

Preparation and Properties of a Sulfobenzylsilica Cation Exchanger for Liquid Chromatography

D. H. Saunders, R. A. Barford, Paul Magidman, L. T. Olszewski, and H. L. Rothbart

Eastern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, Philadelphia, Pa. 19118

The syntheses of sulfobenzylsilicas are described in which the organic moiety is bound to silica surfaces through a carbon-silicon bond. The preparation involved the chlorination of reactive silanol groups, the reaction of this product with benzyl lithium, and finally sulfonation. Special techniques were developed for determining the extent of reaction at each step in the synthesis. One useful method was based on the mixing of a liquid having a refractive index similar to that of silica with the dried solid product so that UV transmission spectra could be obtained. The liquid chromatographic properties of preparations having an ion-exchange capacity of ca. 0.4 mequiv/gram were evaluated. When either pH or ionic strength was varied, organic cations were eluted from these surface-bonded supports in a manner predicted by expressions which had been developed for ion-exchange resins. Although the particles were not optimal in terms of shape and porosity, columns packed with a modified 12-micron fraction of a general purpose commercial silica gave excellent separations with HETP comparable to those observed when commercially-prepared HPLC columns or packings were utilized under similar conditions.

Several approaches to the stable binding of organic layers to supports have been made. Among them are the "brush" phases reported by Halász (1), the pellicular ion-exchangers reported by Horvath (2), the controlled surface porosity exchangers reported by Kirkland (3), and the bonded phases reported by Locke (4).

Organosilica derivatives having a carbon-oxygen-silicon bond are subject to attack by water and some Lewis bases in the same way as are the analogous derivatives of silanes. The "brush" phases produced by Halász are of this type and, although they appear to be useful in anhydrous chromatographic separations, the carbon-oxygen-silicon bond is cleaved by traces of water and alcohols in the mobile phase. Generalizing from organosilicon chemistry (5), it appears that stationary phases bonded through siloxane bonds would be more stable but subject to attack by nucleophilic or electrophilic reagents. Therefore, such derivatives may be modified but choice of reactants is limited.

Organosilica derivatives bonded through carbon-silicon bonds are more readily modified. Even here the choice of reactants to introduce functional groups is circumscribed. On benzylsilica derivatives, Si is subject to nucleophilic attack because it is electropositive relative to carbon, but electrophilic reagents attack the benzene ring resulting in substitution rather than breakage of the Si-CH₂ bond.

EXPERIMENTAL

Materials. Three readily available silicas were used in this work. These included Davison 62 (Davison Division, Grace Corporation) with a particle diameter range of 60-200 μ and a surface area of 340 m²/gram, Cab-O-Sil (Cabot Corporation) with an average particle diameter of 0.012 μ and a surface area of 198 m²/gram and Syloid 74 (Davison Division, W. R. Grace) with a stated particle diameter of 8 μ and a surface area of 320 m²/gram. A fraction (38%) of the Syloid 74 containing particles predominantly 12 μ in diameter was used for the surface modification reactions. The fraction was obtained by a sedimentation procedure from aqueous slurries. ACS grade solvents and reagents were used throughout.

Syntheses. Silica Chloride. At 200 °C, 135 grams of Davison 62 silica were heated in a 1-liter, 3-neck flask for 5 hr under vacuum at 1 mm (Hg) pressure to a dry weight of 129.4 grams. Thionyl chloride (585 grams) was added to the dry silica and the suspension stirred slowly at reflux for 24 hr. After filtration through a medium frit glass Buchner, the product was washed with successive portions (total 600 ml) of dry benzene under an atmosphere of dry nitrogen gas.

The remaining traces of SOCl₂ and benzene were removed by drying first at 100 °C under vacuum and then under the same conditions previously employed with the unreacted silica. We obtained 131.3 grams of dry product.

Benzylsilica. Benzyl lithium was prepared by the dropwise addition of a solution of 0.106 mole of freshly distilled benzyl methyl ether in 32 ml of dry ethyl ether to a stirred suspension of 1.66 g-atoms of cut lithium wire (1% Na) in 64 ml of freshly distilled dry tetrahydrofuran (THF) at -5 °C in a dry argon atmosphere (6). The yield of benzyl lithium as determined by a double titration technique (7) was 58.7%.

