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COMPARISON OF POLYMER-BASED STATIONARY PHASES WITH SILICA-BASED STATIONARY PHASES IN REVERSED-PHASE LIQUID CHROMATOGRAPHY

SELECTIVE BINDING OF RIGID, COMPACT MOLECULES BY ALKYLATED POLYMER GELS

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SUMMARY

Alkylated stationary phases for reversed-phase liquid chromatography were prepared from polymer gels having hydroxyl groups, and their chromatographic properties were examined in terms of steric selectivity and preference for aromatic and saturated compounds. Polymer-based stationary phases were less hydrophobic than silica-based phases, but they showed preferential retention of aromatic compounds. The preference shown by the polymer-based stationary phases toward rigid, compact molecules over flexible and/or bulky molecules can be explained by the contribution of the polymer network structure to the retention process. The polymer-based stationary phases showed greater variation of selectivity due to changes in the composition of the mobile phase.

INTRODUCTION

Currently, chemically bonded silica packings are most widely used in high-performance liquid chromatography (HPLC). A number of advantages, including high performance, reproducibility, little irreversible adsorption and fast equilibrium with mobile phases, make alkylsilylated silica packings indispensable in reversed-phase liquid chromatography (RPLC).

However, silica-based materials do have limitations. They can only be used in a relatively narrow pH range, between 2 and 8. Secondary effects, such as those due to silanols and metal ions in the stationary phase, produce tailing of protonated amines and carboxylic acids[1,2]. Although these subjects have been extensively studied, there are still some unsolved problems of tailing, presumably caused by charged sites on recent packing materials based on silica gel[3]. With respect to chemical stability, some silica particles coated with a polymer layer have recently become available[4,5], which have a longer life at higher pH than conventional reversed-phase packings, but not as long as the organic polymer gels.

Considerable attention has been focused on polymer-based stationary phases

in RPLC. They are supposed to be free of charged-sites and stable in the pH range 2–12, or even wider. Although the conventional polymeric styrene–divinylbenzene (PS–DVB) gel used to give problems when used with aqueous eluents, current materials can be applied in separations with such mobile phases[6–14]. These materials are chemically stable and have been applied to biopolymer separations[12–14]. Such separations are most susceptible to secondary effects with silica-based phases, and need relatively severe elution conditions. Polymer-based stationary phases of much lower hydrophobicity has been used in hydrophobic interaction chromatography[14].

One potential limitation associated with the PS–DVB phase is the contribution of the aromatic functionality to chromatography, which may make elution patterns more complicated than those obtained with alkylsilylated silica gel[15]. In RPLC with a silica-based C_{18} stationary phase, the major factor in retention is the hydrophobic nature of the solutes, which is relatively easy to estimate from their structure. Recently, C_{18}-type packing materials based on PS–DVB, polyacrylamide and poly(vinyl alcohol) have become available[16–19], but detailed retention characteristics of these packing materials are not yet well elucidated.

It was thought to be desirable to synthesize alkyl-type phases, based on polymer particles with aliphatic backbones, and to examine their chromatographic properties. We report here a comparison of these polymer-based stationary phases with silica-based phases. Considerable differences were seen between the polymer-based and silica-based stationary phases in their selectivity toward hydrocarbons with various structural features, in size, rigidity, unsaturation and planarity.

EXPERIMENTAL

Materials

Silica-based C_1, C_8 and C_{18} phases were prepared from Develosil (Nomura Chemical, Seto, Japan) (5 μm, 330 m^2/g) by reacting the silica particles with alkyldimethylchlorosilane in toluene in the presence of pyridine at refluxing temperature[20]. As shown in Table I, all the stationary phases are maximally covered with alkylsilyl groups of specified chain length. The C_8 and C_{18} phases were further trimethylsilylated.

Hydrophilic, porous polymer gels, TSK G3000PW, G4000PW and G5000PW (Toyo Soda, Tokyo, Japan) (8–12 μm) and Asahipak GS520 (Asahi Chem Ind., Kawasaki, Japan) (9 μm) for aqueous-phase gel permeation chromatography (GPC) were kindly supplied by the manufacturers.

Asahipak is a cross-linked poly(vinyl alcohol). The structure of the cross-linking reagent is not disclosed. Wada[21] assumed that the cross-linking reagent is one of the isocyanurate derivatives shown in one of the patents applied for by the manufacturer. The GS520 gel possesses an exclusion limit of molecular weight 300 000 for pullulan in water. The pore size was estimated to be about 300 Å[16].

