rates on the columns are illustrated by the found distribution ratios, D_{found}, calculated from the found capacity factors and measured phase volumes. The D values do not indicate any significant differences between the two columns, but the cellulose column is superior to the Celite column as it has a considerably higher separating efficiency and good peak symmetry for all the samples. The rather high H value on the Celite column may be due to the large particle diameters, but the strongly tailing benzilic acid peak indicates a more specific effect.

Similar differences between cellulose and Celite columns were also obtained with picrate as the stationary phase and a quaternary alkylammonium ion as the sample[12] (Table V). There is good agreement between the found and calculated capacity factors, particularly on the cellulose column and, at a high stationary phase loading, also on the Celite column. This shows that both supports have a negligible influence on the migration rate of the sample, but the asymmetry factors indicate the important difference that the Celite column gives peaks with a very pronounced tailing while the peaks on the cellulose column show a slight leading effect.

Tests on Porasil supports with large pore diameters gave results that indicated strong adsorption effects.

Separation efficiency

The ethanolised cellulose has advantages as a support in ion-pair chromatography owing to its inert character, but it also has a drawback in its non-rigid structure, which prevents its use in high-pressure liquid chromatography. However, the moderate flow-rates will to some extent be compensated by the high selectivity of the ion-pair systems and by the good separation efficiency.

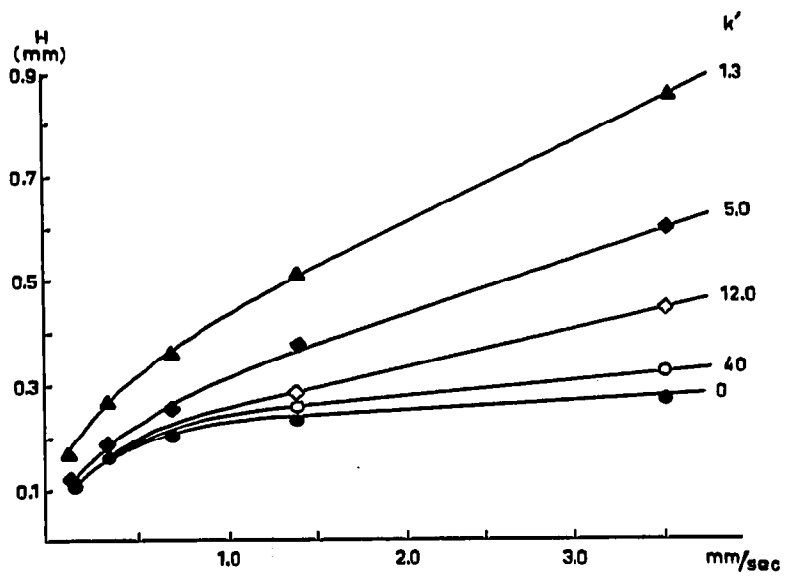


Fig. 2. Separation efficiency of columns with cellulose as support. Sample: quaternary alkylammonium ions (as picrate), 10^{-4} M. Stationary phase: picrate, 0.06 M, pH 11.2 (25% of support weight). Mobile phase: chloroform–1-pentanol (19:1). Column: I.D. 2.7 mm, length 300 mm.

The relationship between H and the mobile phase speed at an optimum stationary phase loading of 25% is shown in Fig. 2. A 0.06 M picrate solution was used as stationary phase and quaternary alkylammonium ions with different capacity factors as samples. Calculations of N_{eff}/t showed that the maximum separation speed was obtained at $k' = 5$, where the limiting mobile phase speed, $v = 3.5$ mm/sec, gave $H = 0.6$ mm.

Columns with smaller diameters had a lower separating efficiency, probably as a result of inhomogenous packing[12].

Sample capacity

The choice of the counter ion has up to now been discussed from one point of view only: as a means of improving the selectivity of detection. However, the main purpose of the counter ion is to give the sample a suitable migration rate. The migration rate depends on the distribution ratio, which is equal to the product of the extraction constant and the counter-ion concentration (eqn. 2), but any combination of E_{ox} and $[X^-]$ that gives a suitable distribution ratio cannot be used.

