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ADSORPTION ISOTHERMS ON SILICA FOR METHANOL AND 1-HEXANOL MODIFIERS FROM SUPERCRITICAL CARBON DIOXIDE

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SUMMARY

Adsorption isotherms were determined for methanol and 1-hexanol on silica from supercritical carbon dioxide at four temperatures and three mobile phase densities. Maximum stationary phase concentrations were extrapolated from linear least squares fits of the data to the Langmuir equation for monolayer adsorption. From these results, maximum surface area coverages were calculated using the mean molecular area of each modifier. The maximum, molar stationary phase concentration of methanol was found to exceed that of 1-hexanol under all experimental conditions; however, in each case the surface area coverage by hexanol was calculated to be larger.

The capacity factors of several substituted and unsubstituted aromatic hydrocarbons were determined in 0–1% (w/v) methanol modifier carbon dioxide. From the linearity of capacity factor *versus* modifier concentration plots, the ability of the solutes to compete with methanol for active column sites was determined. Unsubstituted aromatic hydrocarbons do not appear to compete with the modifier for direct adsorption onto modifier-sorbing active sites.

INTRODUCTION

Modifiers have been widely used in supercritical-fluid chromatography (SFC) to increase the solvent strength of the mobile phase, enhance selectivity, and improve peak shape and column efficiency[1–12,14–16]. With packed columns, the effects of modifier addition on these parameters may result from changes in the nature of the stationary phase surface because of localized adsorption of the modifier onto active stationary phase sites, *e.g.*, silanols, or solvation of bonded phase moieties, as well as changes in the physical properties of the mobile phase. Localized adsorption on active sites is believed to be primarily responsible for the changes in retention and selectivity observed with packed columns at low mobile phase modifier concentrations[6,7,11]. The extent of these changes depends on the type of solute, modifier and stationary phase examined. Several comparisons of methanol and hexanol as mobile phase modifiers in carbon dioxide have been reported. The retention of polycyclic aromatic and nitroaromatic hydrocarbons on ODS-modified silica columns was reported to be lower using 1-hexanol as the modifier in comparison to methanol at equivalent mobile phase concentrations[7]. This was attributed to better masking of active sites on the

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silica surface by the longer, lipophilic chain of 1-hexanol. The retention of 4-nitroaniline on a cyano-modified silica column was also reported to decrease with increasing length of the alkyl chain of the alcohol modifier[1]. This was suggested as being due to better access to active silanol sites and bonded cyano groups of the cyano column by the more lipophilic alcohols. Methanol, in contrast, was shown to decrease the retention of aromatic hydrocarbons on a diol-modified silica column more effectively than 1-hexanol[3]. This was thought to be the result of a more efficient interaction of the smaller methanol modifier with active column sites. Variations in temperature and mobile phase density can also have an effect on the degree to which a modifier interacts with an alters the stationary phase surface. The effect of mobile phase density on the adsorption of ethyl acetate modifier onto silica from supercritical carbon dioxide has been reported[17].

In liquid chromatography, localized adsorption of the modifier has been shown to have a direct effect on the retention of solutes which compete with the modifier for active column sites[18]. Over the modifier concentration range in which localized surface coverage by the modifier is constant, the capacity factor of a solute can be related to the mobile phase modifier concentration using the equation

$$1/k' = a\phi + b \tag{1}$$

where k' is the capacity factor, ϕ is the concentration of modifier in the mobile phase, and a and b are constants. A plot of inverse capacity factor *versus* mobile phase modifier concentration is predicted to be linear. Over the modifier concentration range in which localized surface coverage by the modifier reaches completion, solutes that absorb through localized interactions with active column sites but do not displace the adsorbed modifier from these sites will show a change in the slope of the line determined from the above equation. Non-linearity results because the interaction energy of the solute with the modified surface will be different than that of the solute with the unmodified surface. Snyder and Glajch[19] have suggested that localized adsorption by the modifier is essentially complete at approximately 75% of the total surface coverage. Surface coverage beyond that point must involve delocalized adsorption due to steric restrictions. On the other hand, solutes which do not adsorb via localized interactions with active column sites and solutes which can displace the modifier from the stationary phase surface and interact directly with these active sites, should show linearity over the same concentration range.

