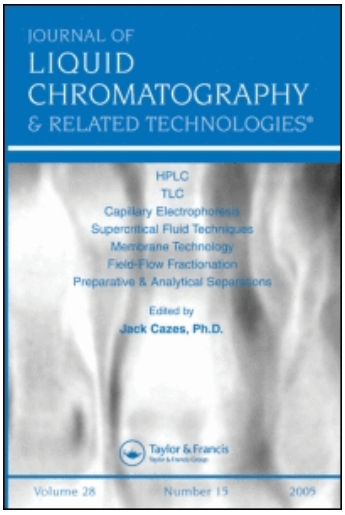

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SOLVENT EFFECTS IN REVERSED PHASE LIQUID CHROMATOGRAPHY FOR HIGH AND LOW LOADINGS OF OCTADECYLSILANE ON MICROPARTICLE SILICA. I.

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ABSTRACT

The selectivity of two ODS bonded phase packings, one with a high carbon load, the other with a low carbon load, were investigated. Many organic solvent/water mobile phases were used with three sample mixtures of varying polarity. When the separations were normalized, trends in the solubility parameters of the various classes of organic solvents were noted. Analyzing the separation factors and considering possible mobile phase, solute, and bonded phase interactions furnished some insight into possible separation/selectivity mechanisms.

INTRODUCTION

More liquid chromatographic separations than ever are being done today on bonded reversed phase packings. The factors most responsible for this trend are: a) ease of mobile phase selection; b) long life of such columns because they retain fewer impurities and are easily cleaned; and, c) large number of compounds soluble in the polar mobile phases used. In spite of its ever-increasing use, little was known until recently about the mechanism of the reversed phase mode

or details of its mobile phase selectivity. Scott and Kucera[1] showed that there is a monolayer of the more nonpolar mobile phase component on the bonded nonpolar phase; thus, partitioning appears to be occurring between the mobile phase and this monolayer. In a study on hydrophobic effects in reversed phase by Karger, et al,[2] it was found that selectivity as a function of composition and organic solvents could be predicted. They also stated, however, "that functional group selectivity in reversed phase LC is dependent to some extent on the organic modifier of the mobile phase."

Some unusual solvent effects in reversed phase separations have been noted. Erni and Frei[3] found better separations of digitalis glycosides with a mixture of dioxane/acetonitrile/water than with either organic solvent alone with water. In separations of cardiotonic steroid conjugates, Shimada, et al,[4] found tetrahydrofuran/water more selective than methanol/water. In the separation of a caffeine-containing analgesic mixture, Baker[5] found that by varying the tetrahydrofuran in the THF/water/acetic acid mobile phase the retention of caffeine was changed. Butts[6] found that only the caffeine peak shifted when the same mobile phase was used for the separation of saccharin, sodium benzoate, and caffeine. No explanation has been offered for these unusual selectivities.

Another variable that must be considered in the mechanism of bonded reversed phase separations is the bonded

reversed phase packing itself. There are now a number of bonded reversed phase packings on the market. A number of workers have investigated the properties of several of these packings.[7-13] They have found significant differences in their performance, indicating that the various bonding techniques and base silica gels used give different products chromatographically. In general, it is found that increased carbon content or chain length lead to increased retention of a given solute under given mobile phase conditions. However, varying amounts of adsorption from non-bonded silanol groups can occur causing different ratios of adsorption/partition.

The present study was undertaken to attempt to provide: a) information on the chromatographic differences between two reversed phase packings; and, b) insight into the mobile phase selectivity in the reversed phase mode. The two packings used were Partisil-10 ODS, bonded with 5% ODS (octadecyl silane) and Partisil-10 ODS-2, bonded with 16% ODS. The physical properties of each packing are given in Table I.

Three groups of solutes of varying polarity were studied. The groups and compounds are given in Table II. The elution order in most systems is as given. Occasionally peak orders were shifted, and these will be mentioned in the discussion.