The chemical structure of the main constituent of TSK PW gels is not disclosed. IR spectra of this material indicated the existence of ester functionalities in addition to hydroxyl groups. ^{13}C NMR spectra showed the existence of carbon atoms in C–C, C–O and C=O linkages, but dit not show the existence of aromatic carbons. Therefore, the major constituent of PW gel is speculated to be a hydroxyalkyl ester of acrylic or methacrylic acid. No information on the structure and content of the

TABLE I

CARBON CONTENTS OF STATIONARY PHASES BASED ON POLYMER GEL AND SILICA
GEL

Stationary phase	Carbon content (%)			Surface coverage ($\mu mol/m^2$)
Silica-C_1	5.5			5.2
Silica-C_8	10.6*			3.5
Silica-C_{18}	18.2*			3.0
PW (native)	55.7			
PW-C_1	I 54.3**;	II	58.0	
PW-C_8	I 59.9**;	II	62.3	
PW-C_{18}	I 58.9**;	II	60.6	
GS (native)	54.7			
GS-C_1	56.6			
GS-C_8	63.5			
GS-C_{18}	66.1			

 * Prior to trimethylsilylation.
 ** Alkylated at 60°C.

cross-linking reagent of this material was available from the results. The 3000PW,
4000PW and 5000PW gels have exclusion limits of molecular weight 60 000, 700 000
and 7 000 000, respectively, for dextran. These values are similar to those obtained
on silica-based phases with pore sizes of 250, 500 and 1000 Å, respectively.

The hydroxyl groups in these polymer gels were utilized to immobilize C_1, C_8
and C_{18} alkyl groups through an ether linkage to form hydrophobic phases. Among
the three PW gels, G4000PW was used to prepare C_1, C_8 and C_{18} stationary phases.
G3000PW and G5000PW were derivatized to C_8 for comparison. The preparation
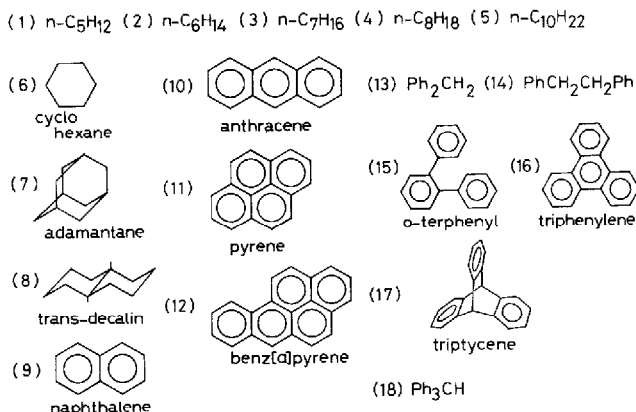method is given below for PW-C_8.

To a suspension of 3 g of G4000PW gel in 50 ml of N,N-dimethylformamide,
1.4 g of sodium hydride was added with stirring under a stream of nitrogen at room
temperature. The mixture was stirred until foaming ceased, then 7.8 g of octyl meth-
anesulphonate were added at 80°C. Stirring was continued for 5 h. The particles were
filtered and washed with water, methanol and chloroform. Other stationary phases
were prepared similarly by using methyl iodide and octadecyl methanesulphonate.
The three stationary phases PW-C_1-I, PW-C_8-I and PW-C_{18}-I were prepared at 60°C.
The particles were packed into stainless-steel columns (100 × 4.6 mm I.D.) by the
conventional slurry technique.

Equipment

An HPLC system was assembled from a TriRotar-V pump and a Uvidec-100-V
UV detector (both from JASCO, Tokyo, Japan), a Model 98.00 refractive index
detector (Knauer, Berlin, F.R.G.) and a 7000A data processor (System Instruments,
Tokyo, Japan).