In this work, the peak maxima method[20] was used to determine adsorption isotherms for methanol and hexanol modifiers onto silica from supercritical carbon dioxide as a function of mobile phase density and temperature. In addition the effects of low concentrations of methanol modifier (0–1%, w/w) on the retention of several substituted and unsubstituted aromatic hydrocarbons on silica from supercritical carbon dioxide were examined. From these results, the propensity of the solutes to compete with methanol for active column sites was also determined.

EXPERIMENTAL

Carbon dioxide was supercritical fluid grade from Scott Speciality Gases (Plumsteadville, PA, U.S.A.). The detector was a Varian Vari-Chrom multiwave-

length detector fitted with a high-pressure UV cell supplied by Hewlett-Packard. The recorder was a Spectra-Physics Model SP4290 integrator.

The experimental set-up used in the determination of adsorption isotherms was described earlier[17]. The column was 10 cm × 4.6 mm I.D. and packed with Whatman Partisil-10 using the upward slurry technique. Methanol was HPLC-grade from Mallinckrodt (Paris, KY, U.S.A.). 1-Hexanol (98%) was obtained from Aldrich (Milwaukee, WI, U.S.A.). Both solvents were dried over 3-Å molecular sieves and distilled. Detection wavelengths for both alcohols were 190 nm at modifier concentrations below 0.10% (g/ml) to achieve adequate sensitivity and 206 nm at higher concentrations to maintain a linear detector response.

Retention data was obtained at 50°C at a carbon dioxide density of 0.60 g/ml. The dual pumping system of a Hewlett-Packard Model 1082B modified liquid chromatograph was used to produce the necessary mobile phase mixtures of carbon dioxide and 1.0% (w/w) methanol in carbon dioxide (Scott Speciality Gases). Capacity factors were measured in the usual manner from retention time (t_R) and dead time (t_0) as $k' = (t_R - t_0)/t_0$, and were reproducible to $\pm 1.5\%$.

RESULTS

Adsorption isotherms for methanol and 1-hexanol were determined at four temperatures and three mobile phase densities as listed in Table I. Examples of these isotherms are shown in Fig. 1. The isotherm data was fitted to a linear equation derived from rearrangement of the Langmuir function for monolayer adsorption:

$$1/C_s = 1/(C_m K^\circ C_s') + 1/C_s' \quad (2)$$

Where C_s is the stationary phase modifier concentration, C_m is the mobile phase concentration, C_s' is the maximum stationary phase concentration, and K° is the thermodynamic equilibrium constant for the distribution of the modifier between the adsorbed and non-adsorbed states. In all cases the correlation coefficient was not less

TABLE I

MAXIMUM MODIFIER CONCENTRATION AND SURFACE COVERAGE WITH TEMPERATURE AND DENSITY

Temperature (°C)	*CO_2 density (g/ml)*	*Maximum stationary phase concentration (%mol/g)*		*Maximum surface coverage[a] (m^2/g)*	
		Methanol	*Hexanol*	*Methanol*	*Hexanol*
40	0.60	0.221	0.136	240	314
60	0.60	0.182	0.141	197	326
80	0.60	0.159	0.130	172	301
100	0.60	0.162	0.112	175	259
60	0.40	0.180	0.117	195	270
60	0.50	0.167	–	181	–
60	0.70	0.169	–	183	–
60	0.80	0.171	0.130	185	301

[a] Mean molecular areas[21]: methanol = 18.0 Å²; 1-hexanol = 38.4 Å².