The eluotropic or solubility parameter tables, based upon the work of Hildebrand and Scott,[14,15] are approximate

TABLE I

Physical Properties of Partisil-10 ODS and ODS-2 Columns

Column	Partisil-10 ODS	Partisil-10 ODS-2
Surface Area, m^2/g	400	400
Carbon Length	18	18
Carbon Content, %	5	16
Dead Volume, ml	2.49	2.03
Density Dry Pack, g/cc	0.500	0.625
Density Slurry, g/cc	0.498	0.621
HETP, mm*	0.053	0.035

*Standard quality control test results: For Partisil-10 ODS the test mixture is benzene (k'=0.4), naphthalene (k'=0.7), and biphenyl (k'=1.0) - mobile phase MeOH/H_2O (70:30). For Partisil-10 ODS-2, the same mixture gives k' = 0.5, 1.0, 1.4 respectively - mobile phase MeOH/H_2O (90:10). HETP is the average for three peaks. Linear velocity ≈0.2cm/sec. Approximate particle diameter for lots of packing used was 8.5μm

measures of solvent polarity. However, "polarity" is really a combination of solvent effects. These effects consist of dispersion interactions, dipole interactions, and hydrogen donor or acceptor interactions. Many workers[16-20] have obtained numerical values for these contributions to "polarity" to characterize solvent properties, but much of this work relates only to adsorption systems. Because of the complexity

TABLE II

Samples Mixtures

Sample:	Low Polarity	Medium Polarity	High Polarity
Solutes:	benzene	acetophenone	phenol
	naphthalene	propiophenone	aniline
	biphenyl	methoxybenzene	nitrobenzene
	phenanthrene		ethyl benzoate

of the interactions, only the total solubility parameter was used in this work.

Because more solvent solubility parameters than were available from the Hildebrand tables were needed, the solubility parameters devised by Hoy[21] were used. These have been used by other workers[17,22] in similar work and, therefore, will allow useful cross-reference in the future. It has been questioned whether they can be used in high pressure systems,[17] but the values are adequate for first approximations. The Hoy solubility parameters for the organic solvents used in this study are given in Table III.

The calculation of the solubility parameter for the mobile phase, δ_{mp}, is given by the equation

$$\delta_{mp} = \phi_{H_2O} \ \delta_{H_2O} + \phi_{org} \ \delta_{org} \qquad (1)$$

where ϕ_{H_2O} = volume fraction H_2O

ϕ_{org} = volume fraction second solvent

TABLE III

Solubility Parameters (Hoy)[21]

Methanol	14.5
Ethanol	12.8
1-Propanol	12.2
2-Propanol	11.4
Acetonitrile	12.1
Tetrahydrofuran	9.4
Dimethoxyethane	8.6
Dioxane	10.1
Ethylene glycol	17.1
Ethylene glycol monomethyl ether	11.7

$$\delta_{H_2O} = 23.5^{(21)}$$

$$\delta_{org} = \delta \text{ of second solvent}^{(21)}$$

The values of the solubility parameter, δ_{mp}, as calculated for the mobile phases which yield normalized retentions, are given in Table IV.

The initial study with the low polarity solutes was done with 10 different organic solvent/water combinations as is seen from Table IV. As will be discussed, many organic solvents in combinations with water gave similar selectivities and separations. Thus, the balance of the study was done with only MeOH, EtOH, CH_3CN, THF, and