Chromatography

Mobile phases were prepared from LC-grade methanol and distilled, deionized

(1) n-C$_5$H$_{12}$ (2) n-C$_6$H$_{14}$ (3) n-C$_7$H$_{16}$ (4) n-C$_8$H$_{18}$ (5) n-C$_{10}$H$_{22}$

(6) cyclo hexane

(7) adamantane

(8) trans-decalin

(9) naphthalene

(10) anthracene

(11) pyrene

(12) benz[α]pyrene

(13) Ph$_2$CH$_2$ (14) PhCH$_2$CH$_2$Ph

(15) o-terphenyl

(16) triphenylene

(17) triptycene

(18) Ph$_3$CH

Scheme 1. Solutes used to examine the stationary phase selectivity.

water. The compounds shown in Scheme 1 were used as samples to examine the selectivity of the stationary phases. They included alkanes (1–5), cycloalkanes (6–8), rigid, planar polynuclear aromatic hydrocarbons (PAHs) (9–12, 16), bulky, flexible aromatic compounds (13–15, 18) and rigid, bulky triptycene (17). o-Terphenyl (15) and triphenylene (16) were used to illustrate the effect of rigidity and planarity on retention.

The column temperature was maintained at 30°C by a thermostated water-bath. Chromatographic experiments were carried out in duplicate with a reproducibility of better than ±0.2%. The k' values were calculated by taking the average elution times of very small amounts of water, methanol and glycerine, determined with the refractive index detector, as t_0. GPC measurements were made in tetrahydrofuran using polystyrene standards (Pressure Chemical, Pittsburg, PA, U.S.A.).

RESULTS AND DISCUSSION

Preparation of alkylated polymer gel

As shown in Table I, alkylation of polymer gel resulted in an increase in carbon content up to 6% with PW gels and up to 11.5% with GS gels. More alkyl groups were introduced into the polymer gel by repeating the alkylation. The PW-C$_{18}$ phase showed a smaller extent of alkylation of hydroxyl groups than the PW-C$_8$ phase, presumably because of steric hindrance. In all instances the ^{13}C NMR and IR spectra indicated the alkylation of hydroxyl groups on polymer gels. The increase in the carbon content of polymer particles was much smaller than for silica particles, which showed as much as 20% of carbon in the case of the C$_{18}$ phase. The PW-based phases I and II, prepared at 60 and 80°C, showed a considerable difference in carbon content. However, these stationary phases showed very similar retention characteristics, as can be seen in Tables II–IV. Further study is needed to explain these results. The results with PW-I phase were utilized in all Figures and in the following discussion.

Typically, a 100 × 4.6 mm I.D. column contains ca. 0.89 g of silica-C$_{18}$ and 0.66 g of PW-C$_{18}$ packing material. The amounts of alkyl groups in the silica-C$_8$ and
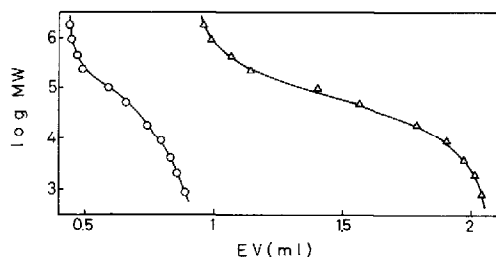
Fig. 1. Elution of polystyrene standards on PW-C_8 phase ($\bigcirc$), 100 × 4.6 mm I.D. column, and on trimethylsilylated LiChrospher Si 500 ($\triangle$), 150 × 4.6 mm I.D. column. Mobile phase: tetrahydrofuran.

-C_{18} columns were estimated to be 3–6 times higher than in the PW-C_8 and -C_{18} columns, respectively.

The pore size of PW-C_8 was found to be similar to that of LiChrospher Si 500, with silica particles having 500 Å pores, as shown by the exclusion limit in GPC in Fig. 1. This agrees with the results for dextran with native PW gel in water, suggesting that the pore structure was preserved during the alkylation.

Hydrophobic properties of polymer-based stationary phases

In order to examine the fundamental properties of polymer-based stationary phases, the retention characteristics of solutes with various structures were studied with a methanol–water mobile phase. They are expected to show a difference in the

TABLE II

CONTRIBUTIONS OF CH_2, OH AND COO GROUPS TO THE k' VALUES OF ALIPHATIC COMPOUNDS

Mobile phase, 80% methanol.