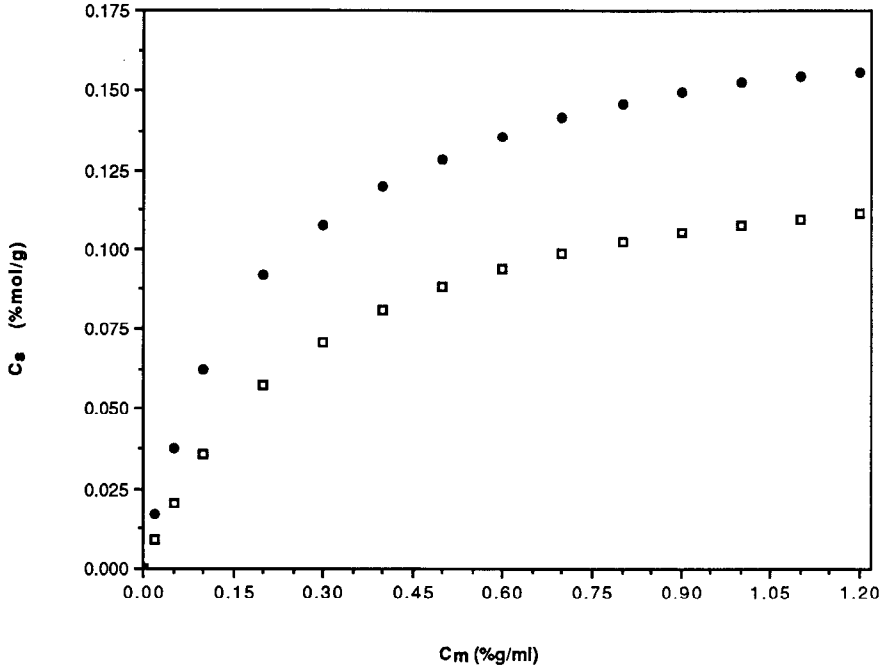

Fig. 1. Adsorption isotherms determined at 60°C at a CO_2 density of 0.60 (g/ml). Key: ⅛ = methanol; □ = hexanol.

than 0.9998. The least-squares fits to these data were used to extrapolate the maximum surface coverage for each modifier. These results as well as the maximum surface area covered by the modifiers calculated from their mean molecular areas[21] are listed in Table I. Under all temperature and density conditions examined, surface coverage by each modifier was more than 60% complete, based on the above calculations, at mobile phase concentrations of less than 1.5% (g/ml), and greater than 90% complete at this mobile phase concentration at 40°C. The importance of reporting concentration units in SFC work is overlooked in many published studies. This is especially important when comparing the effects of surface coverage on retention at low modifier concentrations, *e.g.*, comparing the isotherms in Fig. 1 at equivalent molar mobile phase concentrations, the stationary phase concentration of 1-hexanol increases more rapidly than that of methanol as determined by the difference in their molecular weights.

The surface coverage by each modifier was also examined as a function of temperature at constant mobile phase density. Plots of stationary phase modifier concentration *versus* temperature are shown in Fig. 2. The change in surface coverage was found to be linear with temperature at all mobile phase concentrations examined with correlation coefficients of 0.980–0.999. The rate of change in surface coverage with temperature at equivalent molar mobile phase concentrations was found to be the same for both modifiers.

Capacity factors were determined for several substituted and unsubstituted aromatic hydrocarbons as a function of mobile phase methanol concentration as