TABLE IV

Solvent Composition and Solubility Parameter of Mixed Solvent

	$k'_{benz} \simeq 1$				$k'_{acet} \simeq 1$				$k'_{phenol} = 1$			
	ODS		ODS-2		ODS		ODS-2		ODS		ODS-2	
	ϕ_{org}	δ_m	ϕ_{org}	δ_m	ϕ_{org}	δ_m	ϕ_{org}	δ_m	ϕ_{org}	δ_m	ϕ_{org}	δ_m
MeOH/H_2O	0.53	18.7	0.80	16.3	0.50	19.0	0.70	17.2	0.25	21.2	0.70	17.2
CH_3CN/H_2O	0.55	17.2	0.80	14.4	0.50	17.8	0.70	15.5	0.35	19.5	0.50	17.8
THF/H_2O	0.50	16.4	0.65	14.3	0.45	17.2	0.48	16.7	0.55	15.7	0.50	16.4
DME/H_2O	0.55	15.3	0.76	12.2	0.47	16.5	0.55	15.3	0.55	15.3	0.50	16.1
EtOH/H_2O	0.45	18.7	0.70	16.0	0.37	19.5	0.51	18.0	0.20	21.3	0.40	19.2
2-PrOH/H_2O	0.40	18.7	0.63	15.9								
1-PrOH/H_2O	0.37	19.3	0.60	16.7								
Diox/H_2O	0.50	16.8	0.70	14.1								
EtGl/H_2O	0.60	19.6	-	-								
EGME/H_2O	0.50	17.6	0.75	14.6								

DME = dimethoxyethane
EtGl = ethylene glycol
EGME = ethylene glycol monomethyl ether
Diox = p-dioxane
THF = tetrahydrofuran
benz = benzene
acet = acetophenone

Dimethyoxyethane/H_2O combinations. The first three mobile phases are often used in reversed phase LC and the latter two appear to have unique selectivities.

The solvent systems for each sample on each column were adjusted to give a $k' \approx 1$ for the first eluting solute in the sample. Thus, the normalized retention data could more easily and sensibly be compared between columns and mobile phases.

In order to compare the separations obtained with the various systems, the separation ratio, α, was calculated for each adjacent pair of peaks. The α values for all mobile phases and samples on Partisil-10 ODS and on Partisil-10 ODS-2 are given in Table VI.

EXPERIMENTAL

To determine the densities of the two packings, samples of each were: a) tap-packed into a 10cc graduate for the tap-pack density; and, b) packed at 10,000 psi into a 4.6mm ID x 25cm 316 tainless steel columns, which were then emptied and the contents weighed.

The carbon content was determined by combustion at a commercial laboratory. This is done with each batch of bonded phase to assure the reaction has proceeded as planned. The final test of its suitability is, of course, its chromatographic performance. Certain capacity ratio (k'), efficiency, and peak symmetry specifications must be obtained before any batch is released for commercial sale.

The Partisil-10 ODS and ODS-2 packings are packed with a Haskel pump at 10,000 psi from a $CHCl_3$/MeOH slurry. After packing, the columns are washed with 10ml methanol before being equilibrated to the mobile phase being used.

Two pumps were used at various times during the study, a Laboratory Data Control (LDC) Constametric IIG, and a Spectra-Physics Model 4800A. A Valco valve (Universal 7000 psi) for making injections, an LDC Spectromonitor II variable wavelength detector set at 254 nm, and either a Varian A-25 or LDC Model 3401 recorder were used during the study.

Three sample mixes were used during the study. The low polarity sample consisted of benzene (10mg/ml), naphthalene (1mg/ml), biphenyl (0.25mg/ml), and phenanthrene (0.50mg/ml). The medium polarity sample was made up of acetophenone (0.13mg/ml), propiophenone (0.13mg/ml), and anisole (2mg/ml). The high polarity sample consisted of phenol (4mg/ml), aniline (2mg/ml), nitrobenzene (5mg/ml), and ethyl benzoate (8mg/ml). The samples were all dissolved in methanol.

All of the solvents were either Burdick and Jackson distilled-in-glass or reagent grade solvents. Distilled water was used for all mobile phases. No extra clean-up of solvents was undertaken.

To prevent bubble formation, each mobile phase, after having been made up by volume-volume addition, was degassed for 7 minutes under water aspiration at ambient temperature. Since the organic component is more volatile, some will

necessarily vaporize from the mobile phase. The degassing time/conditions must therefore be carefully controlled for reproducibility.

DISCUSSION

On examining the physical properties of Partisil-10 ODS and ODS-2 from Table I, it can be seen that the Partisil-10 ODS-2 density is about 20% greater than the density of Partisil-10 ODS. One might have expected the ODS-2 with the 16% loading to be bulkier and less dense than the ODS with the 5% loading.