Stationary phase		k' $(\alpha)^{*}$		
		$n\text{-}C_{15}H_{32}$	$n\text{-}C_{14}H_{29}OH$	$n\text{-}C_{13}H_{27}COOCH_3$
Silica-C_1		7.17(1.25)	2.03(0.35)	2.77(0.48)
Silica-C_8		53.3 (1.41)	8.17(0.22)	12.6 (0.33)
Silica-C_{18}		280** (1.54)	20.9 (0.11)	38.2 (0.21)
PW-C_1	I	0.73(1.14)	0.17(0.27)	0.35(0.55)
	II	0.99(1.13)	0.24(0.27)	0.51(0.58)
PW-C_8	I	22.6 (1.35)	2.06(0.12)	4.24(0.25)
	II	25.2 (1.36)	2.25(0.12)	4.67(0.18)
PW-C_{18}	I	44.0 (1.44)	3.20(0.11)	5.95(0.20)
	II	48.0 (1.47)	3.00(0.09)	5.84(0.18)
GS-C_1***		—	—	—
GS-C_8		39.5** (1.38)	4.13(0.14)	6.28(0.22)
GS-C_{18}		64.7**(1.57)	3.64(0.09)	6.27(0.15)

* The α-values (given in parentheses) were calculated by dividing the k' value of each solute by the k' value of n-$C_{14}H_{30}$.
** Extrapolated.
*** Retention too small.

properties of the hydrophobic stationary phase due to differences in the structures of the supports, silica gel and polymer gel.

The hydrophobic properties of a stationary phase can be expressed by the retention increment due to one methylene group, α_{CH_2}, in the solute structure[22,23]. Log α_{CH_2} values correspond to the free energies of transfer of one methylene group from the aqueous mobile phase to the hydrophobic stationary phase. As shown in Table II, the C_{18} phase showed the largest α_{CH_2} values, $k'_{pentadecane}/k'_{tetradecane}$, among the three silica-based stationary phases, and the C_1 phase showed the smallest value. The polymer-based stationary phases showed smaller values than silica-based materials, except GS-C_{18}.

The polar-group selectivity, or the contribution of COO and OH groups in the solutes, indicates how hydrophilic the stationary phases are, *i.e.*, how the polar groups are stabilized on the stationary phase. The C_1 phase showed the largest α_{COO} and α_{OH} values, indicating lower hydrophobicity of the stationary phase. There is a general trend of preferential retention of polar compounds by stationary phases with short alkyl groups. The PW-C_8 and GS-C_8 phases showed slightly less polar group selectivity and lower α_{CH_2} values than the silica-C_8 phase.

The k' values of alkanes may reflect the amount of alkyl groups on the support. The amounts of alkyl groups in the column of polymer-based C_8 and C_{18} are estimated to be about 15–70% of those of silica-based phases. The actual k' values of pentadecane support this estimate. There seems to be little contribution of the polymer backbone to the retention of alkyl compounds. The very small retention of these aliphatic compounds on the polymer-based C_1 phase is notable. This is due to the low hydrophobicity of the methoxy groups in the stationary phase.

Retention characteristics of hydrocarbons of various structure

The structural selectivity of the stationary phases was examined by eluting the hydrocarbons shown under Experimental. From Table III, it is readily seen that the polymer-based stationary phases showed much lower retentions for aliphatic solutes. Polymer-based C_{18} and C_8 showed about 15–75% of the retention shown by silica-C_{18} and -C_8. The difference in k' values between the silica-based and polymer-based phases is much smaller for alicyclic compounds, *trans*-decalin and adamantane. The separation factors $\alpha_{decalin/decane}$ and $\alpha_{adamantane/decane}$, clearly indicate that these cycloalkanes were favoured by polymer-based phase compared with alkanes. This tendency becomes apparent with aromatic solutes. The polymer-based stationary phases actually gave larger k' values than silica-based materials for some aromatic compounds. Note the extraordinary large k' of triptycene on PW-C_1 phase compared with other solutes. This stationary phase, having a low hydrophobicity, showed the largest retention for the rigid, bulky solute triptycene (17). The retention of triptycene (17) and triphenylmethane (18) decreased considerably on the polymer-based C_{18} phases, presumably owing to the bulkiness of the molecules.

The preferential retention of these rigid and/or planar solutes over flexible compounds provides information on the structure of polymer-based stationary phases. This type of selectivity of a polymer-based C_{18} phase was also noted by Wada[21]. The interaction between a carbonyl group and an aromatic group is expected to increase the retention of aromatic solutes[24]. However, the similar tendency between cycloaliphatic compounds and aromatic compounds indicates that the effect is not

TABLE III

EFFECT OF SOLUTE STRUCTURE ON RETENTION

Mobile phase, 80% methanol.