A stable system with a high sample capacity can only be achieved if the ion-pair system has a high counter-ion buffer capacity, $\beta = dC_Q^0/dpX$ (ref. 12). The high buffer capacity is obtained by using a high counter-ion concentration in the stationary phase. Consequently, the extraction constant must be low, and it is in practice suitable to use a counter ion that gives $E_{ox} < 10$ for the most hydrophobic component of the sample.

An illustration of the sample capacity of a column constructed according to these principles is given in Fig. 3. With 0.06 M picrate as the stationary phase and a quaternary alkylammonium ion as the sample, up to 10^{-7} mole of sample (10 μl of a 10^{-2} M solution) could be injected without more than a very slight increase in H

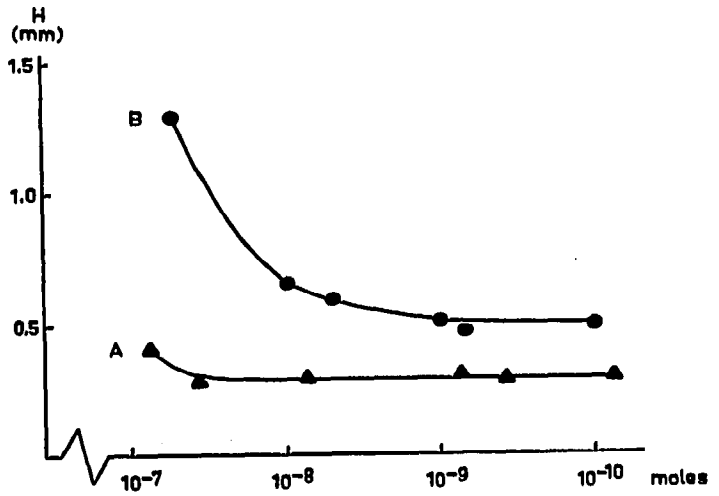


Fig. 3. Sample capacity at different counter-ion concentrations. Sample: A, trimethylethylammonium picrate; B, trimethylpropylammonium picrate (volume, 10 μl in each case). Stationary phase: A, 0.06 M picrate; B, 0.014 M picrate (both pH 11.2, 25% of support weight). Mobile phase: chloroform–1-pentanol (19:1). Mobile phase speed: 1.4 mm/sec. Column: I.D. 2.7 mm, length 300 mm.

A column with a four times lower picrate concentration in the stationary phase had a sample capacity about 40 times lower.

Counter ion

The selection of a group of compounds that are suitable as counter ions can often be based on published constants (*cf.*, ref. 11). The final choice is always combined with the selection of mobile phase composition, and it will usually require determinations of extraction constants by chromatographic tests or batch extraction procedures (*cf.*, ref. 1).

Aprotic counter ions often have advantages, as they can be used at any pH without the risk of a disturbing bleeding. Inorganic anions are ideal in this respect, and they are very suitable for separations of hydrophobic cations, as most of them give low extraction constants[8,13,14].

Samples with low UV absorbance often require a counter ion with high molar absorptivity. Picrate gives ion-pairs with a molar absorptivity of 10^4 (254 nm), which means that samples of less than 0.1 nmole can be quantified with acceptable precision by peak area measurements[12]. A picrate column with chloroform and 1-pentanol (19:1) as the mobile phase gives, with quaternary alkylammonium samples, a separation factor of about 4 for a difference of one CH_2 group. The high selectivity has made such columns very useful in the isolation of acetylcholine and choline in biological material[4,15]. Picrate cannot be used for ion-pair separations of amines, as the bleeding is too disturbing in the pH ranges where the distribution of the amines as bases is negligible.