A 500-ml, 3-neck flask containing 25.0 grams of Davison 62 silica chloride (23 mequiv chloride) was equipped with a thermometer, a motor-driven glass stirrer, and a reflux condenser with a gas inlet tube for the introduction of dry argon and cooled to -20 °C. Benzyl lithium solution (60 mequiv) was added with stirring to the silica chloride using a glass syringe for the transfer. Three 15-ml portions of 2:1 dry THF-ethyl ether mixtures were used to wash residual benzyl lithium from unreacted lithium particles and added to dilute the benzylation slurry. The chocolate-brown mixture was allowed to warm to room temperature, stirred slowly for 16 hr, and finally heated 6 hr at reflux.

The reaction mixture was then cooled to -30 °C and 25 ml of concentrated hydrochloric acid added to decompose the excess benzyl lithium. The mixture was filtered and washed with successive portions (total 300 ml) of acetone, then methanol (200 ml). The benzylsilica was freed of acid, chloride ion, and most of its buff grey color by suspending it in distilled water. Gravity percolation of the product was continued with portions of distilled water (1500 ml). It was finally washed with ethanol and dried for 5 hr under vacuum (0.1 mm Hg pressure) at 100 °C. The weight of the dry product was 24.7 grams.

Sulfobenzylsilica. An 11.6 gram sample of dry benzylsilica (Davison 62) was suspended in 100 ml of dry chloroform in a 3-neck flask fitted with a motor-driven glass stirrer, thermometer, and dropping funnel. A solution of 8.8 grams of chlorosulfonic acid in 80 ml of dry chloroform was added dropwise in 10 minutes. The light brown-yellow sulfonating mixture was stirred 21 hr at room temperature. After filtering off the sulfonating mixture, the product was washed with small portions of chloroform totaling 300 ml. The washing was continued using a total of 300 ml of acetone. Residu-

(1) I. Halász and I. Sebastian, *Angew. Chem., Int. Ed.*, **8**, 453 (1969).
(2) C. Horvath and S. R. Lipsky, *Anal. Chem.*, **41**, 1227 (1969).
(3) J. J. Kirkland and J. J. De Stefano, *J. Chromatogr. Sci.*, **8**, 309 (1970).
(4) D. C. Locke, J. F. Schmermund, and B. Banner, *Anal. Chem.*, **44**, 90 (1972).
(5) C. Eaborn, "Organosilicon Compounds," Butterworths, London, 1960.

(6) H. Gilman and G. R. Schwebke, *J. Org. Chem.*, **27**, 4259 (1962).
(7) H. Gilman and A. H. Haubein, *J. Amer. Chem. Soc.*, **66**, 1515 (1944).

Table I. Typical Analyses of Surface Modified Silicas

Product	Type of silica	Weight %			Mequiv of bound moiety per gram of product	
		C	S	Cl	Elemental analysis	Titration
Silica chloride	Davison 62			3.30		
Silica chloride	Cab-O-Sil			1.53		0.93
Silica chloride	Syloid 74			4.09		0.43
Benzylsilica	Davison 62	4.11			0.49	1.15
Benzylsilica	Syloid 74	5.33			0.63	
Phenylsilica	Cab-O-Sil	4.0			0.55	
Sulfobenzylsilica	Davison 62	3.63			0.43	
Sulfobenzylsilica	Syloid 74 (12- μ fraction)	4.82	1.30		0.41	0.40 (H ⁺)
			1.22		0.57	
					0.38	0.39 (H ⁺)

Table II. Typical Chromatographic Data

Column and packing	L, cm	Ion exchange capacity, mequiv/gram	Flow rate		HETP, mm ^c		k' ^d	
			ml/min	cm/sec	1	2	1	2
A. Sulfobenzylsilica, (60–200 μ)	91.4	0.40	0.90 ^a	1.1	23.7	27.0	1.5	5.6
B. Sulfobenzylsilica, (12 μ)	50.0	0.39	0.20 ^a	0.21	5.2	3.0	1.8	6.1
C. Low capacity CSP Pellicular, (37 μ)	100.0	<0.01 ^e	0.33 ^a	0.36	...	6.7	0	0.5
D. High capacity Pellicular, (37–50 μ)	91.4	0.06 ^e	1.6 ^a	1.4	26.5	18.2	3.0	21.4
E. Unmodified silica, (12 μ)	25.0	...	0.40 ^b	0.42	0.91	0.50	2.8	16.2
F. Pellicular adsorbent, CSP, (37 μ)	100.0	...	1.0 ^b	1.1	0.41	0.49	2.2	2.5
G. Porous adsorbent, (10 μ)	25.0	...	0.23 ^b	0.27	0.56	1.1	2.8	8.9