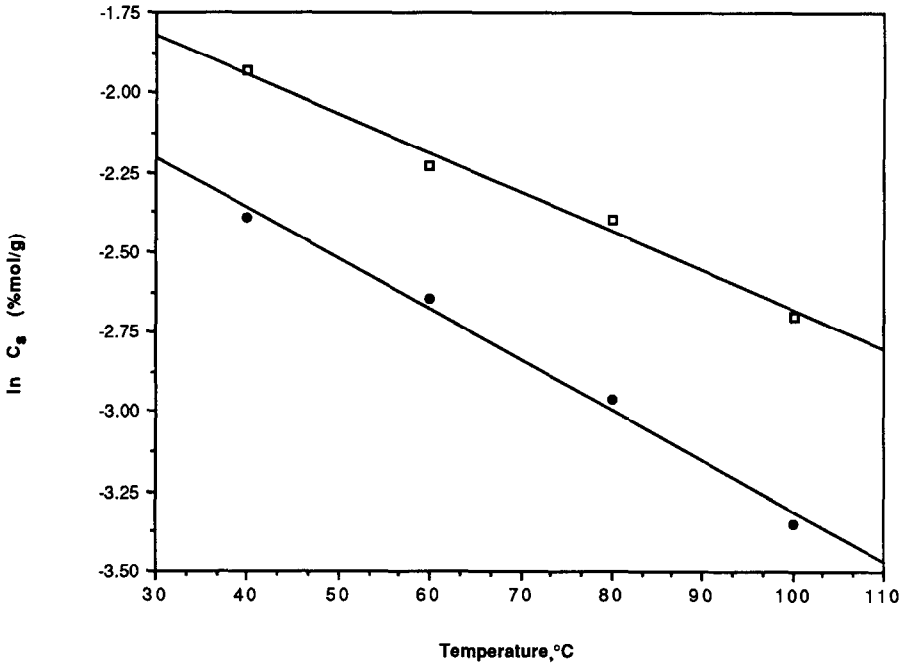

Fig. 2. Stationary phase coverage plotted *versus* temperature at a mobile phase concentration of 0.30% (g/ml) at a CO_2 density of 0.60 (g/ml). Key: □ = methanol, r = 0.994; ⅛ = hexanol, r = 0.995.

shown in Table II. Plots of inverse capacity factor *versus* mobile phase modifier concentration are shown in Figs. 3–6. A linear least squares fit was made to the data using the four largest modifier concentrations measured, at which the surface coverage changes slowly relative to lower concentrations. Eqn. 1 was then used to predict the capacity factors at zero modifier concentration. The calculated values of the capacity factor for solutes that compete with the modifier for active column sites will be lower than the actual values. This is expected since the availability of silanols increases rapidly as the mobile phase modifier concentration approaches zero resulting in an increase in the interaction energy of the solute with the stationary phase. This type of retention behavior was exhibited by methoxynaphthalene and nitronaphthalene as shown in Fig. 3, and for chloromethylnaphthalene as shown in Fig. 4. For direct comparison of solute retention and modifier surface coverage, a methanol adsorption isotherm, determined under the same experimental conditions, is shown in Fig. 4 overlaid on the inverse capacity factor plot of chloromethylnaphthalene. In contrast to the above solutes, the calculated value of the capacity factor of chloronaphthalene in Fig. 5 is larger than the actual value with a resulting maximum in k'. Similar retention behavior has been observed in this laboratory for aromatic hydrocarbons on silica using ethyl acetate modifier in carbon dioxide, and has also been reported for aromatic hydrocarbons from tetrahydrofuran modified carbon dioxide on several stationary phases[22]. This behavior is likely the result of increased dispersive interactions with modifier covered silanols relative to free silanols, or lateral interactions of the adsorbed solute with the adsorbed modifier as has been suggested

TABLE II

SOLUTE CAPACITY FACTORS *VERSUS* MOBILE PHASE METHANOL CONCENTRATION

%Methanol (g/ml)	*k'*							
	Phenol	*Chrysene*	*Naphthalene*	*1-Methoxy-naphthalene*	*2-Chloro-naphthalene*	*1-Nitro-naphthalene*	*1-Chloromethyl-naphthalene*	*Isobutyro-phenone*
0.00	12.3	5.40	0.534	1.93	0.679	6.50	2.21	10.1
0.10	11.3	5.38	0.527	1.71	0.667	6.35	1.96	9.49
0.20	9.60	4.95	0.488	1.40	0.661	3.46	1.46	3.84
0.30	8.50	4.70	0.497	1.28	0.667	2.77	1.32	2.77
0.40	7.55	4.29	0.461	1.23	0.648	2.51	1.16	1.82
0.60	6.35	3.83	0.431	1.13	0.596	2.18	1.03	1.18
0.80	5.35	3.40	0.394	1.03	0.569	1.88	0.946	0.916
1.00	4.76	3.08	0.375	0.971	0.544	1.76	0.857	0.795