Separations of different types of ammonium ions, both aprotic and protolytic, can be made with aromatic sulphonates as counter ions. The separation of alkylammonium ions of different degrees of substitution on a column with a β-naphthalene sulphonate solution as the stationary phase is demonstrated in Table VI. β-Naphthalene sulphonate can be used at low pH without disturbing bleeding and gives ion-pairs with a molar absorptivity of about $3 \cdot 10^3$ at 254 nm. The found separation factors, given with trimethylammonium as reference, agree well with calculations based on extraction constants, except for the two primary amines *n*-pentyl- and *n*-hexylamine. It is possible that the deviations are due to adsorption on the support, and an increase in the pentanol content of the mobile phase to 10% increased the separation factor between the two amines to the expected value of about 4.

Separations of anions can be achieved with quaternary ammonium compounds as counter ions (*cf.*, ref. 16 and Fig. 1). Alkylation of an amine is often a convenient way of obtaining a quaternary ammonium ion of suitable properties, as amines with different hydrophobic characters and absorptivities are readily available, *e.g.*, within the pharmaceutical field (*cf.*, refs. 11 and 16).

Ion-pair dissociation effects

Small amounts of samples will sometimes give rise to leading peaks in ion-pair chromatography (*cf.*, Table V), and it has also been observed that the tendency for leading to occur increases with decreasing amount of sample. Effects of this kind may be due to dissociation of the ion-pair in the organic phase, and calculations based on dissociation constants showed that this assumption was probably correct[5,17].

The degree of dissociation will vary with the nature of the ion-pair, quaternary

TABLE VI

ION-PAIR CHROMATOGRAPHY OF ALKYLAMMONIUM IONS

Sample concentration: 10^{-4} *M* (20 μl). Stationary phase: β-naphthalene sulphonate, 0.1 *M*, pH 2.4 (25% on cellulose). Mobile phase: chloroform–1-pentanol (19:1). Column: I.D. 2.7 mm, length 300 mm.

Ammonium ion	k'_{found}	*Asymmetry factor*	α_{calc}	α_{found}
Tetraethyl	7.7	0.85	1.7	1.8
Trimethyl	14.3	1.1	—	—
Di-isopropyl	0.66	1.0	23	22
n-Pentyl	2.07	1.2	13	7
n-Hexyl	1.15	1.2	49	13

TABLE VII

DISSOCIATION EFFECTS IN ION-PAIR CHROMATOGRAPHY

Sample: picrate (as ion-pair), $7 \cdot 10^{-5}$ *M* (10 μl). Support: cellulose (stationary phase loading 25%). Column: I.D. 2.7 mm, length 300 mm.

No.	*Stationary phase**	*Mobile phase*	*k'*	*Asymmetry factor*
1	TMPrABr, $10^{-2.1}$ *M*	Chloroform–1-pentanol (19:1)	< 0.1	—
2	TMPrABr, $10^{-2.1}$ *M* $NaClO_4$, 10^{-2} *M*		11.1	0.91
3	Choline citrate, $10^{-1.1}$ *M*	Methylene chloride–1-pentanol (49:1)	< 0.1	—
4	Choline citrate, $10^{-1.1}$ *M* $NaClO_4$, 10^{-2} *M*		15	0.91

* TMPrABr = trimethylpropylammonium bromide.

ammonium ions usually giving higher dissociation constants than amines[17]. The dissociation also depends on the polarity of the organic phase: methylene chloride and 1-pentanol give higher ion-pair dissociation than chloroform and cyclohexane[5]. The dissociation can be suppressed if other ion-pairs containing the counter ion are present in the organic phase[18].

Some experiments that illustrate the effect of ion-pair dissociation in chromatography are presented in Table VII. A small amount of picrate (0.1 nmole) was used as the sample and quaternary ammonium compounds as counter ions. In experiments 1 and 3 there was a very low bleeding of quaternary ammonium ion into the organic phase, as the only anions present in the aqueous phase were the hydrophilic bromide and citrate. The dissociation of the picrate ion-pair was not suppressed to any great extent, and chromatograms with capacity factors much lower than the calculated values were obtained.