For columns A–D, 1 = *p*-nitrobenzoic acid, 2 = *p*-aminobenzoic acid; for columns E–G, 1 = benzene, 2 = nitrobenzene. ^a Eluent: NaNO₃, pH 3.7, ionic strength 0.0168. ^b Eluent: Hexane. ^c HETP = $Lw^2/16V_R(V_R - V_M)$. ^d $k' = (V_R - V_M)/V_M$. ^e Data obtained from manufacturer.

al acid, color, and chloride ion were removed by suspending the product in five successive 100-ml portions of distilled water and allowing water to filter through by gravity percolation.

The product was washed with acetone and chloroform and extracted 96 hr in a Soxhlet apparatus with chloroform. It was finally dried in a 500-ml flask with a controlled bleed of dry nitrogen gas at 100 °C for 5 hr under 50-mm (Hg) pressure. Final weight recovered was 11.5 grams.

Similar steps were used to prepare the sulfobenzyl derivative of Syloid 74 silica.

Phenylsilica. Reactions using Cab-O-Sil required different procedures than those used for Davison 62 because of the former's small particle size and because of its ability to sorb large volumes of liquids, thereby forming undesirable gels. Cab-O-Sil (24.4 grams) was dried and refluxed 24 hr with a mixture of 586 grams of thionyl chloride and 150 ml of dry benzene. Most of the chlorinating agent was then removed by distillation until only a moist residue remained. Dry benzene was added and the mixture was distilled to dryness. At the end, a small amount of dark yellowish liquid distilled (80–126 °C). Two 500-ml portions of benzene were swirled with the silica chloride and decanted. Then a final portion of 500 ml of benzene was distilled from the product which, after drying in vacuum at 200 °C, was composed of dark yellow-tinted particles. The bulk of the reaction product was reacted with phenyllithium (8). The main portion of phenylsilica, after drying at 200 °C in vacuum for 16 hr, weighed 17.6 grams.

Analyses of Surface Bonded Materials. Titrations. The extent of chlorination of the silica surfaces was determined by suspending 1-gram samples in 25 ml of boiled distilled water and titrating with 0.100N sodium hydroxide. Ion exchange capacity of the sulfobenzyl derivatives was determined similarly by titration of the H⁺-form material with base. Benzylsilica treated, but not reacted, with chlorosulfonic acid was washed and extracted as described for sulfobenzylsilica. It was titrated with base, and demonstrated little or no ion-exchange capacity. This indicated negligible sorption of acid and the material was used as a suitable blank.

Elemental Analyses. C, H, Cl, and S were determined both in-house and by a commercial analytical laboratory. In our laboratory, carbon was determined by a Pregl type determination, and percent Cl by oxygen flask combustion followed by mercurimetric titration (9). Determination of sulfur required the addition of about 25% stearic acid in order to increase temperature in the oxygen-flask combustion procedure. This was followed by a volumetric microchemical sulfur determination using tetrahydroxyquinone disodium salt as the indicator (10). The data are reported in Table I.

Spectrophotometry. Dry silica particles were added to UV-transmitting cuvettes with 1-mm path lengths. The cells were held upright and tapped against a firm horizontal surface until settling stopped. Levels were noted and CHCl₃ was added. Trapped air was removed by placing the cells in a vacuum desiccator and carefully controlling the pressure with a bleed arrangement. The closed cells were then tapped once again to resettle the silica to original levels and the now reasonably transparent mixtures were examined in a Beckman DB spectrophotometer. Successfully modified silicas absorbed too strongly for accurate measurements although unmodified silicas were used in the reference beam. Hence, each sample of modified silica was diluted by adding a known quantity of unmodified silica (ca. 20%) to it, and thoroughly mixing them prior to spectrometry. Silicas were removed readily from the cells after directing a stream of N₂ into the cells' openings to dry the particles. The small, 12 μ , particles tended to become tightly packed and had to be blown gently out of the cell with a stream of gas.