Stationary phase		k' (α)*					
		Decane	trans-Decalin	Adamantane	Pyrene	Triphenylmethane	Triptycene
Silica-C_1		2.02	1.27(0.63)	0.96(0.48)	0.58(0.28)	0.71(0.35)	0.43(0.21)
Silica-C_8		9.97	6.04(0.61)	4.43(0.44)	2.36(0.24)	3.12(0.31)	1.52(0.15)
Silica-C_{18}		31.9	17.6 (0.55)	11.5 (0.36)	7.54(0.24)	7.35(0.23)	3.07(0.10)
G4000PW-C_1	I	0.38**	0.77(2.03)	0.86(2.26)	4.96(13.1)	2.66(7.00)	3.70(9.74)
	II	0.54**	0.96(1.78)	1.02(1.89)	5.59(10.4)	3.19(5.91)	4.19(7.76)
G4000PW-C_8	I	5.17	4.06(0.79)	3.42(0.66)	7.55(1.46)	3.97(0.77)	3.06(0.59)
	II	5.46	4.61(0.84)	3.94(0.72)	9.94(1.82)	5.20(0.95)	4.35(0.80)
G4000PW-C_{18}	I	7.11	4.93(0.69)	3.69(0.52)	7.05(0.99)	3.11(0.44)	2.35(0.33)
	II	7.22	5.26(0.73)	3.98(0.55)	7.93(1.10)	3.55(0.49)	2.81(0.39)
GS-C_1		_***	_***	_***	4.70	0.80	0.86
GS-C_8		5.77	4.51(0.78)	3.71(0.63)	12.0 (2.08)	4.60(0.80)	3.23(0.56)
GS-C_{18}		7.91	6.09(0.77)	5.00(0.63)	9.53(1.20)	2.90(0.37)	2.01(0.25)
G3000PW-C_8		9.96	7.82(0.79)	6.47(0.65)	15.5 (1.56)	9.08(0.91)	4.15(0.42)
G5000PW-C_8		3.38	2.95(0.87)	2.52(0.75)	6.38(1.89)	3.23(0.96)	1.73(0.51)

  * The α-values (given in parentheses) were calculated by dividing the k' value of each solute by the k' value of decane.
  ** Extrapolated.
  *** Retention too small.

only caused by the electronic effect of the participation of π-electrons but also by steric factors. The more rigid the solute, the greater is the preference by polymer-based stationary phase. The polymer-based C_{18} phases showed less preference for the rigid solutes, especially bulky ones, compared with C_1, as shown in Table III.

Previously, we reported the preferential retention of rigid, planar solutes by silica-C_{18} phase compared with silica-C_1 or -C_8 phase[25]. As shown in Scheme 1, triphenylene (16) has a planar structure. o-Terphenyl (15) possesses the same number of carbon atoms and unsaturated bonds as triphenylene, but this molecule cannot assume a planar structure, owing to the steric repulsion between the two phenyl rings[26]. This compound also is flexible, making it more bulky.

As shown in Table IV, triphenylene was preferentially retained by silica-C_{18} compared with non-planar o-terphenyl, $\alpha_{t/o}$ being 1.44, whereas the silica-C_1 and -C_8 phases showed similar $\alpha_{t/o}$ values of about unity between these two solutes. The results were attributed to the contribution of extended C_{18} chains in this solvent system[25]. The tendency was more pronounced in 100% methanol. The planar solute was preferentially retained between the long alkyl chains on the silica surface. The C_1 phase cannot provide such retention.

In contrast to the silica-based materials, all the polymer-based stationary phases showed much greater preference toward the rigid, planar solute. The PW-based phases showed $\alpha_{t/o}$ of 3–3.5 and GS-based phases gave $\alpha_{t/o}$ of 4–7. The fact that large $\alpha_{t/o}$ values were found for all the polymer-based phases, regardless of alkyl chain

TABLE IV

PREFERENCE FOR o-TERPHENYL AND TRIPHENYLENE SHOWN BY STATIONARY PHASES

Mobile phase, 80% methanol.