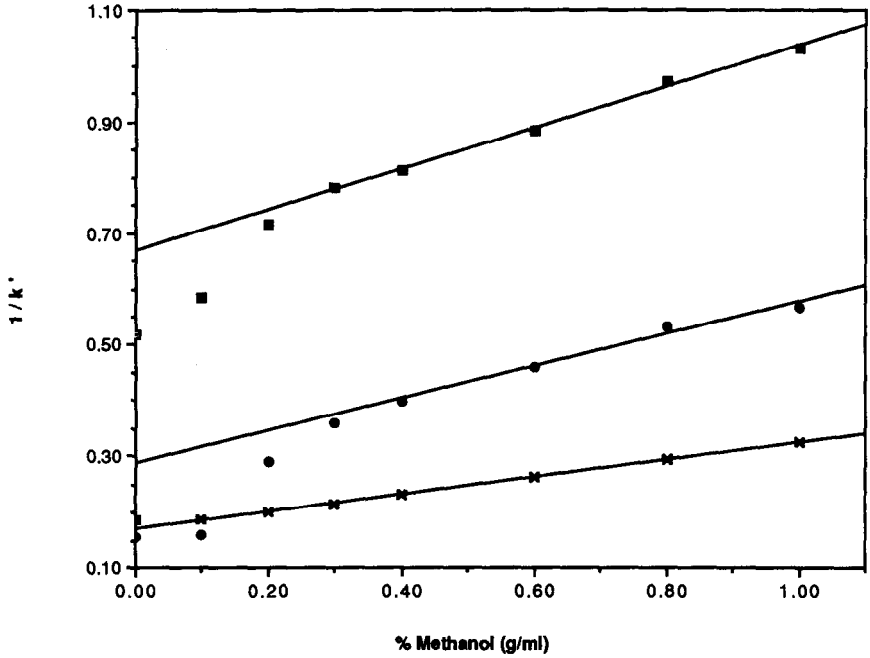

Fig. 3. Plots of inverse capacity factor of three solutes *versus* mobile phase methanol concentration at 50°C at a CO_2 density of 0.60 g/ml. Key: ⅜ = methoxynaphthalene; ⅛ = nitronaphthalene; × = chrysene.

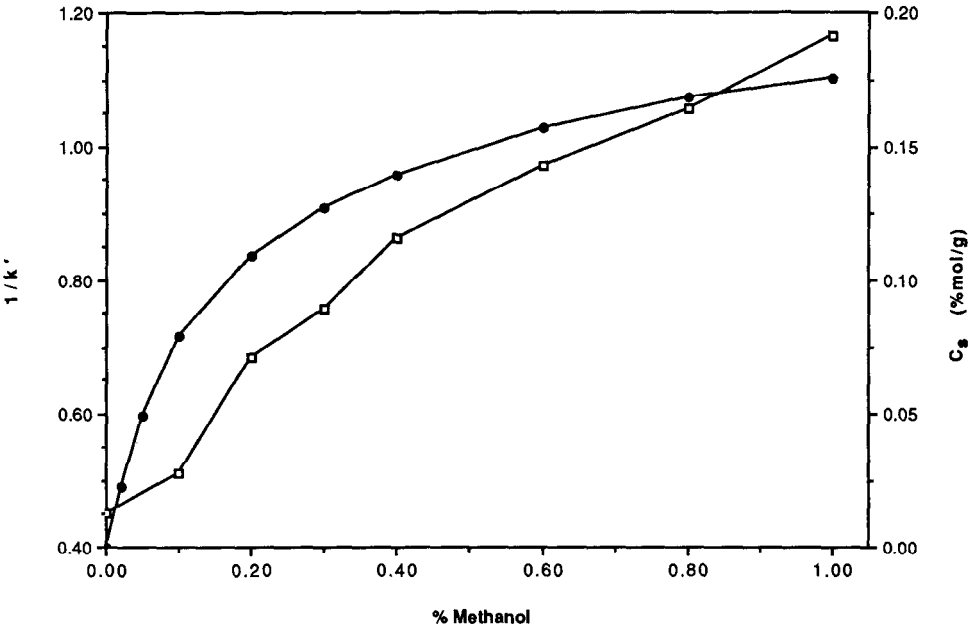

Fig. 4. Plot of inverse capacity factor of 1-chloromethylnaphthalene overlaid with an adsorption isotherm for methanol. Same conditions as in Fig. 3. Key: ⅛ = C_s; □ = $1/k'$.