In experiments 2 and 4, perchlorate was added to the aqueous phase at a high concentration. Perchlorate gives much higher extraction constants than bromide and citrate (*cf.*, Table I) and the bleeding of the quaternary ammonium ions from the stationary phase increased considerably. The dissociation of the picrate ion-pairs in these experiments was suppressed with a drastic increase in *k'* and symmetrical peaks resulted.

Reversed-phase separations

By ion-pair chromatography with reversed phases, a solution of the counter ion is used as the mobile phase and the sample is extracted into the stationary phase as an ion-pair[19]. The technique has particular advantages in gradient elution as the migration rate of the sample is easily controlled by the concentration of the counter ion in the mobile phase. However, the possibilities of improving the sensitivity of detection by using a counter ion with a high UV absorbance are lost because the ion-pair is present in the stationary phase.

The reversed-phase technique can also have practical advantages when the sample is a highly hydrophilic compound dissolved in an aqueous solution. The counter ion must, in such cases, have a very high extraction power and extraction agents that give adducts with the ion-pair in the organic phase may be particularly useful. The principle is illustrated in the equation that shows the extraction of a cation, HA^+, with an ion-pairing and adduct-forming acid, HX:

$$HA^+_{aq} + nHX_{org} = HAX \cdot (HX)_{n-1,\,org} + H^+_{aq} \qquad (4)$$

HX is mainly present in the organic phase, as it must be very hydrophobic. The distribution of HA^+ is controlled by pH and the concentration of HX.

A chromatographic separation based on this principle is shown in Fig. 4

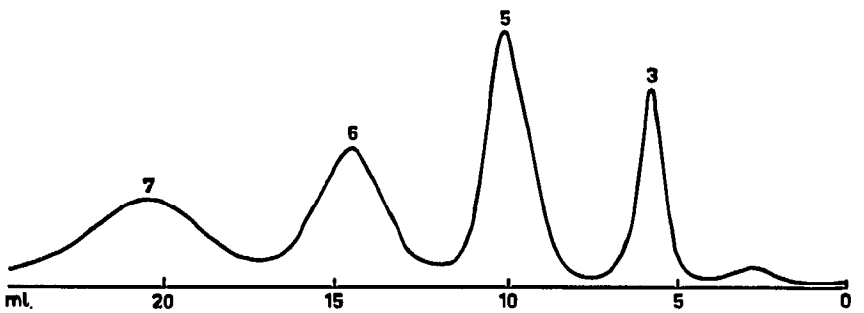


Fig. 4. Reversed-phase chromatography of aminophenols. Samples: 3 = epinephrine; 5 = synephrine; 6 = norphenephrine; 7 = *p*-hydroxynorephedrine (10^{-7} moles of each) (for structures, see Fig. 5). Stationary phase: bis-(2-ethylhexyl)phosphoric acid in chloroform. Mobile phase: citrate buffer, pH 3.8. Column: I. D. 4 mm, length 300 mm.

The samples are four highly hydrophilic derivatives of phenylethanolamine, three monophenols and one diphenol. Silicon-treated cellulose was used as the support carrying a stationary phase of the extracting agent, bis-(2-ethylhexyl)phosphoric acid, dissolved in chloroform.

The selectivity of the system will change with the concentration of the adduct-forming agent, as this changes the composition of the adducts. In a 0.1 *M* solution of the alkylphosphoric acid, mainly adducts with the composition $HAX \cdot (HX)_2$ are present, but an increase in the concentration of the solution will increase the content of HX in the adducts[20]. In chromatographic work, this higher solvation leads to a decrease in the selectivity, as illustrated in Fig. 5.