The density of either packing is the same whether it was dry or slurry packed. As theory and experience have shown,[23-27] slurry packing of any microparticle material gives more efficent columns than dry packing. This is presumably because a greater bed density is obtained at high pressures. It is postulated that the orientation of the particles or swelling of the bonded phase may be responsible for the greater efficiency of the slurry packed columns.

Table I also shows that the HETP is smaller for the Partisil-10 ODS-2 than for the Partisil-10 ODS column. The smaller value for HETP is probably due to the increased density of the Partisil-10 ODS-2, as is shown by the smaller V_o of the ODS-2 column. The average increase in column efficiency of ODS-2 over ODS is 25%.

The chromatographic effects of the high and low carbon loadings using the same mobile phase composition can be seen in Figure 1. As expected, with a higher ODS content, the polyaromatic hydrocarbons are retained to a greater extent.

Figure 2 shows the change in the capacity ratio (k') of the low polarity solutes when higher chain length alcohols are used in the mobile phase. There is no change in the elution order of the solutes in these alcohol/water systems. Although not an alcohol, acetonitrile fits fairly well in the alcohol series with eluting powers between that of methanol and ethanol.

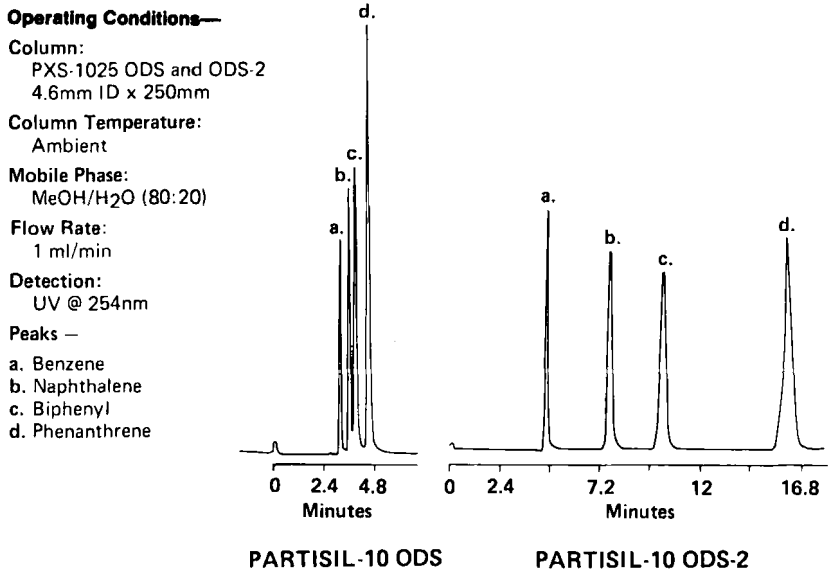

FIGURE 1

Comparison chromatograms on Partisil-10 ODS and Partisil-10 ODS-2.

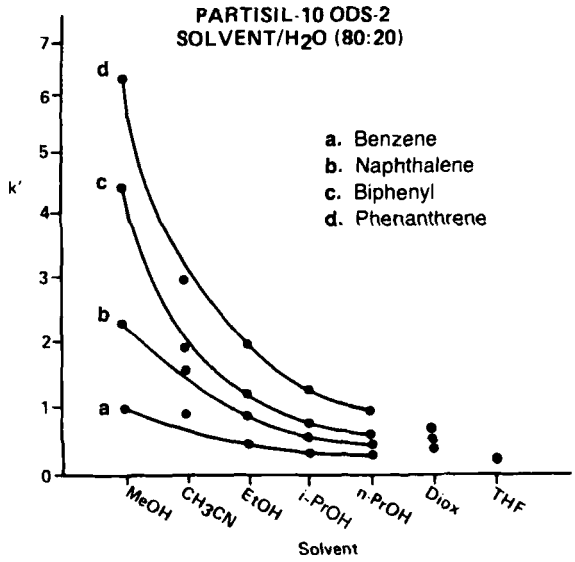

FIGURE 2

Capacity factors (k') of low polarity solutes with various organic solvents.