Stationary phase		k'			
		Triphenylene	o-Terphenyl	$\alpha_{t/o}$*	
Silica-C_1		0.66	0.72	0.92	
Silica-C_8		2.69	2.82	0.95	
Silica-C_{18}		9.72	6.74	1.44	
4000PW-C_1	I	7.49	2.11	3.55	
	II	8.47	2.60	3.26	
4000PW-C_8	I	10.7	3.42	3.13	
	II	14.3	4.32	3.31	
4000PW-C_{18}	I	9.96	2.83	3.52	
	II	11.1	3.16	3.51	
GS-C_1		7.42	1.04	7.13	
GS-C_8		17.8	4.16	4.29	
GS-C_{18}		16.5	2.88	5.72	
3000PW-C_8		23.0	7.46	3.08	
5000PW-C_8		9.03	2.68	3.37	

* The α-values were calculated by dividing the k' value of triphenylene by the k' value of o-terphenyl.

length, indicates that shape selectivity was not only provided by the alkyl chains but also by the polymer gel structure. An extremely large retention of triptycene was found on the C_1 phase, in spite of the small retentions of alkyl compounds.

That the retention of triptycene on PW-C_{18} or -C_8 phase was smaller than on PW-C_1 phase indicates the importance of steric compatibility in determining retention. As indicated by the lower degree of substitution with C_{18} than with C_8 alkyl groups, the pores in the portion of the polymer gel, which supposedly provides the mechanical strength of polymer particles, are relatively small[27]. The contribution of this part of the support structure seems to provide a preference toward rigid, compact solutes. In the sense that steric compatibility between the pore size and the solute size determines the partition coefficients, this resembles the formation of inclusion complexes. Then, it is readily understandable that the relatively hydrophilic C_1 phase showed a very large retention for rigid, bulky solutes.

Apparently, the pores utilized for GPC of much larger molecules are outside these small pores. The difference in surface area between the estimate based on the exclusion limit and that measured by nitrogen adsorption has been explained by the microporosity of polymer gels[27]. Macroreticular resins consist of a highly cross-linked microgel structure and a macroporous structure which is constructed with the microgels. The present observation seems to indicate the participation of the microgel structure in the chromatographic process under RPLC conditions. The existence of a small-pore structure in polymer gels may be one of the reasons for the smaller number of theoretical plates observed with most polymer-based packing materials

compared with silica particles. The portions of polymer gels which showed a preference for rigid, compact solutes could also be the polymer chains on the surface of the microgel structure[28].

The three C_8 stationary phases derived from 3000PW, 4000PW and 5000PW with different exclusion limits for large molecules showed very similar retention characteristics. This suggests a similar microscopic pore structure in the portion providing the shape selectivity and a different pore structure in much larger structural units of these three polymer gels. The alkylation of hydroxyl groups made the contribution of the microgel structure of the polymer gel predominant in RPLC. The three C_8 phases, based on 3000PW, 4000PW and 5000PW, behaved like silica-based stationary phases of different surface area and similar pore size.

Comparison between polymer-based phases with different alkyl chain lengths

The plot of log k' values under one set of conditions against those under another set of conditions indicates a difference in behaviour, caused by alteration in the conditions. As shown in Fig. 2, the silica C_{18} phase showed a preference toward planar solutes compared with the C_1 or C_8 phases. Polynuclear aromatic hydrocarbons (PAHs) were retained much longer on the C_{18} phase than non-planar aromatic compounds, whereas C_1 and C_8 phases showed no such preference, eluting 15 and 16 in a similar k' range. It has been reported that the chain length and surface density of alkyl groups on silicas had large effects on the shape selectivity of RPLC stationary phases[25,26,29].

In contrast to silica-based stationary phases, the PW-C_{18} phase showed a preference toward alkanes over aromatic solutes when compared with the PW-C_1 phase, as shown in Fig. 3a. Pyrene (11) showed a similar retention to decane on PW-C_{18}, while its retention on PW-C_1 phase was more than ten times larger than that of decane. The PW-C_{18} phase showed a structural selectivity very similar to that of the PW-C_8 phase, as shown in Fig. 3b. These facts indicate that the alkyl portions of PW-C_8 or -C_{18} phase contribute to the retention of aliphatic hydrocarbons, whereas
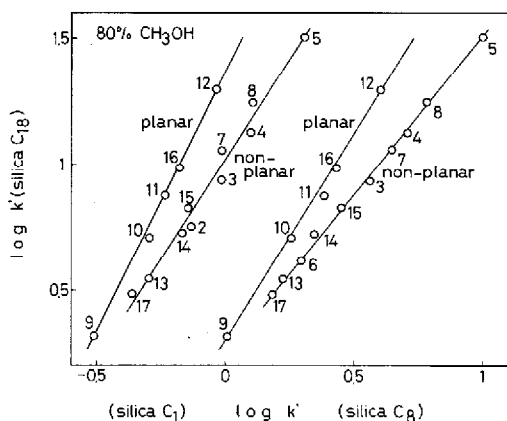