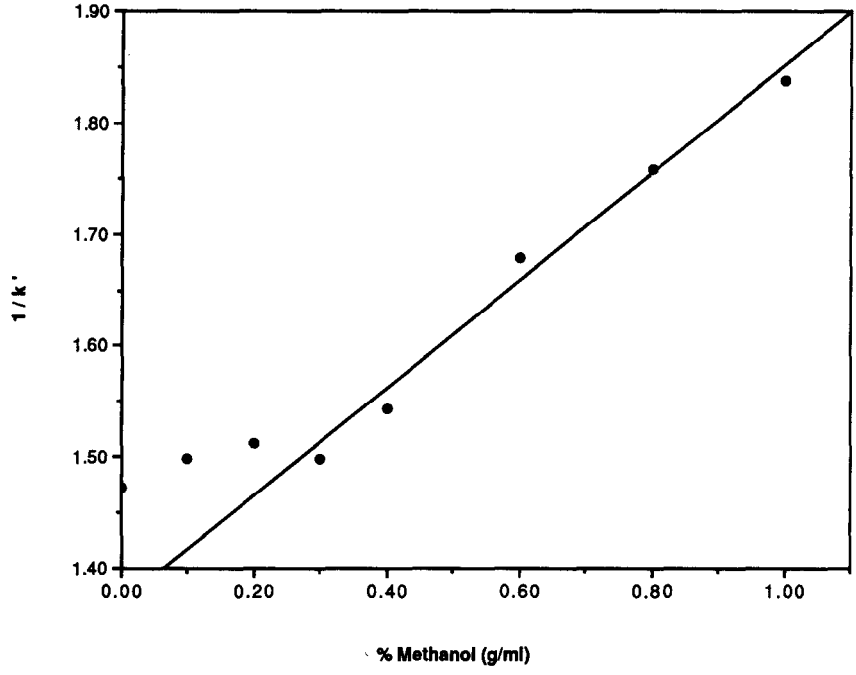

Fig. 5. Plot of inverse capacity factor of 2-chloronaphthalene *versus* mobile phase methanol concentration. Same conditions as in Fig. 3.

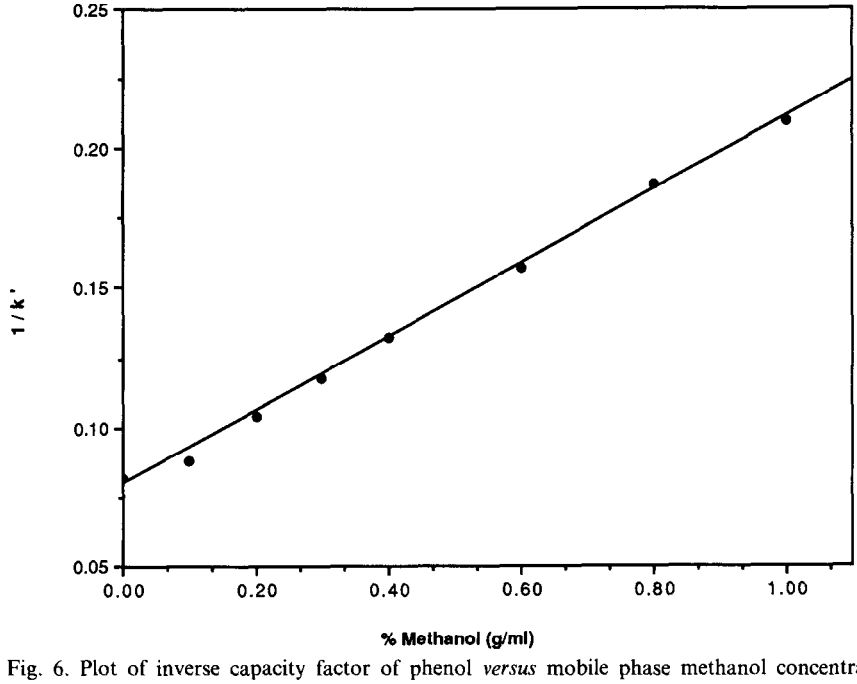

Fig. 6. Plot of inverse capacity factor of phenol *versus* mobile phase methanol concentration. Same conditions as in Fig. 3.