The slight difference in selectivity of acetonitrile is seen in the data points which do not lie on the curves for each compound.

At the 80:20 composition, dioxane/water and tetrahydrofuran/water cause some or all of the peaks to overlap; thus these organic solvents are too nonpolar. By lowering the percent organic component, separation of the low polarity solutes was obtained. The α values for these separations are given in Table VI. As this Table and Figure 3 show, tetrahydrofuran/water will not separate the biphenyl from phenanthrene on Partisil-10 ODS and elutes biphenyl after

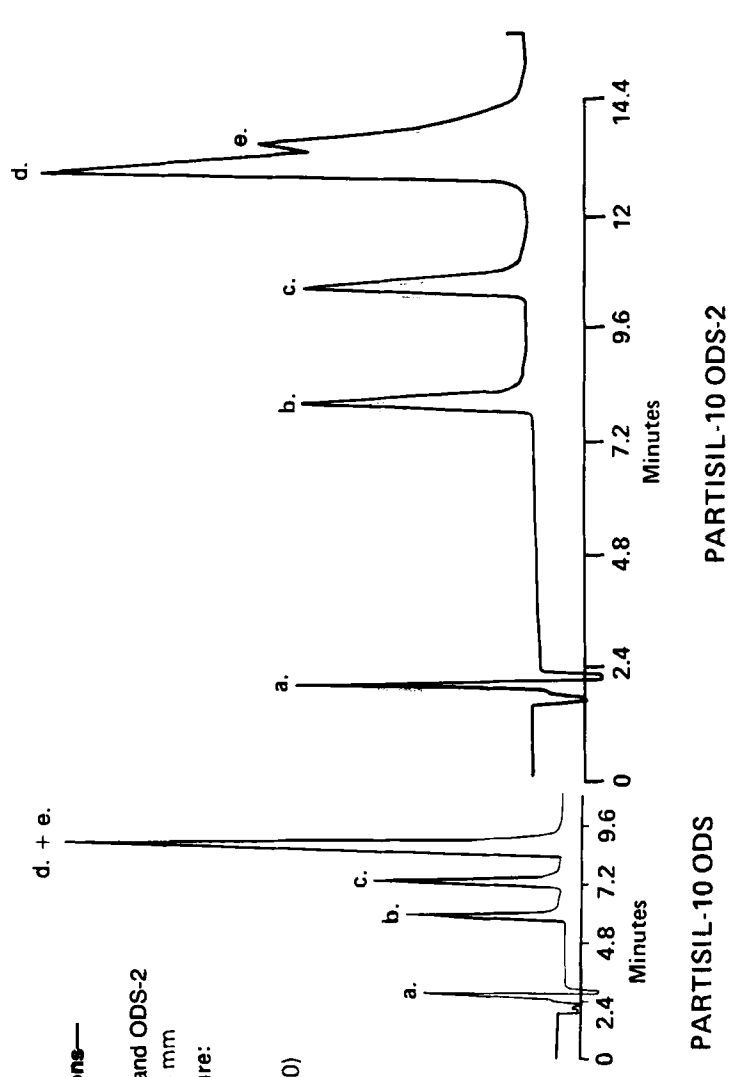

FIGURE 3

Separation of hydrocarbons on Partisil-10 ODS and ODS-2 with THF/water.

phenanthrene on Partisil-10 ODS-2. This elution order is the reverse of that found for these two peaks with any other mobile phase on either packing.

When all the separations were normalized (so that the k' of the first eluting solute equaled 1), the ϕ_{org} and δ_{mp} values were calculated. (Table IV) The δ_{mp} values are quite similar for each mobile phase made up of any alcohol/water on either packing material with normalized retention of the test solutes. Table V shows the average δ_{mp} values for molecule phases containing alcohols and ethers for the low polarity sample on both Partisil-10 ODS and ODS-2.