Fig. 2. Plot of log k' values on silica-C_{18} phase against log k' values on silica-C_1 and silica-C_8 phases in 80% methanol. Compound numbers as in Scheme 1.
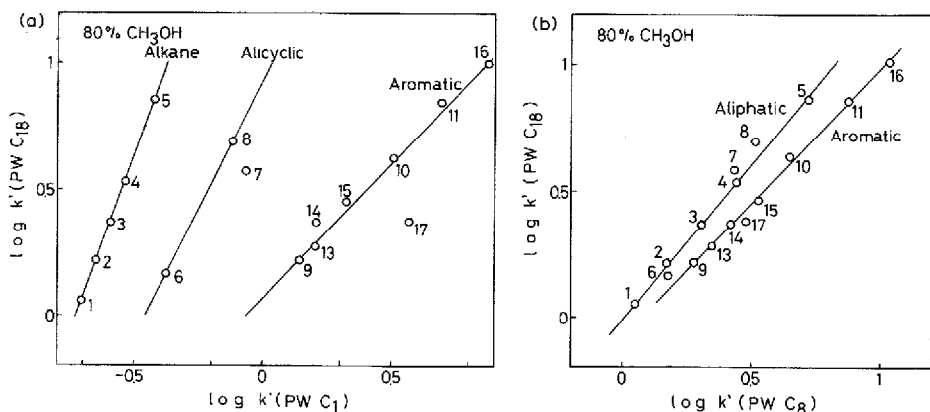
Fig. 3. Plot of log k' values on the PW-C_{18} phase against log k' values on (a) the PW-C_1 phase and (b) the PW-C_8 phase in 80% methanol. Compound numbers as in Scheme 1.

the preference for the rigid, compact compounds shown in Tables III and IV was provided by the polymer support itself. With silica-based material, shape selectivity was provided by alkyl chains in their ordered structure on the solid silica surface, making what are called slots between the alkyl chains[25,29–31]. On the polymer-based phase it seemed to be provided by the polymer network structure in the microgel. The facts that the shape selectivity is not much dependent on the alkyl chain length of the PW phase and that the C_1 phase showed a large retention for bulky compounds lead to this interpretation.

Comparison between polymer-based and silica-based C_8 phases

Fig. 4 shows the plot of log k' values on silica-C_8 against those on PW-C_8. Rigidity in molecular structure in the order alkanes < cycloalkanes < non-planar aromatic compounds < PAHs is the factor which determines the selectivity between
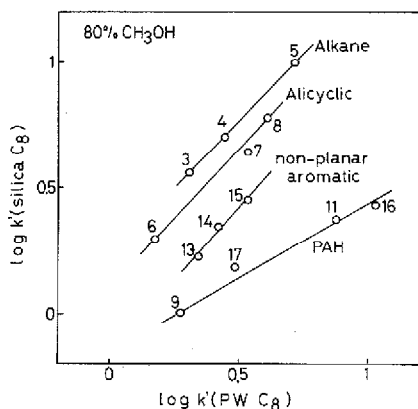

Fig. 4. Plot of log k' values on silica-C_8 phase against log k' values on the PW-C_8 phase in 80% methanol. Compound numbers as in Scheme 1.
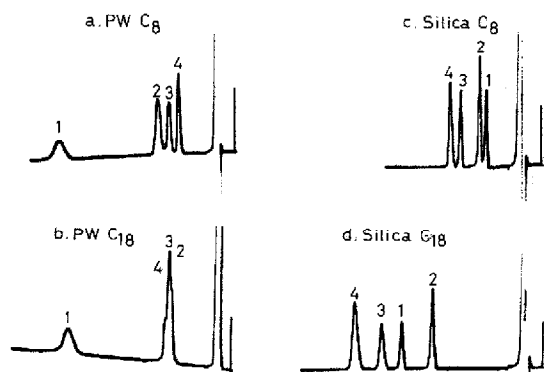
Fig. 5. Chromatograms of (1) triphenylene, (2) triphenylmethane, (3) adamantane and (4) octane on (a) PW-C_8, (b) PW-C_{18}, (c) silica-C_8 and (d) silica-C_{18} in 80% methanol.

these two stationary phases. The PW-C_8 phase showed some similarity to the silica-based pyrenylethyl phase, which has very rigid pyrene rings bonded to the silica surface[31].