It must be observed that the reversed-phase columns gave a lower separation

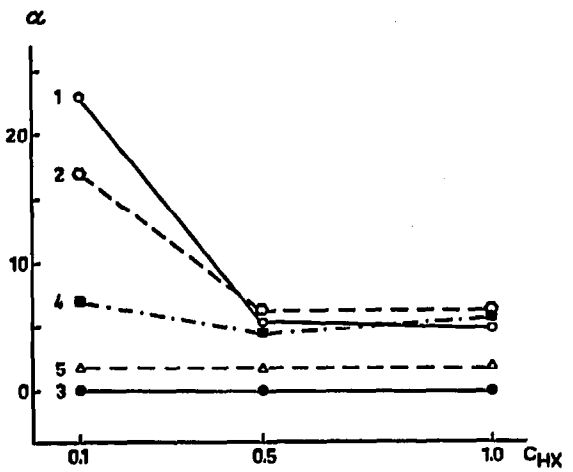
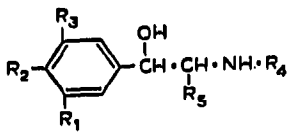


Fig. 5. Separation factors by reversed-phase chromatography with bis-(2-ethylhexyl)phosphoric acid as adduct-forming agent. Mobile phase: phosphate buffer, pH 3–5. Stationary phase: bis-(2-ethylhexyl)phosphoric acid (HX) in chloroform. Column: I.D. 4 mm, length 300 mm.
Sample:



No.	R_1	R_2	R_3	R_4	R_5
1	OH	H	OH	*tert.*-Butyl	H
2	OH	OH	H	Isopropyl	H
3	OH	OH	H	CH_3	H
4	H	OH	H	CH_3	CH_3
5	H	OH	H	CH_3	H
6	OH	H	H	H	H
7	H	OH	H	H	CH_3

efficiency than the straight-phase columns, *H* usually being about 2 mm. The stability of the column packing was not always satisfactory.

REFERENCES

1 R. Modin and G. Schill, *Acta Pharm. Suecica*, 4 (1967) 301.
2 K. O. Borg and D. Westerlund, *Z. Anal. Chem.*, 252 (1970) 275.
3 R. Modin, B.-A. Persson and G. Schill, in J. G. Gregory, B. Evans and P. C. Weston (Editors), *Proc. Int. Solvent Extr. Conf., 1971*, Vol. II, Society of Chemical Industry, London, 1971, p. 1211.
4 S. Eksborg and B.-A. Persson, *Acta Pharm. Suecica*, 8 (1971) 205.
5 K. Gustavii, *Acta Pharm. Suecica*, 4 (1967) 233.
6 R. Modin and A. Tilly, *Acta Pharm. Suecica*, 5 (1968) 311.
7 B.-A. Persson, *Acta Pharm. Suecica*, 5 (1968) 335.
8 G. Schill, R. Modin and B.-A. Persson, *Acta Pharm. Suecica*, 2 (1965) 119.
9 B. A. Persson and G. Schill, *Acta Pharm. Suecica*, 3 (1966) 281.
10 R. Modin and S. Bäck, *Acta Pharm. Suecica*, 8 (1971) 585.

11 G. Schill, *Advan. Ion Exchange Solvent Extr.*, 6 (1973) in press.
12 S. Eksborg and G. Schill, *Anal. Chem.*, in press.
13 B.-A. Persson, *Acta Pharm. Suecica*, 5 (1968) 343.
14 K. O. Borg and A. Mikaelsson, *Acta Pharm. Suecica*, 7 (1970) 673.
15 S. Eksborg and B.-A. Persson, *Acta Pharm. Suecica*, 8 (1971) 605.
16 K. O. Borg and G. Schill, *Acta Pharm. Suecica*, 5 (1968) 323.
17 K. Gustavii and G. Schill, *Acta Pharm. Suecica*, 3 (1966) 259.
18 K. O. Borg, *Acta Pharm. Suecica*, 6 (1969) 425.
19 K. G. Wahlund and K. Gröningsson, *Acta Pharm. Suecica*, 7 (1970) 615.
20 R. Modin and M. Johansson, *Acta Pharm. Suecica*, 8 (1971) 561.