Since the bonded phase is nonpolar, its predominant interaction with solutes of low polarity is dispersive, and there is more nonpolar bonded phase on Partisil-10 ODS-2, it will exhibit more dispersive interactions. To overcome these increased interactions, the mobile phases for Partisil-10 ODS-2 separations must contain more of the organic

TABLE V

δ_{mp} Averages for Different Classes of Solvents

(Sample: Low Polarity)

	#	Column: δ_{mp}	ODS SD	#	Column: δ_{mp}	ODS-2 SD
Alcohols	6	18.8	0.7	5	15.9	0.8
Ethers	3	16.2	0.8	3	13.5	1.2
CH_3CN		17.2			14.4	

component, yielding smaller δ_{mp} values, than for the same normalized separation on Partisil-10 ODS.

The trend of smaller δ_{mp} values for ODS-2 over ODS separations holds true for all mobile phase combinations for the low polarity and the medium polarity samples. This trend is also apparent for methanol/water, acetonitrile/water, and ethanol/water combinations with the high polarity sample. However, with tetrahydrofuran/water or dimethoxyethane/water with the high polarity sample, the δ_{mp} for ODS is smaller than for ODS-2. These two ethers must interact differently with the solutes or packings during the chromatographic process than do methanol, ethanol, or acetonitrile.

The general trend of increasing δ_{mp} with increased solute polarity is observed for either packing. With average δ_{mp} values as given in Table V, it is possible to rearrange equation (1) to obtain the equations

$$\frac{\delta_{mp} - \delta_{org}}{23.5 - S_{org}} = \phi_{H_2O} \qquad (2)$$

$$\frac{23.5 - \delta_{mp}}{23.5 - \delta_{org}} = \phi_{org} \qquad (3)$$

These equations can be used to calculate the approximate mobile phase composition which will give solute retention under these normalized conditions for low polarity solutes. Using estimates of higher δ_{mp} values (as indicated from Table V),

the approximate mobile phase compositions for the normalized separation of more polar solutes can also be calculated. Although useful, more information regarding selectivity is needed to choose between the many organic solvent/water combinations.

The data in Table VI relating the separation factors, α, for various mobile phases on ODS and ODS-2 indicate possible selectivity effects. If the low polarity sample is being separated on the nonpolar bonded phase, the only possible interactions between mobile phase, bonded phase, and solute are dispersion interactions. As the chain length of the alcohol in the mobile phase increases, the alcohol will introduce larger and competing dispersion interactions. As a result, the low polarity components will not be separated as well from one another with a mobile phase of longer chain alcohols; e.g., methanol/water separates the low polarity solutes from one another better than other alcohol/water mixtures. The separation of the low polarity solutes can also be enhanced if more polar interactions are introduced into the chromatographic system. In going from ethanol/water to ethylene glycol/water on Partisil-10 ODS, the α values increase. Further work on propanol/water, propylene glycol/water, and glycerine/water mobile phases is now underway to investigate this trend.

In separating the moderate polarity sample, $\alpha_{1,2}$ increases on ODS and decreases on ODS-2 when going from MeOH to EtOH.

TABLE VI

Separation Factors, α, on Partisil-10 ODS and ODS-2
With Various Mobile Phases

($\alpha_{\text{Partisil-10 ODS}}/\alpha_{\text{Partisil-10 ODS-2}}$)