Chromatographic Procedures. Studies were carried out with a duPont Model 820 liquid chromatograph. The columns used are listed in Table II: all but columns C, F, and G were constructed in this laboratory. Tubing, Swagelok fittings, and frits at column exits were all made of 316 stainless steel. Packings were contained in tubing (2.1-mm i.d., 6.35-mm o.d.) by glass wool and a Swagelok union fitted with a frit which had a pore size of 10 μ (0.25 μ in

(8) J. Wartmann and H. Deuel, *Helv. Chim. Acta.* **42**, 1166 (1959).

(9) F. W. Cheng, *Microchem. J.* **3**, 537 (1959).

(10) AOAC Official Methods, 11th ed., 42.027 (1970).

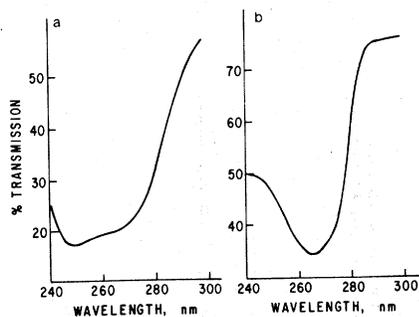


Figure 1. (a) UV spectrum of sulfobenzylsilica by RI matching method. (b) Benzylsilica

columns B and E). The union was then connected to the inlet of the UV detector through the fitting normally used, with a 50-mm length of tubing (0.51-mm i.d., 6.35-mm o.d.). A plug of glass wool, about 0.5 cm long was inserted at the head of the column. Columns A and D were dry packed.

Because of the small particle size, column B was slurry packed with the aid of N_2 at 1000 psig. A 40% aqueous solution of sodium nitrate provided a slurry medium with density sufficient to retard the settling of the silica particles during the packing process. It also reduced the time of adjustment to the buffered sodium nitrate-containing mobile phase. Column E was slurry-packed with $CHCl_3$ which provided a medium of sufficient density and was easily displaced by hexane, the intended mobile phase. Columns C and F were purchased, already packed, from the duPont Company. The packing of C is composed of controlled surface porosity (CSP) particles coated with a strong acid type cation-exchange polymer and has an ion-exchange capacity of <0.01 mequiv/gram. Column D was prepared with Pellionex, a pellicular cation exchanger with a capacity of 0.06 mequiv/gram, purchased from Reeve Angel, Inc. Column G, purchased from the Varian Company, was connected to the injector by means of a low-dead-volume adaptor.

RESULTS AND DISCUSSION

Our studies have shown that thionyl chloride is a satisfactory chlorinating agent for silica if used undiluted. When diluted with dry benzene, a lesser number of silanol groups were replaced by chlorine. The analyses for typical chlorinations using undiluted thionyl chloride are shown in Table II. According to Boehm (11), the extent of the reaction with thionyl chloride gives values for active silanols per unit area of silica surface, comparable to other methods for determining available activities. In our investigations, we consistently have been able to replace a higher percentage of silanol groups with chlorine than we have been able to bind benzyl groups to the same sites. This, in part, may be due to the difficulty of positioning larger organic groups close enough for reaction at all sites. The reaction of some of the chlorinated sites with lithium methylate, present in the benzyl lithium reagent, may result in the attachment of labile methoxy groups, later to be hydrolyzed in purification steps. Analyses (Table I) indicated that slightly more than half of the available chlorine was replaced by benzyl groups. The UV spectrum of the product, shown in Figure 1b, is similar to that of toluene and is supporting evidence for the presence of the benzyl group.

Benzyl methyl ether may be used to prepare benzyl lithium without further purification (6). However, we found that on repeated exposure to air, it oxidized extensively and the organolithium reagent prepared from the contaminated ether would not benzylate silica. Pure benzyl methyl ether may be reclaimed by fractional vacuum distillation. This is accomplished after reduction of peroxidized material, extraction of water soluble contaminants, and removal of the organic solvent.

(11) H. P. Boehm, *Angew. Chem., Int. Ed.*, 5, 533 (1966).