Fig. 5 shows the chromatograms of triphenylene, triphenylmethane, adamantane and octane in 80% methanol on PW-C_8 and -C_{18} and on silica -C_8 and -C_{18}. Silica-C_8 and -C_{18} showed a peak reversal between triphenylene and triphenylmethane due to the recognition of planarity by the C_{18} phase. The retention order on PW-C_8 and -C_{18} was completely different from that on the silica-based stationary phases, indicating the preferential retention of rigid molecules.

Effect of the mobile phase on selectivity

Figs. 6 and 7 show the plots of log k' values in 70% methanol against those
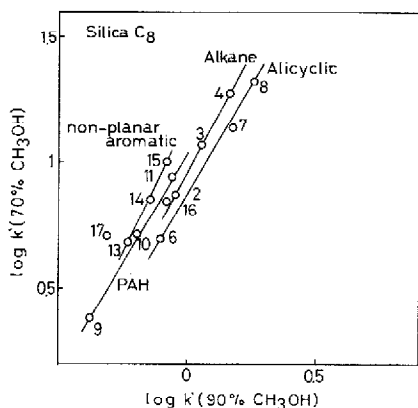

Fig. 6. Plot of log k' values on silica-C_8 phase in 70% methanol against log k' values in 90% methanol. Compound numbers as in Scheme 1.
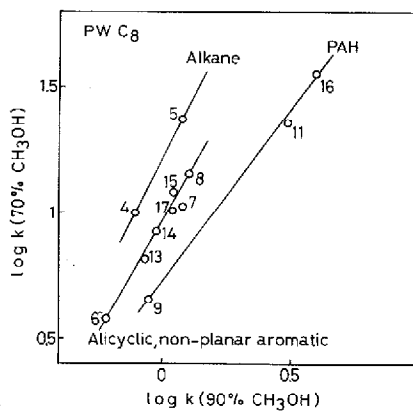
Fig. 7. Plot of log k' values on the PW-C_8 phase in 70% methanol against log k' values in 90% methanol. Compound numbers as in Scheme 1.

in 90% methanol on silica-C_8 and PW-C_8, respectively. In these plots, the difference in selectivity reflect the effect of the change of the mobile phase and also the effect of any change in stationary-phase structure caused by the solvent change. We have previously shown that a change in mobile phase did not cause changes in shape selectivity on silica-C_1, which has relatively small trimethylsilyl groups on the silica surface[25]. The increase in methanol content resulted in a decrease in the relative retention of aromatic solutes compared with saturated solutes on this phase due to an increase in solvation in the mobile phase.

The difference in shape selectivity seen in Figs. 6 and 7 between alkanes and cycloalkanes and between non-planar aromatic compounds and PAHs should be attributed to the change in stationary phase structure brought about by the solvent change. The tendency is common to the two stationary phases. Rigid, compact compounds, PAHs and cycloalkanes, were preferentially retained at higher methanol contents. The extent of the change is much greater with PW-C_8.

The selectivity change caused by the solvent change between aromatic solutes and saturated solutes is just the opposite for the two phases. On the PW-C_8 phase, aromatic compounds were preferentially retained at higher methanol contents. As shown above, an increase in methanol content always caused more solvation in the mobile phase for aromatic solutes, resulting in a smaller relative retention on silica-C_8 and silica-C_1 phases with higher methanol contents. At present, it is not possible to explain the difference in the solvent effects seen on the two C_8 phases in Figs. 6 and 7. As the variation of methanol content caused significant changes in selectivity, the effect of other organic solvents, especially good solvents for the polymer chains, will be particularly interesting to study.

CONCLUSION

C_{18}, C_8 and C_1 alkyl-bonded stationary phases were prepared from hydrophilic polymer gels, and their retention characteristics were examined with hydrocarbons of various structures as samples. The polymer-based phases were less hydrophobic than silica-based materials, and showed a preference for aromatic compounds. The retention of rigid, compact molecules was much greater than that of flexible and/or bulky compounds, owing to the contribution of the polymer network structure in the microgel. The effect of organic solvents in the mobile phase on the shape selectivity was large and different from that on silica-based stationary phase. The difference in selectivity between silica- and polymer-based stationary phases is expected to increase the usefulness of RPLC.

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