Mobile Phase/Sample	$k'_{benzene} = 1$			$k'_{acetophenone} = 1$		$k'_{phenol} = 1$		
	$\alpha_{1,2}$	$\alpha_{2,3}$	$\alpha_{3,4}$	$\alpha_{1,2}$	$\alpha_{2,3}$	$\alpha_{1,2}$	$\alpha_{2,3}$	$\alpha_{3,4}$
MeOH/H_2O	3.15/2.34	1.82/1.47	1.73/1.76	1.24/1.68	1.32/1.10	1.31/1.26	2.67/2.04	4.27/1.87
CH_3CN/H_2O	1.63/1.75	1.27/1.24	1.33/1.52	1.28/1.52	1.00/1.00	1.47/1.56	1.43/2.04	2.02/1.87
THF/H_2O	1.43/1.14	1.32/1.22	1.00/1.07	1.08/2.04	1.62/1.30	1.00/1.00	1.58/2.75	1.00/1.20
DiM/H_2O	1.68/1.22	1.56/1.20	1.00/1.04	1.76/2.33	1.12/1.24	1.74/2.75	1.00/2.48	1.11/3.20
EtOH/H_2O	2.59/2.04	1.66/1.40	1.41/1.57	1.60/1.60	1.00/1.47	1.22/1.46	2.63/2.88	3.90/2.78
1-PrOH/H_2O	1.73/1.75	1.31/1.24	1.24/1.56					
2-PrOH/H_2O	2.07/1.92	1.44/1.30	1.27/1.60					
Diox/H_2O	2.05/1.41	1.52/1.26	1.22/1.09					
EtGl/H_2O	3.41/ -	2.04/ -	1.67/ -					
EGME/H_2O	2.69/1.60	1.76/1.44	1.28/1.14					

Greater dispersion interactions are introduced which appear to aid the separation of acetophenone and propiophenone on ODS but hinder this separation on ODS-2. The reverse effects are occurring for the separation of propiophenone and methoxybenzene ($\alpha_{2,3}$) where other selectivities apparently operate because of the difference in the carboxy and ether moieties. The reversing of these effects might be expected in the separation of moderately polar solutes because of more subtle balancing of solvent effects.

In separating the high polarity sample, $\alpha_{1,2}$ and $\alpha_{3,4}$ decreases ($\alpha_{2,3}$ remains the same) on ODS and all three α values increase on ODS-2 when going from MeOH to EtOH. Greater dispersion interactions appear to hinder the separation of these polar solutes on ODS, but aids their separation on ODS-2. Perhaps the explanation of this observation lies in the reversed phase mechanism of Scott and Kucera.[1]

On ODS-2, more bonded phase will attract a greater amount of the organic component of the mobile phase. Likewise, the longer the chain length of the alcohol, the greater the amount of the alcohol to be attracted as a stationary liquid phase. Although ODS with its low carbon content would attract less of the organic component of the mobile phases, perhaps the greater polar interactions present allow methanol to be attracted far in excess of its capacity based upon carbon content alone.

If the separation efficiency can be based upon the amounts of the "attracted" organic solvent, then each packing in its turn can more effectively perform the separation. Of interest, too, are the subtle solvent effects observed in a few of the separations. For instance, the reversal of elution order in THF containing mobile phases, and the significantly large α values when separating the high polarity sample with dimethoxyethane/water on Partisil-10 ODS-2 than on Partisil-10 ODS. Hopefully, continuing studies of solvent effects in reversed phase separations will allow more definitive conclusions to be drawn as to mechanism.

From this study, however, there are generally larger α values, hence better resolution, for low polarity solutes when separated on Partisil-10 ODS. Conversely, there is a trend to higher α values for moderate to high polarity solutes when separated on Partisil-10 ODS-2. Thus, both high and low loaded bonded ODS microparticle packings can provide unique selectivities in addition to simple changes in retention.

REFERENCES

1. Scott, R.P.W., and Kucera, P. The Examination of Five Commercially Available Liquid Chromatographic Reverse Phases (Including the Nature of the Solute/Solvent/ Stationary Phase Interactions Associated with Them). J. Chromatogr., 142, 213 (1977).

2. Karger, B.L., Gant, J.R., Hartkopf, A., and Weiner, P.H. Hydrophobic Effects in Reversed Phase Liquid Chromatography. J. Chromatogr., 128, 65 (1976).

3. Erni, F., and Frei, R.W. A Comparison of Reversed Phase and Partition High Performance Liquid Chromatography of Some Digitalis Glycosides. J. Chromatogr., 130, 169 (1977).

4. Shimada, K., Hasegawa, M., Hasebe, K., Fujii, Y., and Nambara, T. Studies on Steroids CXIV. Separation of Cardiotonic Steroid Conjugates by High Performance Liquid Chromatography. J. Chromatogr., 124, 79 (1976).