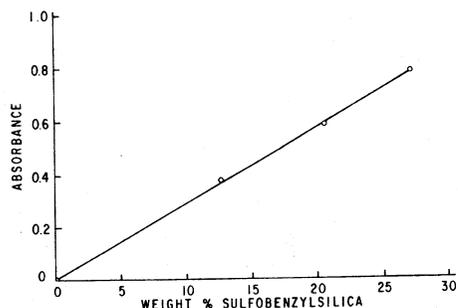


Figure 2. UV absorbance of modified silica-silica mixtures

Carbon analyses indicated a small loss of benzyl groups after sulfonation of the benzylsilica (Table I). If it is assumed that one sulfonic acid group is incorporated on each benzyl, 95% of the remaining bonded aralkyl groups were sulfonated when Davison 62 silica was used, and 66% were sulfonated when Syloid 74 was treated. Electrometric titrations of the products with standard base indicated 0.40 and 0.39 mequiv/gram, respectively, in good agreement with values calculated from sulfur analyses. An attempt to sulfonate using chlorosulfonic acid dissolved in acetone (4) resulted in little or no sulfonation.

The UV spectrum of a sulfobenzylsilica is shown in Figure 1a. Its general features are supporting evidence for the continued presence of benzyl groups modified by sulfonation. The absorbances, at 265 nm, of samples treated in this manner were comparable to those for the original benzylsilicas and correlated with the carbon analyses before and after sulfonation. The extent of the reaction of phenyllithium (Table I) with silica chloride was greater than that of benzyl lithium. However, the sulfonation conditions resulted in cleavage of the carbon-silicon bond with loss of the aryl group.

The spectrophotometric method was a valuable analytical technique throughout this work. It is based upon the increase in light transmission when a powdered solid is wetted with a liquid which has a closely matching refractive index. To evaluate its quantitative characteristics, the relationship between absorbance and percent modified silica in mixtures with unmodified silica was plotted for samples with three different proportions of the same benzylsilica. The results are shown in Figure 2. A straight line fit to the data points by least squares analysis had a sum of squares of residuals of 1.7×10^{-4} . Absorbance was sensitive to the resettled level after the degassing procedure; thus, some care in this regard had to be exercised, especially when the smallest particle materials were utilized. Several preparations of benzyl- and sulfobenzylsilicas were evaluated by this technique. The absorbance, divided by the fraction of modified silica in the dry mixture, was found to be proportional to the corresponding percentages of carbon determined by elemental analysis. However, resettling problems encountered with small particles led to some scatter in the absorbance data and the technique, in these cases, served as a rapid semiquantitative measure of extent of reaction at the end of a number of steps in the synthetic process.

Attempts were made to gain further qualitative data concerning the nature of groups bound to silicas. IR reflectance data were generally uninformative because of extensive light scattering. The hardness of the silica particles prevented the preparation of films at pressures up to 40,000 psig. Dispersion of the particles in halocarbon mulls followed by IR spectrometry resulted in some absorption in the O-H stretching region, due most likely to unreacted hydroxyl moieties on the silica, but no useful