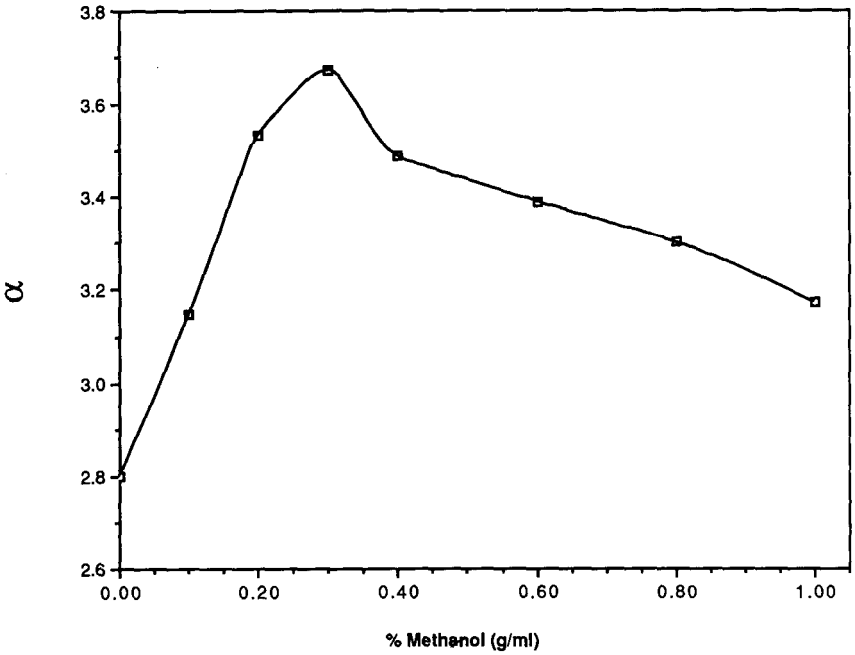

Fig. 7. Selectivity calculated for chrysene and 1-methoxynaphthalene plotted as a function of mobile phase methanol concentration.

for binary adsorption onto stationary phases in gas chromatography[23]. The unsubstituted aromatics, which likely do not adsorb through localized interaction with the silanols, show linearity throughout this concentration range as indicated for chrysene in Fig. 3. Phenol, which is expected to interact strongly with the silanols as suggested by its relatively large capacity factor, exhibits linearity, as shown in Fig. 6, probably because of its ability to displace methanol rather than compete with it for active sites[21].

The selectivity for a given solute pair can vary considerably over the modifier concentration range examined here. In the case of chrysene and methoxynaphthalene, in which one solute competes with the modifier for stationary phase active sites and the other does not, a maximum in selectivity is observed as shown in Fig. 7. As a result of its competition for active column sites, the capacity factor of methoxynaphthalene, which elutes first over this concentration range, decreases more rapidly than that of chrysene as the modifier is initially added to the mobile phase. As the active sites become covered with increasing modifier concentration, the capacity factor of chrysene decreases more rapidly than that of methoxynaphthalene with a resulting decrease in selectivity.

CONCLUSIONS

From the adsorption isotherms presented here and the values of surface coverage calculated from mean molecular surface area, 1-hexanol is apparently more

effective in masking stationary phase active sites than methanol. It must be noted, however, that the calculation of surface area coverage assumes that the molecules are spherical. This is probably a reasonable assumption for methanol, but the orientation of the adsorbed hexanol molecule, which has a greater length-to-breadth ratio, will have an effect on the apparent surface coverage. Orientation of the lipophilic chain away from the surface would increase its apparent coverage, while flat, horizontal, adsorption would have the opposite effect. Nevertheless, assuming that the actual orientation of the adsorbed hexanol molecules is somewhere between these extremes, calculations based on mean molecular area are a reasonable first approximation. The ability of 1-hexanol to restrict solute interaction relative to methanol will depend to some extent on the relative distribution of the modifiers between adsorbed and non-adsorbed states as a function of temperature and mobile phase density. The size, structure and functionality of the adsorbing solute, and its ability to intercalate between the longer lipophilic chains of 1-hexanol are also important.

The retention data presented here indicate that for certain solutes and functional groups competitive, solute–modifier, active site adsorption occurs in SFC with a carbon dioxide mobile phase. Non-substituted aromatics do not appear to interact directly with these alcohol-sorbing active sites under the conditions examined. The changes in retention for chrysene and naphthalene at low modifier concentrations are likely due to enhanced solubility of these solutes in the mobile phase and are not a response to the changing sorbent surface.

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