5. Baker, D.R., ASTM Committee E-19 Meeting, Chicago, October, 1976.

6. Butts, E.T., personal communication, 1977.

7. Karch, K., Sebestian, I., and Halasz, I. Preparation and Properties of Reversed Phases. J. Chromatogr., 122, 3 (1976).

8. Unger, K.K., Becker, N., and Roumeliotis, P. Recent Developments in the Evaluation of Chemically Bonded Silica Packings for Liquid Chromatography. J. Chromatogr., 125, 115 (1976).

9. Majors, R.E., and Hopper, M.J. Studies of Siloxane Phases Bonded to Silica Gel for Use in High Performance Liquid Chromatography. J. Chromatogr. Sci., 12, 767 (1974).

10. Done, J.N. Sample Loading and Efficiency in Adsorption, Partition, and Bonded-Phase High Speed Liquid Chromatography. J. Chromatogr., 125, 43 (1976).

11. Kikta, E.J., and Grushka, E. Retention Behavior on Alkyl Bonded Stationary Phases in Liquid Chromatography. Anal. Chem., 48, 1098 (1976).

12. Hemetsberger, H., Maasfeld, W., and Ricken, H. The Effect of Chain Length of Bonded Organic Phases in Reversed Phase High Performance Liquid Chromatography. Chromatographia, 9, 303 (1976).

13. Karch, K., Sebestian, I., Halasz, I., and Engelhardt, H. Optimization of Reversed Phase Separations. J. Chromatogr., 122, 171 (1976).

14. Hildebrand, J.H., and Scott, R.L., Regular Solutions, Prentice Hall, Englewood Cliffs, N.J., 1962.

15. Hildebrand, J.H., and Scott, R.L., Solubility of Non-Electrolytes, Dover, N.Y., 1964.

16. Mikes, O., and Vespalec, R., in Liquid Column Chromatography - A Survey of Modern Techniques and Applications, Z. Deyl, K. Macek, and J. Janak (Editors), Elsevier, N.Y., 1975, pp. 233-280.

17. Tijssen, R., Billiet, H.A.H., and Schoenmakers, P.J. Use of the Solubility Parameter for Predicting Selectivity and Retention in Chromatography. J. Chromatogr., 122, 185 (1976).

18. Karger, B.L., Snyder, L.R., and Eon, E. An Expanded Solubility Parameter Treatment for Classification and Use of Chromatographic Solvents and Adsorbents. Parameters for Dispersion, Dipole and Hydrogen Bonding Interactions. J. Chromatogr., 125, 71 (1976).

19. Snyder, L.R. Classification of the Solvent Properties of Common Liquids. J. Chromatogr., 92, 223 (1974).

20. Bakalyar, S.R. Solvent Selectivity in Reversed Phase LC, in Chromatographic Reviews, Spectra-Physics, 1977.

21. Hoy, K.L. New Values of the Solubility Parameters from Vapor Pressure Data. J. Paint Technol., 42, 76 (1970).

22. Gere, D.R., ASTM Committee E-19 Meeting, Chicago, October, 1976.

23. Giddings, J.C. Dynamics of Chromatography, Part 1, Principles and Theory, Marcel Dekker, N.Y., 1965.

24. Majors, R.E. High Performance Liquid Chromatography on Small Particle Silica Gel. Anal. Chem., 44, 1722 (1972).

25. Kirkland, J.J. High Performance Liquid Chromatography with Porous Silica Microspheres. J. Chromatogr. Sci., 10, 593, (1972).

26. Majors, R.E. Effect of Particle Size on Column Efficiency in Liquid-Solid Chromatography. J. Chromatogr. Sci., 11, 88 (1973).

27. Cassidy, R.M., LeGay, D.S., and Frei, R.W. Study of Packing Techniques for Small Particle Silica Gels by High Speed Liquid Chromatography. Anal. Chem., 46, 340 (1974).