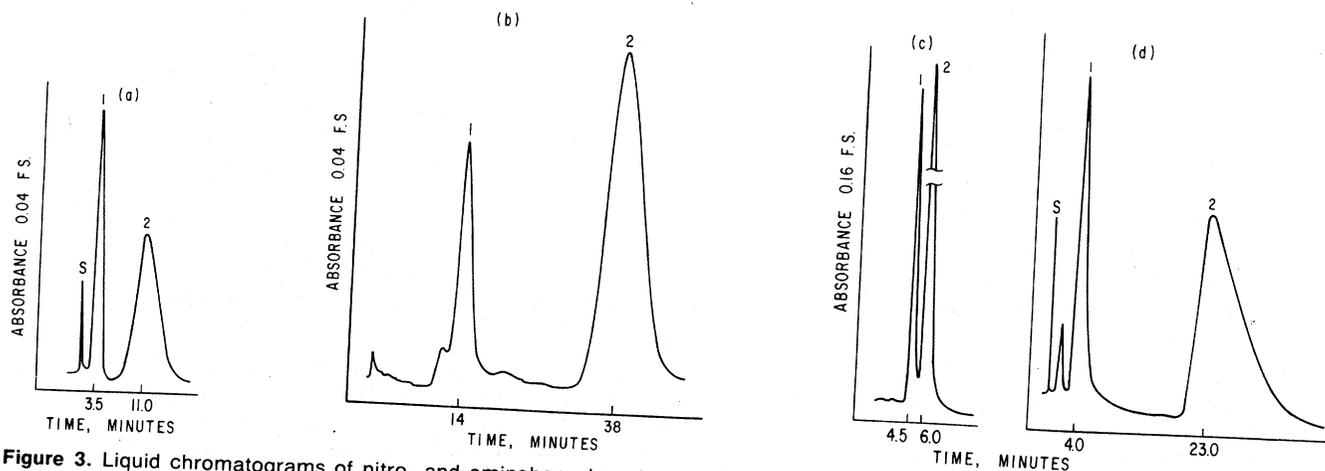


Figure 3. Liquid chromatograms of nitro- and aminobenzoic acid. See Table II for conditions. (a) Col. A, (b) Col. B, (c) Col. C, (d) Col. D

indications of C-H stretching absorbance appeared although elemental analysis supported the presence of benzyl moieties. A 1-mm NaCl cavity cell was utilized in conjunction with CCl_4 as the refractive index-matching solvent. Spectral range was limited to 4000 to 2500 cm^{-1} by the absorbance of the silica. The broad O-H stretching frequency region exhibited strong absorption but the C-H stretching frequency was weak and inconclusive. The latter may be associated with the normally weak bands exhibited by benzyl compounds in the IR.

Attempts to remove bound groups from the silicas by treatment with HF or KOH were generally inconclusive, although an isoctane extract of an HF treated benzylsilica demonstrated a UV absorbance spectrum consistent with typical benzyl-containing species.

Typical liquid chromatograms obtained using the modified porous silicas are shown in Figure 3 along with profiles obtained using two commercial supports. *p*-Nitro- and *p*-aminobenzoic acids were a convenient and important solute-pair for studying the chromatographic properties of the supports (12, 13).

The first organosilica reactions were performed on Davison 62 (60–200 μ) to develop the methods for synthesis. Since no attempt was made to optimize shape, porosity, or particle size ($d_c \ll [2.4 dpL]^{1/2}$) (14), it was not surprising to find HETP (Table II) for this preparation to be much higher than HETP's for the smaller supports. The porous silica values are in the same order of magnitude as the high capacity pellicular support values. Two advantages of higher capacity supports should be considered: large sample sizes may be used in conjunction with detectors of lower sensitivity; and anomalous effects caused by trace cationic contaminants in the chromatographic system are less likely to occur.

It can be shown that for ordinary bulk ion exchangers (15), weakly acidic species such as *p*-aminobenzoic acid are retained in accordance with the expression:

$$V_R^C = V_M + V_S K_{EI}^C [E_1^+]/[E_1^+](1 + K_1[H_M^+]) \quad (1)$$

in which V_R^C = the retention volume of solute C
 V_M = the volume of mobile phase in the column (the dead volume)

- (12) H. H. Weetall and L. S. Hersh, *Biochim. Biophys. Acta.* **185**, 465 (1969).
 (13) C. Venter, J. E. Dixon, P. R. Maroko, and N. O. Kaplan, *Proc. Nat. Acad. Sci.*, **69**, 1141 (1972).
 (14) J. J. De Stefano and H. C. Beachell, *J. Chromatogr. Sci.*, **8**, 434 (1970).
 (15) H. L. Rothbart in "Introduction to Separation Science," B. L. Karger, L. R. Snyder, and C. Horvath, Ed., Wiley Interscience, New York, N.Y., 1973, p 356.

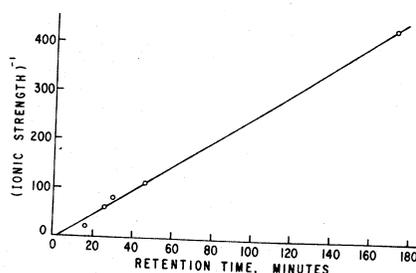


Figure 4. Behavior of *p*-aminobenzoic acid on column A at pH 2.7, aqueous NaNO_3 eluent, 0.01M citric acid buffer

V_S = the volume of stationary phase

K_{EI}^C = the selectivity coefficient = $[E_1^+][C_S^+]/[E_1^+][C_M^+]$

K_1 = dissociation of (protonated) *p*-aminobenzoic acid cation

$[E_1^+], [C_M^+]$ = concentrations of eluent ion and solute ion C, respectively, in the mobile phase

$[E_1^+], [C_S^+]$ = concentrations of eluent ion and solute ion C, respectively, in the stationary phase

Under the conditions of chromatography when the pH of the mobile phase is kept constant, V_R^C should be linearly dependent upon $[E_1^+]$ for bulk ion exchangers. This relationship was demonstrated to be applicable also to the surface bonded exchangers (Figure 4). Similarly when pH was increased, V_R^C approached V_M (Figure 5) since the *p*-aminobenzoic acid cation is essentially absent at high pH. Although the surface-bonded ion exchangers obey the retention equations derived for bulk exchangers, temperature effects were much greater than those generally encountered using resin beads (16, 17). Figure 6 depicts a fourfold decrease in V_R as temperature was varied over 25 $^\circ\text{C}$.

The unmodified, 12- μ silica was evaluated as a packing for adsorption chromatography and as a comparison for the ion exchanger derived from it. Both *p*-aminobenzoic acid and *p*-nitrobenzoic acid emerged from columns packed with Syloid at the void volume when aqueous eluents were utilized. There was no evidence of separation or ion exchange behavior in these cases. Since the small particle size seemed favorable from mass transfer considerations, it was evaluated under conditions in which solutes of similar molar volume had capacity factors of the same order of magnitude as those previously studied. An

- (16) P. Hamilton, *Anal. Chem.*, **35**, 2061 (1963).
 (17) W. Riemann III and H. F. Walton, "Ion Exchange in Analytical Chemistry," Pergamon Press, Elmsford, N.Y., 1970, p 52.

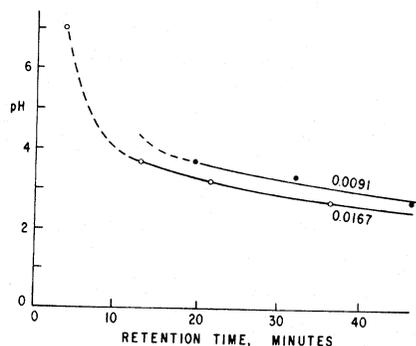


Figure 5. Behavior of *p*-aminobenzoic acid on column A, 0.01M citric acid buffer, NaNO_3 used to adjust ionic strength to values shown

excellent separation with efficiencies comparable to cases utilizing specially designed surfaces (18) was observed (Table II, columns E, F, G).

In conclusion, it may be pointed out that reactions may be carried out with inexpensive porous silicas in order to prepare useful stationary phases for chromatography. The extent of reaction may be rapidly determined by the reported spectrophotometric technique. Although the unmodified silica appears to be the support of greatest efficiency, ion exchangers prepared from small particles have good characteristics for chromatography and obey the retention equations derived for resin beads. HETP values from such an ion exchanger changed only slightly al-

(18) J. J. Kirkland, *J. Chromatogr. Sci.*, 7, 361 (1969).

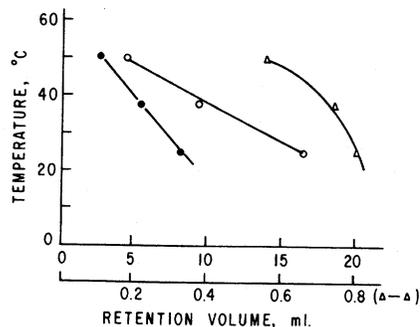


Figure 6. Variation of V_R with temperature. (Δ) acetylsalicylic acid, (\bullet) caffeine, (\circ) phenacetin. Column B, mobile phase— H_2O . Constant flow 0.2 ml/min

though flow rate varied from 0.1–0.5 ml/min. The capacity was much greater, and hence more useful, for the study of natural product mixtures; but the high pressure drops encountered with relatively viscous aqueous eluents obviated greater flow rates and are a disadvantage when these small particles are used.

Current investigations of the utility of these types of supports in the separation of complex mixtures such as lipids in GLC and TLC will be reported soon.

Received for review, September 17, 1973. Accepted January 28, 1974. Presented in part at the 164th National ACS Meeting, New York, Fall 1972. Mention of commercial products does not constitute an endorsement by the United States Department of Agriculture over others of a similar nature not mentioned.